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Résumés des communications affichées

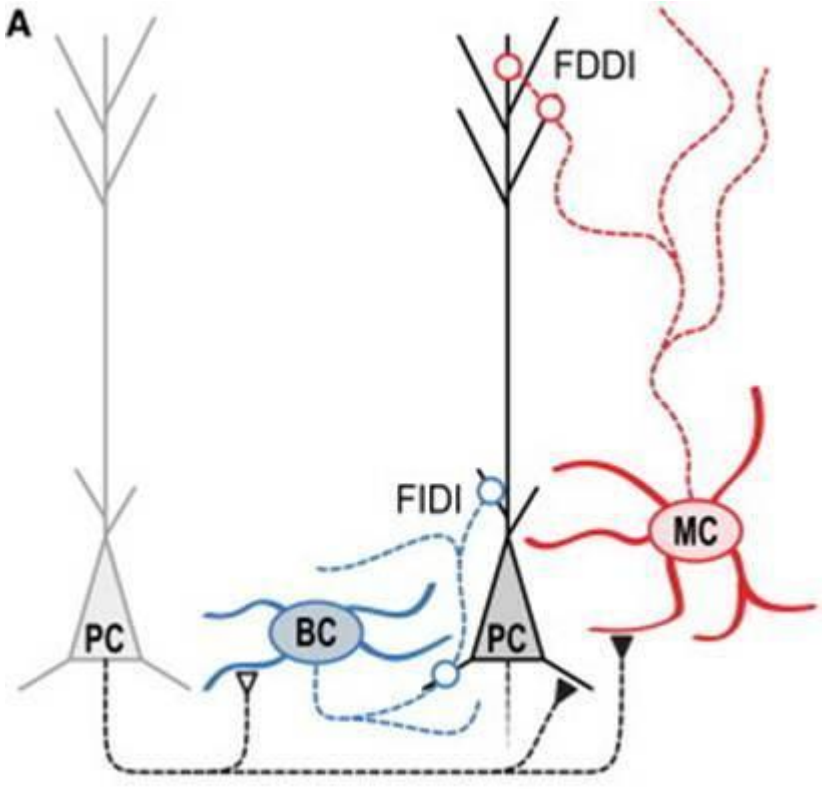
P1.001

Target-specific expression of presynaptic NMDA receptors in neocortical microcircuits

Moreau, A.W. (London)[1], Buchanan, K.A. (London)[1], Blackman, A.V. (London)[1], Elgar, D. (London)[1], Costa, R.P. (London)[1,2], Lalanne, T. (Montreal)[3], Tudor Jones, A.A. (London)[1], Oyrer, J. (London)[1], Sjöström, P.J. (London)[1,3]

[1]University College London, Department of Neuroscience, Physiology, and Pharmacology, London, United Kingdom, [2]School of Informatics, University of Edinburgh, Neuroinformatics Doctoral Training Centre, Institute for Adaptive and Neural Computation, Edinburgh, United Kingdom, [3]The Research Institute of the McGill University Health Centre, Montreal General Hospital, Centre for Research in Neuroscience, Department of Neurology and Neurosurgery, Montreal, Canada

Postsynaptic NMDA receptors are ideal coincidence detectors, as they need both glutamate and depolarisation to open, flux calcium, and elicit plasticity. But presynaptic NMDA receptors (preNMDARs) have also been found in several brain regions. We investigated the expression and role of preNMDARs in mouse visual cortex layer 5 using two-photon laser-scanning microscopy, paired recordings, and neurotransmitter uncaging. The NMDAR blocker AP5 reversibly suppressed AMPA EPSPs at 30Hz between connected pyramidal cells (PC), whereas PC to interneuron (IN) connections were unaffected. Analysis of paired pulse ratio (PPR) and coefficient of variation (CV) indicated a presynaptic action of AP5. We then used transgenic mice specifically expressing GFP to target IN types. At connections from PCs to somatostatin INs (JAX #003718), EPSPs were reversibly suppressed by AP5 with a presynaptic locus, showing that PC inputs to putative Martinotti cells (MCs) have preNMDARs. The effect of AP5 at PC connections to parvalbumin INs (JAX #007677) clustered in two types and this was predicted by postsynaptic IN morphology: type 1 IN (characterized by their ascending axonal arbours) exhibited reversible EPSP suppression due to AP5 (with a presynaptic locus) whereas type 2 IN did not. Morphometry showed that type-2 INs were Basket Cells (BC), which thus have no preNMDARs. We next used internal MK801 to block NMDARs in individual cells. At PC-PC pairs, pre- but not post-synaptic MK801 suppressed EPSPs. Presynaptic MK801 in PC-IN pairs, however, was ineffective. PreNMDARs are thus in the presynaptic PC, but are absent from some PC-IN connections. To locate preNMDARs, 2-photon imaging was combined with MNI-NMDA uncaging. Supralinear calcium signals were found in 55% of boutons, confirming that preNMDAR expression is heterogenous. A tuned network model predicted that preNMDARs impact PC-PC disynaptic inhibition mediated by MCs but not by BCs. Indeed, we experimentally showed that MC disynaptic inhibition was reversibly supressed by AP5. We conclude that preNMDARs exist in L5 PC axons, in a target-specific manner, to regulate information flow in local circuits. PreNMDARs may thus be implicated in pathology due to e.g. schizophrenia or drug use.



[circuit jpg]

P1.002

Effects insulin resistance and obesity on Alzheimer´s disease pathogenesis

Planel, E. (Québec)[1], Gratuze, M. (Québec)[1], El Khoury, N.B. (Québec)[1], Dickler, M. (Québec)[1], Papon, M.-A. (Québec)[1], Poitras, I. (Québec)[1], Marcouiller, F. (Québec)[1], Morin, F. (Québec)[1], Julien, C. (Québec)[1]

[1]*Université Laval, Département de Psychiatrie et Neurosciences, Québec, Canada*

Tau is a microtubule-associated protein that is abundant in the central nervous system and expressed mainly in axons. Tau hyperphosphorylation can induce its aggregation *in vitro*, and is thought to induce neurofibrillary tangles (NFT) formation in Alzheimer´s disease (AD) and other tauopathies. Understanding the causes and consequences of tau pathology is important because its extent shows a strong relationship to dementia in AD, and to memory loss in normal aging and mild cognitive impairment.

The causes of sporadic AD are likely to be multifactorial, with external and biological factors interacting with genetic susceptibilities to accelerate the manifestation of the disease. Diabetes and obesity might be such accelerating factors since both are independently linked to cognitive decline, and since diabetes has been shown to increase the risk of AD. It has been suggested that the effects of diabetes and obesity on AD pathogenesis might be mediated by the concommitant insulin resistance that can be present in both medical conditions. However, recent data data suggest that obesity could accelerate tau pathology in the absence of insulin resistance.

To address the controversy, we used genetic models of diabetes and obesity (db/db mice and ob/ob mice) that present different degrees of insulin resistance and examined tau phosphorylation in their brains. We found that both db/db and ob/ob mice had tau hyperphosphorylation, but also mild hypothermia, which is a powerful promoter of tau hyperphosphorylation. Maintaining the mice normothermic resulted in total rescue of tau phosphorylation in db/db mice, but not in ob/ob mice.

Our results suggest that insulin resistance is inducing tau hyperphosphorylation through hypothermia in both mouse models, but that ob/ob mice have additional upregulation of tau phosphorylation independant of temperature and insulin resistance.
This research will help understanding the link between diabetes, obesity and AD, and the development of future treatments or life style strategies destined to check the advance of the disease.

P1.003

NMDA-R dependent synaptic activity is regulated by neuronal D-serine and glycine via the Asc-1 transporter

Billard, J.-M. (Paris)[1], Rosenberg, D. (Haifa)[2], Artoul, S. (Haifa)[2], Segal, A.C. (Haifa)[2], Kolodney, G. (Haifa)[2], Radzishevsky, I. (Haifa)[2], Dikopoltsev, E. (Haifa)[2], Foltyn, V.N. (Haifa)[2], Inoue, R. (Toyama)[3], Mori, H. (Toyama)[3], Wolosker, H. (Haifa)[2], Dutar, P. (Paris)[1]
[1]Institut de Psychiatrie et de Neurosciences, INSERM UMR 894, Paris, France, [2]Institute of Technology, Department of Biochemistry, Haifa, Israel, [3]University of Toyama, Department of Molecular Neuroscience, Toyama, Japan

Although d-serine is now viewed as a preferential co-agonist of NMDA receptors (NMDAR), the glial vs neuronal contribution to d-serine dynamics are yet to be determined. d-serine was originally identified as a gliotransmitter but recent data show that the synthesizing enzyme serine racemase (SR) is predominantly expressed in glutamatergic neurons, challenging the notion that d-serine is exclusively released from astrocytes. d-serine levels are controlled by two types of antiporters, the Na^+-dependent ASCT1 and ASCT2 and the Na^+-independent Asc-1, the latter known to be restricted to neurons. Data obtained with knockout (KO) mice indicate that Asc-1 is the main d-serine transporter in the brain. In the present study, we investigated the role of Asc-1 in controlling the levels of d-serine in the context of NMDAR-dependent synaptic activity.
Asc-1 antiporter activity is enhanced by d-isoleucine (d-Ile), which releases d-serine but also glycine from Asc-1-transfected cells, primary neuronal cultures and hippocampal slices. d-Ile has no effect on astrocytes. Besides, d-Ile enhances the long-term potentiation (LTP) at hippocampal CA1 in rats and mice by stimulating Asc-1-mediated endogenous d-serine release. d-Ile effects on synaptic plasticity are abolished by enzymatically depleting d-serine or by employing serine racemase knockout (SR-KO) mice, confirming its specificity and supporting the notion that LTP depends mostly on d-serine release. Interestingly, our data also disclose a role of glycine in activating synaptic NMDARs. Although acute enzymatic depletion of d-serine also drastically decreases the isolated NMDAR synaptic potentials, these responses are still enhanced by d-Ile. Furthermore, NMDAR synaptic potentials are preserved in SR-KO mice and are also enhanced by d-Ile, indicating that glycine overlaps with d-serine binding at synaptic NMDARs.
Altogether, our results disclose a novel role of Asc-1 in regulating NMDAR-dependent synaptic activity by mediating concurrent non-vesicular release of d-serine and glycine. Our data also highlight an important role of neuron-derived d-serine and glycine, indicating that astrocytic d-serine is not the sole responsible for activating synaptic NMDARs.

P1.004

Combinatorial patterns of gene expression define new classes of cortical parvalbumin interneurons in the mouse somatosensory cortex

Urban, A. (Paris)[1], Bernard, A. (Seattle)[2], Perrenoud, Q. (Paris)[3], Sorensen, S.A. (Seattle)[2], Gallopin, T. (Paris)[3], Hawrylycz, M. (Seattle)[2], Rossier, J. (Paris)[1], Lein, E. (Seattle)[2]
[1]Centre de Psychiatrie et de Neurosciences, INSERM UMR 894, Paris, France, [2]Allen Institute of Brain Science, Seattle, United States, [3]ESPCI, Laboratoire de Neurobiologie, Paris, France

Interneurons of the neocortex are highly heterogeneous not only in their anatomy and firing properties but much more in their pattern of gene expression. In previous studies the expression of glutamate decarboxylase (GAD), parvalbumin (PV), vasointestinal peptide (VIP), neuropeptide Y (NPY), somatostatin (SOM), was analysed using patch-clamp and single-cell reverse transcription multiplex polymerase chain reaction (RT-mPCR) in acute slices of rodent neocortex. Classification of neurons by unsupervised clustering based on the analysis of multiple electrophysiological and molecular properties disclosed 4 GABAergic interneuron clusters. In this study we added more than 20 genes in our RT-mPCR. These genes were selected after mining the large database of the Allen Institute obtained by fluorescent in situ hybrydization (FISH) of mouse brain. Genes with expression patterns varying primarily by density and to a lesser extent by laminar specificity in the cortex were selected. Markers included Adamts15, Adamts8, MyBPC, pNOC, Reelin, Btbd11, Mme, KIT, Sema4g, Sema3c, Nxph1, Cox6A2, Mchr1, Calbindin, NOS1, Nr2f2, Calretinin, 5HT3a, Btbd14a, AKR1C18 and the traditional markers GAD65 and 67, PV, SOM, NPY, VIP, CCK and VGluT1. More than 200 neurons including 50 pyramids were analysed by RT-mPCR after patch clamp on slice prepared from the somatosensory cortex of juvenile mice. We found a clear clustering of cells based on these markers into molecularly defined groupings, and perfect division of excitatory and inhibitory neurons. VIP bipolar interneurons were identified in a single cluster. Neurogliaform interneurons were identified as a single cluster with high occurrence of 4 markers KIT, Reelin, Sema3c and pNOC. SOM was found associated to two clusters. PV was found associated to few pyramids and distributed at least in two clusters, one expressing SOM and the other expressing the two metallopeptidases Adamts8 and 15. NPY was found in all the clusters including the one containing the pyramids. The presence of metallopeptidase in PV cells could be of importance in view of the reorganization of the perineuronal net with plasticity.

P1.005

Changes in D3 dopamine receptor expression and dopamine transporter function: putative influence on epileptogenesis in absence-epilepsy prone rats?

Cavarec, F. (Grenoble)[1], Krauss, P. (Grenoble)[1], Broisat, A. (Grenoble)[2], Toczek, J. (Grenoble)[2], Ghezzi, C. (Grenoble)[2], Depaulis, A. (Grenoble)[1], Deransart, C. (Grenoble)[1]
[1]Grenoble Institut des Neurosciences, INSERM U. 836 - Equipe 9, Bât. Edmond J. Safra, Grenoble, France, [2]Laboratoire Radiopharmaceutiques Biocliniques Unité Mixte UJF-INSERM U1039 Faculté de Médecine de Grenoble 38700 La Tronche, Grenoble, France

Purpose: Genetic Absence Epilepsy Rats from Strasbourg (GAERS) and Non Epileptic Control rats (NEC) derive from an original Wistar-Hannover rat strain (WH). The age of onset of spike-and-waves discharges in GAERS is about 25 days post-natal (P25). Dopamine has been shown to play a modulatory role in seizure expression in adult GAERS, which display an over-expression of dopaminergic D3-receptors (D3R) as compared to NEC. Because expression and function of D3R and dopamine transporter (DAT) are closely related, we hypothetized that D3R and DAT are involved in absence epileptogenesis during brain maturation. To this aim, we measured their expression and functionality in GAERS pups before the onset of epilepsy (P14 and P21).
Method: D3R expression and functionality was investigated by [^{125}I]-PIPAT autoradiography and quinpirole-induced yawning, respectively, in the three strains at P14 and P21. DAT expression in GAERS and NEC was investigated by [^{123}I]-Ioflupane SPECT imaging and [^{3}H]-GBR12935 autoradiography. DAT activity was assessed by ^{3}H-dopamine reuptake on synaptosomal living fractions of striatum, cortex and hippocampus.
Results: [^{125}I]-PIPAT autoradiography showed an over-expression of D3R in several structures involved in seizure initiation and control, at P14 and P21 in GAERS, as compared to NEC and WH. Similarly, we measured an increase in the number of quinpirole-induced yawns in GAERS at P14 and P21 as compared to age-matched NEC. Neither SPECT imaging nor autoradiographic data showed any differences in DAT expression between the three strains at any ages. However, we observed a consistent increase in ^{3}H-dopamine reuptake in adult GAERS, as compared to NEC and WH in… (quelles structures?).

Conclusion: Our results suggest that an over-expression of functional D3R already exists before the onset of seizures in GAERS and that functional changes in DAT occur in adults, despite a lack of changes in their expression. They further support that changes in basal ganglia function might be a conditional factor for epileptogenesis.

P1.006

Development of the respiratory central pattern generator: an optogenetic approach in the mouse embryo

Ruffault, P.-L. (Gif sur Yvette)[1], Hayes, J. (Gif sur Yvette)[2], Papagiakoumou, E. (Paris)[3], Goridis, C. (Paris)[4], Kiehn, O. (Stockholm)[5], Emiliani, V. (Paris)[3], Brunet, J.-F. (Paris)[4], Fortin, G. (Gif sur Yvette)[2]
[1]Institut de Neurobiologie Alfred Fessard, CNRS UPR 3294-Neurobiologie & Développement, Equipe 12, Gif sur Yvette, France, [2]Institut de Neurobiologie Alfred Fessard, N&D, Gif sur Yvette, France, [3]Université Paris Descartes, Neurophsiology and New Microscopies, Paris, France, [4]ENS, IBENS, Paris, France, [5]Karolinska Institute, Department of Neuroscience, Stockholm, Sweden

Previous investigations from our, and other, labs have started characterizing the distinct modular developmental programs that specify cells forming the pFRG/RTN (also known as e-pF in embryos) and the preBötC. The functional significance of developmental schemes requires a better understanding of intercellular connectivity in order to bridge cellular identity to network assembly and function.
We report on an optogenetic approach *in vitro* to begin dissecting connectivity in between, and within, respiratory oscillators in embryos. We make use of the *vGlut2::ChR2-YFP* mouse line in which the e-pF, but not the preBötC glutamatergic neurons, express Channel-Rhodopsin2 fused to the yellow fluorescent protein. We show that photostimulation of the e-pF evokes typical bursting activity in e-pF neurons, resets its activity, and through the preBötC leads to respiratory-like C4/Phrenic nerve discharges. Light-induced responses are lost in mutants lacking e-pF neurons or in preparations where the bursting pattern of e-pF cells is impaired by riluzole. However, light-induced phrenic responses are preserved when e-pF intercellular synchronisation is disrupted under carbenoxolone. Finally, we show that the above nerve responses can result when light targets a very limited number of e-pF cells (typically a single digit number) using holographic laser stimulation patterns. These data demonstrate that an efficient spread of activity within the e-pF network underlies its all or none mode of collective activation, a property whereby “modular” developmental programs may simply here translate into “modular” functional units.

P1.007

PGE2 modulates synaptic plasticity at hippocampal mossy fiber - CA3 synapse

Maingret, V. (Bordeaux)[1], Mulle, C. (Bordeaux)[1], Amédée, T. (Bordeaux)[1]
[1]Univ. Bordeaux, IINS UMR 5297, Bordeaux, France

There are considerable evidences that neuroinflammation and Alzheimer´s disease (AD) are intimately related. Among different inflammatory molecules highly expressed in the course of the pathology, prostaglandin E_2 (PGE_2) may play a key role.
Interestingly PGE_2 has recently emerged as a potent modulator of synaptic plasticity in the hippocampus and numerous studies have lead to the hypothesis that the early stage of AD was caracterized by hippocampal synaptic dysfunction. PGE_2 acts on four G protein coupled receptors (EP1R-EP4R) subtypes. At the canonical Schaffer collateral - CA1 synapse, PGE_2 modulates basal synaptic transmission and plasticity such as long term potentiation (LTP). However the effect of PGE_2 has not been studied at the mossy fiber (Mf) pathway which connects dentate gyrus granule cells to

CA3 pyramidal neurons. Remarkably Mf-CA3 LTP is presynaptic, independant of NMDA receptors and due to an increase of neurotransmitter release probability.
In this study we investigated the effect of acute application of PGE_2 on presynaptic plasticity at the Mf-CA3 synapse. PGE_2 (10 µM) and either ONO-AE1-249-01 (1 µM), a specific agonist of EP2R, or sulprostone (10 µM), a potent agonist of EP3R, had no effect on basal synaptic transmission. On contrast, PGE_2 (10 µM) or sulprostone (10 µM) strongly impaired Mf-CA3 LTP and those effects were blocked by the co-application of a specific antagonist of EP3R (ONO-AE3-240, 1 µM).
EP3R is negatively coupled to cAMP production through G-proteins, therefore our data suggest that PGE_2 by altering presynaptic level of cAMP and PKA activity decreases the release probability of neurotransmitter. Next experiments will investigate if EP3R is involved in synaptic dysfunction at the Mf-CA3 synapse in a transgenic model of AD-like pathology, the APP/PS1 mouse.

P1.008

Septins control the morphological differentiation of dorsal root ganglia neurons

Boubakar, L. (Villeurbanne)[1], Badouin, H. (Villeurbanne)[1], Falk, J. (Villeurbanne)[1], Castellani, V. (Villeurbanne)[1]
[1]*Universite Claude Bernard Lyon 1, CGphiMC UMR 5534, Villeurbanne, France*

During development, neurons elaborate long processes called axons that enable distant neurons to interconnect. Process initiation appears to be tightly controlled so that the right number of processes is formed at the right place from the start. However, the mechanisms controlling axon initiation remain elusive. We investigated this in chick dorsal root ganglion (DRG) neurons. Monitoring of cells morphology in nascent and more mature ganglia indicates that DRG neurons form a single axon at the ventral pole and another at the dorsal pole. Thus, initiation sites are likely to be induced in specific regions of DRG neurons. Surprisingly, these neurons acquire the same morphology in vitro, suggesting that a significant part of the mechanism is intrinsic. In addition, the centrosome position indicates that DRG neurons are polarised along the dorso-ventral axis *in vivo* before axon formation. Molecular polarity may prefigure morphological differentiation, thus similarly to budding in yeast, some molecules may accumulate at the future site of axon initiation. Among the vertebrate molecules homologous to those involved in budding site selection, we identified the septins. These GTP-ases form filaments that act as diffusion barriers and molecular scaffolds. Septins, homologous with those controlling budding, are expressed in DRG early in morphological differentiation. We found that a pharmacological inhibitor or a dominant-negative construct block axon formation *in vitro* and *in vivo* respectively. In conclusion, septins appear to regulate early morphological differentiation of DRG neurons possibly by controlling axon initiation site selection.

P1.009

Characterization of 3'UTR sequence isoforms of odorant receptor mRNAs in the mouse olfactory system

Dubacq, C. (Paris)[1], Weber, L. (Paris)[1], Trembleau, A. (Paris)[1]
[1]*Université Pierre et Marie Curie - UPMC, Développement et Plasticité des Réseaux Neuronaux UMR CNRS 7102, Paris, France*

Olfactory sensory neurons (OSNs) project from the olfactory epithelium to stereotyped targets within the olfactory bulb such that OSNs that express the same odorant receptor (OR) project to specific glomeruli. Thus, ORs have two main functions in olfactory sensory neurons in mice: besides their conventional role in the detection and transduction of odorant molecules, they also play a critical role

in controlling the sorting and coalescence into glomeruli of axons of the same OR identity (Mombaerts, 2006). Indeed, OR proteins are present in terminal parts of developping and mature OSN axons, precisely where their homotypic fasciculation is initiated. We previously demonstrated that axonal OR mRNAs are translated in this compartment, indicating that axonal OR proteins may have a local origin (Dubacq et al., 2009). Furthermore, we observed that the transport and local translation of OR mRNAs in axons are developmentally regulated, and this regulation may be important for the formation of homotypic bundles of OSN axons just at the time they reach the olfactory bulb. So far, the molecular mechanisms involved in these regulatory processes are unknown, and we are now characterizing OR mRNAs, looking for potential cis-regulating sequences. We first focused on 3' untranslated regions (3'UTRs), since it has been reported in other systems that long 3'UTR isoforms of RanBP1 or IMPA-1 mRNAs are specifically addressed to axons, where they are locally translated (Yudin et al, 2008; Andreassi et al., 2010). Studying a couple of ORs, we demonstrated the expression in the olfactory epithelium of multiple isoforms of these mRNAs, due to alternative polyadenylation. Combining RT-qPCR, *in situ* hybridization and northern blot analyses, we are investigating the relative expression and localization of these isoforms in the OSNs. In future work, understanding the regulation of transport and local translation of OR mRNAs in the OSN axons will allow to further investigate the role of this local translation in the formation and maintenance of the olfactory primary map.

Andreassi C et al. Nat Neurosci. 2010 Mar;13(3):291-301.
Dubacq C et al. J Neurosci. 2009 Aug 19;29(33):10184-90.
Mombaerts P. Annu Rev Cell Dev Biol. 2006;22:713-37. Review.
Yudin D et al. Neuron. 2008 Jul 31;59(2):241-52.

P1.010

5-HT1A and dopamine receptors direct the orientation of plasticity in layer 5 pyramidal neurons of the mouse prefrontal cortex

Meunier, C. (Orsay)[1], Amar, M. (Orsay)[1], Fossier, P. (Orsay)[1]
[1]*CNRS, CNPS Centre de Neuroscience Paris-Sud UMR8195, Orsay, France*

The prefrontal cortex (PFC) is the region of brain that control cognitive functions. Cortical networks are endowed with cellular machinery controlling the equilibrium between excitation (E) and inhibition (I) and synaptic plasticity. In the PFC, the induction of synaptic plasticity is dopamine-dependent and the dopamine release is regulated by serotoninergic receptors as 5-HT_{1A}Rs. Dysfunctional 5-HT_{1A}Rs has been observed in anxiety disorder, schizophrenia and bipolar disorder. Troubles of plasticity are show in neuronal disorders and are linked to dopaminergic alteration. Interaction between dopamine and serotonin is poorly known. Postsynaptic 5-HT_{1A}Rs, dopaminergic receptors D_1R and D_2R are located on pyramidal neurons of layer 5 (L5PyNs) and GABAergic interneurons.
In PFC, we has examined the combining role of 5-HT_{1A}R, D_1R and D_2R in the determination of the E-I balance and the plasticity in L5PyNs in wild-type (wt) and KO 5-HT_{1A}R mice. We performed electrophysiological recordings responses of L5PyNs evoked by electrical stimulation of layers 2-3. An algorithmic decomposition of the global conductance change of the recorded response permits to determine the E-I balance. In L5PyNs, the E-I balance value is 20-80% in wt and significantly modified (23-77%) in KO 5-HT_{1A} mice. In an attempt to modulate the E-I balance by acting on D1R (agonist, antagonist), we found that the E-I is not altered in both strains. In presence of the D2 agonist, the E-I balance was significantly modified to 15-85% in both strain. These results suggest that 5-HT_{1A}Rs and D_2R regulate the E-I balance. To study synaptic plasticity, we apply a HFS protocol, which resulted either in a LTP, a LTD or no plasticity. The analysis of the neuronal population recorded show a concomitant role of the 5-HT_{1A}R and D_1R reduce selectively the induction of LTD (by 26%) and we show a concomitant role 5-HT_{1A}R and D_2R to reduce the induction of the LTP (by 38%). This study reveals new insight in the interaction of dopamine and serotonin receptors to control plasticity and could give the conceptual background for new antipsychotics strategies which associated 5-HT1AR and D1R agonists with D2R antagonists.

P1.011

Collapsin response mediator protein 5 expression induces mitophagy

Amélimoradi, M. (Lyon)[1,2], Brot, S. (Lyon)[1,2], Auger, C. (Lyon)[1,2], Bentata, R. (Lyon)[1,2], Rogemond, V. (Bron)[3], Honnorat, J. (Lyon)[1,2,3]
[1]Centre de Recherche en Neuroscience de Lyon, Equipe ONCOFLAM, Lyon, France, [2]Universite Claude Bernard Lyon 1, Villeurbanne, France, [3]Hospices Civils de Lyon, Hôpital Neurologique, Bron, France

The degradation of damaged mitochondria by mitophagy is an essential process to ensure cell homeostasis. Because neurons, metabolically active cells with a high energy demands, are dependent on the mitochondrial dynamics, mitophagy represents a key mechanism in neuronal function. The collapsin response mediator proteins (CRMPs), highly expressed during brain development, are involved in axon guidance and neurite outgrowth signaling. Among them, CRMP5, a cytosolic protein, regulates neuronal polarity by inhibiting dendrite growth. In this study we demonstrate that CRMP5 can be present, *in vivo,* in brain mitochondria. Electron microscopy shows that CRMP5 over-expression triggers a drastic change in mitochondrial morphology in cells, within which some structures resembling autophagosomes are distinguishable. In addition, CRMP5 over-expression enhances the expression of LC3-II, an autophagosome marker, which is recruited at the mitochondrial level and overlapped CRMP5 labeling. These data added to the recruitment of lysosomal marker at the mitochondrial level strengthens the initiation of mitophagy, resulting in a decrease of mitochondrial content over time. In cultured hippocampal neurons, strong expression of endogenous CRMP5 at an early developmental stage correlates with low mitochondrial content. At later stages, when CRMP5 expression is absent, mitochondrial content enhances, suggesting that CRMP5 modulates the mitochondrion numbers. These results suggest a new function for CRMP5 as an initiator of mitophagy, safeguarding the maintenance of proper dendrite outgrowth.

P1.012

Formation and stability of paranodal junctions in myelinating culture of dorsal root ganglia

FaivreSarrailh, C. (Marseille)[1], Hivert, B. (Marseille)[1], Labasque, M. (Marseille)[1], Tricaud, N. (Montpellier)[2], Goutebroze, L. (Paris)[3]
[1]CNRS CRN2M UMR 7286, Faculté de médecine secteur Nord, CS 80011, Marseille, France, [2]INM INSERM U1051, Montpellier, France, [3]Institut du Fer à Moulin INSERM S839, Paris, France

The node of Ranvier is organized into several subdomains: the nodal gap, flanked by the paranodes and juxtaparanodes. Each domain contains a unique set of cell adhesion molecules (CAMs), scaffolding molecules and ion channels at the basis of the saltatory conduction. With the aim of analyzing the trafficking and clustering of nodal, paranodal, juxtaparanodal CAMs, we set up a system of adenovirus-mediated expression of GFP-CAMs in myelinating co-cultures of DRG sensory neurons and Schwann cells. We observed that Caspr-GFP was strongly recruited at the paranodal junctions of myelinated axons 12 days after the onset of myelin induction with ascorbic acid. Using FRAP, we demonstrated that Caspr-GFP displays a very low turnover when recruited at non-compacted and mature paranodal junctions. Next, we tested the paranodal stability of Caspr in knock-out mice for the scaffolding protein 4.1B. Both paranodal Caspr and juxtaparanodal Caspr2 proteins contain a binding site for 4.1B protein. The 4.1B-deficient mice show major alterations of juxtaparanodes and only slight perturbation of paranodes maybe due to functional redundancy with 4.1G (Cifuentes-Diaz et al., 2012). In accordance, the membrane stability of Caspr-GFP was not altered at paranodes in DRG cultures from 4.1B deficient mice. We also showed that the stability of paranodal Caspr-GFP was not affected by treatment with methyl-ß-cyclodextrine a drug that disrupts lipid rafts. We plan now to analyze the formation of juxtaparanodes and stability of Caspr2-GFP recruited in these Kv1.2-enriched subdomains.

P1.013

Low-excitable lumbar motoneurons are selectively affected in the spinal cord of postnatal transgenic SOD1-G93A mice

Durand, J. (Marseille)[1], Filipchuk, A. (Marseille)[1], Pambo-Pambo, A. (Marseille)[1], Liabeuf, S. (Marseille)[1], Brocard, C. (Marseille)[1], Kulagina, I. (Dniepropetrovsk)[2], Korogod, S. (Dniepropetrovsk)[2], Guéritaud, J.-P. (Marseille)[1]
[1]CNRS UMR7289 Aix Marseille Université, Institut Neuroscience Timone, Marseille, France, [2]International Center for Molecular Physiology, Dniepropetrovsk Section, NASU, Dniepropetrovsk, Ukraine

In superoxide dismutase 1 (SOD1) transgenic mice, standard model of amyotrophic lateral sclerosis, recent studies showed early alterations of spinal motoneurons during the postnatal period supporting the hypothesis that pathological signs begin significantly earlier than overt clinical symptoms. In this work we compared the electrical properties of lumbar motoneurons recorded in the entire isolated brainstem-spinal cord preparation from WT and SOD1 G93A low expressor mice at postnatal days P7-10. We classified the lumbar motoneurons according to their firing pattern (transient, sustained, delayed). The three types of motoneurons also present different excitability during this period of maturation. The motoneurons of transient firing type are the most excitable having the lowest rheobase and the highest input resistance. The motoneurons of the delayed firing type have the highest rheobase, input conductance and membrane potential. The motoneurons of the sustained firing type were the most frequently recorded motoneurons (45 % of the population). In SOD1 G93A mice, the proportion of the sustained firing type was comparable (37%) to that of the WT whereas more delayed firing type were recorded in the SOD1 (42%) compared to 27% in WT. Interestingly, the delayed firing type in SOD1 markedly differed in terms of membrane potential, rheobase, spike threshold and gain (slope of the current intensity-frequency curve as measured in the steady state). Further we demonstrated that the SOD1 G93A motoneurons exhibit dendritic overbranching at the postnatal age P8-P9. The impact of geometry of the dendrites on the discharge patterns was studied on two motoneuron models (WT and SOD1) using NEURON environment. The reconstructed dendritic arborizations were connected to a standardized cylinder shape soma with attached axon hillock, initial segment and a myelinated axon. The simulations using pulse or ramp current injections suggest that the different geometry of dendrites (overbranching in SOD1) could be directly responsible for the higher spike threshold in SOD1 motoneurons. In this work, we show that the SOD1 mutation already affected the delayed firing motoneurons in the second postnatal week, confirming a specific deficit of the largest motoneurons in the SOD1 mice.

P1.014

Respective contribution of d-serine and glycine in synaptic plasticity in the visual cortex of young rat

Fossier, P. (Orsay)[1], Meunier, C. (Orsay)[1], Le Roux, N. (Paris)[2], Levasseur, G. (Marseille)[3], Dallerac, G. (Marseille)[3], Amar, M. (Orsay)[1], Sacchi, S. (Varese)[4], Pollegioni, L. (Varese)[4], Mothet, J.-P. (Marseille)[3]
[1]CNRS, CNPS Centre de Neurosciences Paris-Sud, Orsay, France, [2]UMR-S 839 INSERM UPMC, Institut du Fer à Moulin, Paris, France, [3]Aix-Marseille University, UMR7286 CNRS, Marseille, France, [4]Dipartimento di Biotecnologie e Scienze Molecolari, Politecnico di Milano and Università degli Studi del l'Insubria, Varese, Italy

D-Serine and glycine are believed to exert neuronal modulations as co-agonists of the NMDA receptor (NMDAR) but glycine on its own also activates glycine receptors (GlyR). Dysfunction of NMDARs disturbs cortical network functions particularly in psychiatric disorder such as schizophrenia related to concentration of D-serine and/or glycine. As a consequence, the role of glycine at tuning excitatory

neurotransmission is till controversial and the relative contribution of d-serine and glycine in the dynamic of neuronal network assemblies has to be established.
We performed electrophysiological recording in layer 5 pyramidal neurons (L5PyNs) of composite responses evoked by electrical stimulation of layer 2-3. We report in the rat visual cortex that a theta burst in layer 2-3 induced in the soma of L5PyNs a NMDA-dependent LTP which relies exclusively on d-serine but not on glycine. In the absence of d-serine, the same protocol of stimulation results in a LTD. We further show that glycine is not a co-agonist of synaptic NMDA receptors but rather plays a crucial role in activating GlyR distributed along the apical dendrite of layer 5 pyramidal neurons in visual cortex. Thereby, glycine through the activation of GlyR produces a shunting inhibition that controls neuronal gain and then the LTP to LTD-switch in the soma after dendritic integration. Therefore, the direction of the synaptic plasticity (LTP or LTD) is directly governed by the relative concentrations and specific actions of d-serine and glycine on their own receptors in the visual cortex.

P1.015

Genome-wide analysis of thyroid hormone receptors shared and specific functions in neural cells: perspectives for neurodevelopment

Chatonnet, F. (Lyon)[1], Guyot, R. (Lyon)[1], Benoît, G. (Villeurbanne)[2], Flamant, F. (Lyon)[1]
[1]Institut de Génomique Fonctionnelle de Lyon, CNRS UMR 5242, Université Lyon 1, ENS Lyon, INRA, Equipe Neurodéveloppement, Lyon, France, [2]Centre de Génétique et de Physiologie Moléculaire et Cellulaire, CNRS, Université Lyon 1, Unité Mixte de Recherche 5334, Villeurbanne, France

Thyroid hormone (3,3′,5-triiodo-L-thyronine or T3) plays an important role in neurodeveloppement, as demonstrated by the severe mental retardation observed in patients affected by a low activity level of thyroid hormone during pregnancy and early life.
Thyroid hormone acts on gene expression through two main receptors in mammals, TRα1 and TRβ1, which are transcription factors that share similar properties. However, their respective functions are very different. This functional divergence might be explained in two ways: it can reflect different expression patterns or result from different intrinsic properties of the receptors. We tested this second hypothesis by comparing the repertoires of T3-responsive genes of two neural cell lines, expressing either TRα1 or TRβ1. Using transcriptome analysis, we found that a substantial fraction of the T3 target genes display a marked preference for one of the two receptors.
So when placed alone in identical situations, the two receptors have different repertoires of target genes. Chromatin occupancy analysis, performed at a genome-wide scale, revealed that TRα1 and TRβ1 cistromes were also different. However, receptor-selective regulation of T3 target genes did not result from receptor-selective chromatin occupancy of their promoter regions. We conclude that modification of TRα1 and TRβ1 intrinsic properties contributes in a large part to the divergent evolution of the receptors' function, at least during neurodevelopment.
These results will be exploited to understand the consequences of mutations in either receptor on cerebral development, to devise new receptor and/or tissue-selective drugs modulating thyroid hormone activity while avoiding unwanted side effects, or to predict the neurodevelopmental deleterious effects of perinatal exposure to chemical compounds known to interfere with thyroid hormone receptors.

P1.016

New molecules at the neuromuscular junction : a large scale proteomic approach on synaptic microsamples

Strochlic, L. (Paris)[1], Elbouchtaoui, M. (Paris)[2], Kraut, A. (Grenoble)[3], Couté, Y. (Grenoble)[4], Legres, L. (Paris)[5], Legay, C. (Paris)[6]
[1]Université Paris Descartes, UMR8194 CNRS, Equipe Legay, Paris, France, [2]Institut Universitaire d'hématologie UMR INSERM S_728, 75, Paris, France, [3]Institut de Recherches en Technologies et Sciences pour le vivant U1038 INSERM/CEA/UJF, Grenoble, France, [4]Institut de Biologie de Recherches en Technologies et Sciences pour le vivant U1038 INSERM/CEA/UJF, Grenoble, France,

[5]*Institut Universitaire d'hématologie UMR INSERM S_728, Paris, France,* [6]*Université Paris Descartes UMR8194, Paris, France*

Synapses formed in three main steps including target recognition, differentiation and synaptic maturation of the synapse. At the neuromuscular junction (NMJ), signals from both the neuron and the muscle orchestrate the formation of the synapse in a chronological sequence through a well-defined and dynamic communication between the two cells. Candidate approaches led to the discovery of a number of these signals, but much remains to be done to understand how the pre and the postsynaptic cells recognize each other and form the synapse. Identification of new players during the development of NMJ is limited by the small size of tissue samples and the expression of some proteins such as morphogens at low level and therefore represents a challenging task. In order to discover new molecules concentrated at the NMJ we combined laser capture microdissection (LCM) technique designed to capture cells and isolate accurately specific cells from large regions of tissue and large scale identification of the proteins using mass spectrometry. We used mouse embryonic. Same size of synaptic and extrasynaptic regions of mouse embryonic diaphragms from two developmental stages E16 and E18.5 previously labelled with alpha bungarotoxin to visualise the endplate band, were cut using LCM. After lysis of the microdissected tissue, analysis of the protein composition from both samples using mass spectrometry was performed. First experiments performed with E18.5 diaphragms (N=20 diaphragms) lead to successful validation of the LCM technique to dissect synaptic and extrasynaptic regions. Mass spectrometry analyses identified approximately 1200 proteins in each sample. Among those proteins, around 800 were common to both regions, 200 proteins were only expressed in synaptic region and 200 proteins only expressed in extrasynaptic regions. This comparative study of the NMJ proteomic composition from two developmental stages should allow us to identify new synaptic proteins whose role in the formation of the NMJ remains to be demonstrated.

P1.017

Involvement of the subplate zone in preterm infants with periventricular white matter injury

Verney, C. (Paris)[1], Pogledic, I. (Paris)[2], Fallet-Bianco, C. (Paris)[3], Gressens, P. (Paris)[4,5]
[1]*Inserm U676, PremUP, Université Paris 7, Hôpital Robert Debré, Paris, France,* [2]*INSERM U676, Paris, France,* [3]*Department of Fetal Pathology, Hôpital Cochin, Paris, France,* [4]*Inserm U676, PremUP, Université Paris7, Hôpital Robert Debré, Paris, France,* [5]*Institute for Reproductive and Developmental Biology, Imperial College, London, United Kingdom*

Infants born before 31 postconceptional weeks (pcw) are at high risk for brain damage and cognitive, behavioral, and/or motor deficits. Periventricular white matter injury (PWMI) is the main white matter (WM) damage associated with cerebral palsy. Imaging studies have shown not only cystic PWMI, but also more subtle noncystic PWMI in preterm infants. Pathological studies reports defects of myelination associated with gliosis and microglial activation (Verney et al., 2012). Also imaging studies reported a significant reduction in grey matter introducing a neuronal injury especially in the cerebral cortex and the transient cortical subplate zone.
We selected pathological cases classified into cystic and noncystic PWMI and controls. Most cases were noncystic PWMI corresponding to diffuse white matter lesions which are the predominant lesions currently detected by imaging. Immunocytochemical profiles of glial cells (GFAP+, MonoCarboxylate Transporter-MCT1+, Iba-1+, CD68+, Olig2+) were analyzed at very preterm (24-29 pcw) and preterm period (30-34 pcw) on postmortem human frontal lobe. In noncystic PWMI glial activation extended into the subplate whereas in cystic PWMI glial activation was restricted to the white matter.
Two major age-related and laminar differences were observed in noncystic PWMI: in very preterm cases an increase of activated microglial cells extended into the subplate adjacent to the lesion while in preterm cases astroglial reaction expanded not only into the subplate, but also throughout the cortical plate. There were no differences in Olig 2+ preoligodendrocytes in the subplate in PWMI compared to controls.
In conclusion, PWMI lead to age related laminar characteristic responses for both microglial and astroglial populations. Involvement of gliosis in the deep subplate supports the concept of complex

cellular vulnerability of subplate zone during preterm period and may explain widespread changes of MR signal intensity in imaging of early PWMI.

P1.018

Prenatal alcohol exposure affects the brain vascular system in mice neonates and pFAS/FAS patients

Jégou, S. (Rouen)[1], Laquerrière, A. (Rouen)[1,2], El Ghazi, F. (Rouen)[1], Kwetieu de Lendeu, P. (Rouen)[1], Marret, S. (Rouen)[1,3], Uguen, A. (Brest)[4], Roy, V. (Mont-Saint-Aignan)[5], Marcorelles, P. (Brest)[4], Gonzalez, B. (Rouen)[1]
[1]Université de Rouen, IRIB, UFR Médecine-Pharmacie, ERI 28, NEO VASC, Rouen, France, [2]CHU Rouen, Department of Pathology, Rouen, France, [3]CHU Rouen, Department of Neonatal Pediatrics and Intensive Care, Rouen, France, [4]CHU Brest, Department of Pathology, Brest, France, [5]University of Rouen, IRIB, EA 4306, PsyNCA, Mont-Saint-Aignan, France

In Humans, prenatal alcohol exposure is known to induce brain developmental abnormalities. Depending on the severity of the symptoms, they are referred to Fetal Alcohol Syndrome (FAS) or partial FAS (pFAS). Numerous studies focused on the deleterious effects of alcohol on neural cells. While recent studies suggested that alcohol could affect angiogenesis in adults, the impact of prenatal alcohol exposure on the brain microvasculature remains poorly understood. We investigated the effects of prenatal alcohol exposure on the mice cortical microvasculature and VEGF system, the action of alcohol, glutamate and VEGF on activity, plasticity and survival of microvessels in neonatal brain slices, and compared the cortical microvascular network in control and pFAS/FAS fetuses at different developmental stages. In mice, prenatal alcohol exposure induced a reduction of the cortical vascular density, a loss of the radial orientation of the microvessels and an alteration of the VEGF receptors expression. Calcimetry and videomicroscopy experiments performed on immature cortical slices revealed that alcohol inhibited glutamate-induced calcium mobilization in endothelial cells, affected microvessel morphology and promoted vascular cell death. These effects were prevented by VEGF. Furthermore, we found that vascular suffering precedes the neural death. In humans, we evidenced a stage-dependent alteration in the cortical vascular network of the pFAS/FAS fetuses. While no modification was observed from weeks's gestation 20 (WG20) to WG22, the radial organization of cortical microvessels was clearly altered in pFAS/FAS patients from WG30 to WG38. In conclusion, our data evidence that fetal alcohol exposure affects cortical vascular development both in mice and in pFAS/FAS patients. In addition, alcohol modifies expression of VEGF receptors, endothelial cell activity, vascular plasticity and survival. Altogether, this study suggests that vascular defects would contribute to alcohol-induced brain abnormalities. This work, published in Ann. Neurol. (December 2012, 72; 952-960), was supported by IREB, INSERM, IRIB, University of Rouen.
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P1.019

K+/Cl- co-transporter KCC2 and NMDA receptor subunit expressions in lumbar cord of SOD1 juvenile mice might explain early changes in motoneuron excitability and dendrite overbranching

Liabeuf, S. (Marseille)[1], Brocard, C. (Marseille)[1], Durand, J. (Marseille)[1]
[1]Institut Neurosciences Timone (INT), CNRS UMR 7289, Campus Santé Timone, Marseille, France

Amyotrophic lateral sclerosis (ALS) is a devastating paralytic disorder caused by dysfunction and death of motor neurons with clinical symptoms starting at adulthood. Most of our knowledge on the initial pathophysiological mechanisms comes from transgenic mice with mutations in the gene encoding superoxide dismutase 1 (SOD1). The SOD1 mutation is a proven causal factor in a subset of sporadic and familial ALS cases. Recent evidence obtained in SOD1 mice supports the hypothesis on the slow disease progression with a long preclinical phase during which the disease, although active, is not symptomatic. In SOD1 mice, we found very early abnormalities during development. Pathological signs such as dendritic overbranching were demonstrated in lumbar motoneurons of postnatal SOD1 mice. NMDA and AMPA receptors and also K+/Cl- co-transporter KCC2 have been linked to dendritic branching during development. In the present work, we aimed to determine whether AMPA, NMDA receptor subtypes and/or K+/Cl- co-transporter KCC2 are normally expressed in the lumbar cord of SOD1 mice at postnatal days (P8-P9) precisely when overbranching occurs.We used specific antibodies directed against KCC2, NR2A subunit (NMDA receptors) and GLUR1 subunit (AMPA receptors). Animals were sacrified at 3 days and 8 days after birth.The lumbar spinal cords (L1-L6) were dissected and immediately frozen at -80°C for immunoblotting. For immunohistochemistry, after fixation, the lumbar spinal cords transversal sections (30µm) from 2 mutant SOD1 and WT mice at P3 and P8 were mounted on the same slide and processed simultaneously. Sections were scanned using laser scanning confocal microscope (Olympus FV 500) at 20 x magnification.
Western blot analysis revealed increased expression of NR2A subunit and decreased of KCC2 transporter in lumbar segments of SOD1 mice at postnatal day 8. Low levels of immunofluorescence were detected in SOD1 lumbar motoneuronal pools at postnatal day 3 for KCC2 and synaptophysin. These data are in favor of a decrease of inhibition in the spinal cord of SOD1 pups suggesting a possible role for KCC2 in the early hyperexcitability of motoneurons. The dendrite hyperbranching of motoneurons occurring between P4 and P8 might be an adjustment to compensate for this hyperexcitability.

P1.020

Roles of Wnts in neuromuscular junction formation

Messeant, J. (Paris)[1], Strochlic, L. (Paris)[1], Delers, P. (Paris)[1], Legay, C. (Paris)[1]
[1]*CESeM, UMR8194 CNRS, Université Paris Descartes, Paris, France*

The neuromuscular junction (NMJ) is a synapse between a motor neuron and a skeletal muscle fiber. Signals originating first from the muscle then from the nerve orchestrate the formation of the NMJ through a dynamic communication. Most of these signals are transduced by MuSK, a specific tyrosine kinase receptor. The best-known function of this kinase is the clustering of acetylcholine receptors (AChR). In mice, AChR aneural clusters appear before innervation around E13.5 in a broad central region of the muscle according to a process called prepatterning. With innervation from stage E14, nerve terminal releases the glycoprotein agrin that induces new synaptic AChR clusters by binding MuSK.
Our group has been the first to show that absence of Wnt4 affects the prepatterning and results in a profound defect of innervation in mammals. Moreover, we showed that Wnt4 binds and phosphorylates MuSK. In Zebrafish, Wnt11 has been involved in the same process. We thus ask two questions:
(1) do Wnt11 also acts with Wnt4 in prepatterning?
(2) What are the signalling pathways induced by these factors to control this first step of synapse genesis?
Our results showed that similarly to Wnt4, Wnt11 induces AChR clustering in vitro in a dose-dependant manner. To decipher signalling pathways by which Wnt4 and Wnt11 enhance AChR clustering in muscle cells, we first explored the role of the canonical Wnt signalling pathway. We observed by Western Blot after subcellular fractionating and immunocytochemistry that overexpression of Wnt4 and Wnt11 on muscle cells induces an increase in beta-catenin nuclear level. In parallel, we found that Dikkopf-1, a specific inhibitor of the canonical Wnt pathway, significantly affects on the clustering activity of Wnt4 and Wnt11. These analyses indicate that the canonical

pathway is involved in Wnt4- and Wnt11-mediated AChR clustering in muscle cell. To further define the mechanisms by which these Wnts stimulate AChR clustering, we investigated the plausible functional relationship between these Wnts and Agrin-initiated pathways. Our results support an inhibitory effect of Wnt4 and Wnt11 on Agrin-induced clustering. From these results, molecular mechanisms driving early synaptogenesis are emerging. We will present a model of this process.

P1.021

Distinct plasticity rules at feed-forward *vs.* feedback excitatory inputs onto hippocampal parvalbumin interneurons

LeRoux, N. (Paris)[1,2,3], Cabezas, C. (Paris)[1,2,3], Böhm, U.L. (Paris)[1,2,3], Poncer, J.-C. (Paris)[1,2,3]
[1]INSERM UMR-S 839, Paris, France, [2]Université Pierre et Marie Curie, Paris, France, [3]Institut du Fer à Moulin, Paris, France

Hippocampal parvalbumin-expressing interneurons (PV INs) provide fast and reliable GABAergic signaling to principal cells and orchestrate hippocampal ensemble activities. Precise coordination of principal cell activity by PV INs relies in part on the efficacy of excitatory afferents that recruit them in the hippocampal network. Feed-forward (FF) inputs in particular from Schaffer collaterals influence spike timing precision in CA1 principal cells whereas local, feedback (FB) inputs may contribute to pacemaker activities. Although PV INs have been shown to undergo activity-dependent long term plasticity, how both inputs are modulated during principal cell firing is unknown. We show that FF and FB synapses onto PV INs are endowed with distinct postsynaptic glutamate receptors which set opposing long-term plasticity rules. Inward-rectifying AMPARs expressed at both FF and FB inputs mediate a form of non-Hebbian long term potentiation (LTP) induced upon synaptic activation in the absence of postsynaptic depolarization. In contrast, FF inputs are largely devoid of NMDARs which are more abundant at FB afferents and confer on them an additional form of LTP with Hebbian properties. Both forms of LTP are expressed with no apparent change in presynaptic function but instead can be induced through purely postsynaptic mechanisms. The specific endowment of FF and FB inputs with distinct coincidence detectors allow them to be differentially tuned by distinct frequency regimes of afferent activity. Thus, high frequency activity (>20Hz) specifically potentiates FB, but not FF afferents. We propose that this differential, input-specific tuning of synaptic efficacy may allow PV INs to adapt to changes in hippocampal activity while preserving their precisely-timed, clockwork operation.

P1.022

Axonal Kv1 channels determine intrinsic neuronal excitability in CA3 pyramidal cells

Rama, S. (Marseille)[1,2], Zbili, M. (Marseille)[1,2], Debanne, D. (Marseille)[1,2]
[1]Faculté de Médecine secteur Nord, UMR 1072, Marseille, France, [2]Aix-Marseille Université, Marseille, France

The fast-on, slow-off D-type voltage-gated K^+ current (I_D) has been shown to create a delay in the onset of the first action potential (AP). It thus defines intrinsic excitability in a number of neuronal types including CA3 pyramidal neurons. CA3 neurons exhibit a voltage-dependent subthreshold ramp and a long delay to the firing of the first Action Potential (AP). The ramp-and-delay phenotype can be abolished by voltage-inactivation of bath application of the K^+ channel blocker 4-aminopyridin (4-AP) or dendroxin-I (DTx-I). The reduced rate of depolarization imposed by I_D disrupts the temporal precision of AP generation and limits network synchronization (Cudmore et al., 2010). We have previously shown that activity deprivation of the hippocampal network down-regulates I_D, indicating that it could be a major actor in the regulation of intrinsic neuronal excitability. Although Kv1 channels

have been found in the axon of L5 pyramidal neurons and neocortical PV-BC, the contribution of axonal Kv1 channels in the ramp-and-delay phenotype remains unknown in CA3 neurons.
CA3 pyramidal neurons were recorded in whole cell patch clamp in organotypic slice cultures. The cells were filled with Alexa-488 and the ramp-and-delay was monitored throughout the experiment. In a first series of experiments, the Kv1.1 inhibitor DTx-K was puffed on the apical dendrites or the axon initial segment (AIS). The ramp-and-delay was significantly decreased when DTx-K was applied on the AIS. In these conditions, the excitability of the neuron was found to increase. In contrast, no modification was observed when DTx-K was puffed on equidistant dendrites. In a second step, we proceeded to resection or laser burning of either axon collaterals or dendritic branches. We found that the ramp-and-delay was decreased only when axonal branches were cut. Third, a model of CA3 neuron was developed with the software Neuron and confirmed that the ramp-and-delay was reduced by virtual section of the axon but not the dendrite.
Altogether, our data indicate that Kv1 channels responsible for the ramp-and-delay phenotype are located in the axon of CA3 neurons. This finding stresses out the importance of the axonal compartment in the regulation of the intrinsic excitability of CA3 pyramidal cells.

P1.023

Effects of autophagic modulation on brain microvessels exposed to ethanol

Girault, V. (Rouen)[1], Lesueur, C. (Rouen)[1,2], Gonzalez, B. (Rouen)[1], Bekri, S. (Rouen)[1,2], Brasse-Lagnel, C. (Rouen)[1,2]
[1]Université de Rouen, NeoVasc ERI28, Rouen, France, [2]Department of Biochemistry, Rouen University Hospital, Rouen, France

Hypothesis: Brain developmental disorders are the most devastating consequences of antenatal alcohol exposure. Numerous studies focused on the neuroteratogen effect of ethanol and a recent study suggested that activation of autophagy, a cellular adaptative stress process, is a neuroprotective response to alleviate ethanol toxicity. Recently, our laboratory highlighted that ethanol exposure affects vasculature development in the neonatal brain with a great impact on microvascular cell death. In this study, the involvement of autophagy will be characterized on the cortical microvascular network after ethanol exposure.
Methods: Brain slices of 2-day old NMRI or GFP-LC3 C57/Bl6 mice were used to analyze cortical vascular death and plasticity under autophagic modulation by using 3-methyladenine (3-MA) and rapamycine, as inhibitor and activator, respectively, of the autophagic process. Videomicroscopy experiments with isolectin B4-FITC labeling (an endothelial cells marker) were used to characterize vascular plasticity and co-labelling with 7-AAD was used to assess cell death. Autophagic process was analyzed by confocale microscopy and Western blotting by following the LC3 protein as an autophagic marker.
Results: Results obtained by videomicroscopy highlight a length decrease of cortical microvessels under ethanol exposure which is blocked by 3-MA 30 mM or rapamycine 200 nM. Western blot analysis allow to observe an ethanol trend to activate autophagic activity in the cortex and this effect is blocked by addition of 3-MA. In culture of brain microvessels, autophagy is activated upon ethanol exposure and this activation is abolished in presence of 3-MA.
Conclusion: These preliminary results emphasize that ethanol exposure activates autophagy in the cortex and specifically in cortical microvessels. Moreover, activation or inhibition of autophagic process seems to modulate ethanol effects on cortical microvessels plasticity and death. The involvement of autophagy will be confirmed in cerebral cortex of prenatal ethanol-exposed P2 pups mice and the molecular mechanism involved will be characterized on endothelial cell.

P1.024

Ketamine administration disturbs the developmental profile of NMDA receptor and delays the synaptic targeting of GluN2A-enriched NMDA receptor in the cortex of mice neonates

Lecointre, M. (Rouen)[1], Vézier, C. (Rouen)[1], Brasse-Lagnel, C. (Rouen)[1,2], Bénard, M. (Mont Saint Aignan)[3], Dupré, N. (Rouen)[1], Henry, V. (Rouen)[1], Marret, S. (Rouen)[1,4], Gonzalez, B. (Rouen)[1], Jégou, S. (Rouen)[1], Leroux-Nicollet, I. (Rouen)[1]
[1]Université de Rouen, UFR Médecine-Pharmacie, IRIB,ERI 28 'NEO VASC', Rouen, France, [2]CHU Rouen, Département de Biochimie, Rouen, France, [3]Université de Rouen, IRIB, Plateforme d'Imagerie Cellulaire de Normandie, Mont Saint Aignan, France, [4]CHU Rouen, Département de pédiatrie néonatale et réanimation, Rouen, France

Preterm and term infants can undergo therapeutic surgery under anesthesia, which could interfere with brain development processes. Ketamine is a non-competitive NMDA receptor (NMDA-R) antagonist used in pediatric anesthesia. Disruption of glutamate transmission during the early post-natal period could have deleterious effects, on account of the role of glutamate transmission in cerebral development. Our study compares the effect of *in vivo* ketamine administration (s.c.; 40mg/kg) in mice aged 2, 5 or 10 days (P2, P5 or P10) on the cortical developmental profile of NMDA-R subunits expression and association with their Membrane Associated Guanylate Kinases (PSD95, SAP102). 24h after ketamine administration, western blot analysis showed a decrease in GluN2A and GluN2B subunits at P5 and P10 respectively, whereas PSD95 and SAP102 were increased at P5 only. Moreover, co-immunoprecipitation data showed a decrease in GluN2A/PSD95 association at P2, P5 and P10 while GluN2B/PSD95 was increased at P5, suggesting that the decreased GluN2A targeting to a synaptic position was replaced by an increased GluN2B targeting at this age. Furthermore, GluN2A/SAP102 and GluN2B/SAP102 associations were decreased at P5 and P10, respectively. qRT-PCR analysis from laser-microdissected cortical subregions in control mice showed an heterogeneity in the NMDA-R subunits distribution pattern: only GluN2A mRNA level was lower in deep layers (V-VI) compared to superficial layers (I-IV) at P5. Lower levels of GluN1, GluN2A and GluN2B mRNA were observed in deep layers compared to superficial layers at P10. Ketamine administration led to a decrease in GluN2A mRNA levels only in superficial layers at P5 and an increase in GluN1, GluN2A, GluN2B and PSD95 mRNA levels only in deep layers at P10, thus abolishing the difference between superficial and deep layers regarding cortical NMDA-R subunits distribution. Our data suggest that ketamine administration disturbs the developmental profile of NMDA-R subunits, by delaying the synaptic targeting of GluN2A-enriched NMDA-R and altering the intracortical distribution of NMDA-R subunits in mice neonates.
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P1.025

Functional characterization of CamKII-alpha positive and negative interneurons in the adult olfactory bulb

Gribaudo, S. (Paris)[1], David, L. (Beauport)[2], Chaker, Z. (Paris)[3], Holzenberger, M. (Paris)[3], Triller, A. (Paris)[4], Trembleau, A. (Paris)[1], Saghatelyan, A. (Beauport)[2], Caillé, I. (Paris)[1,5]
[1]Univ. Pierre et Marie Curie Paris 6, UMR CNRS 7102 -Neurobiologie des Processus Adaptatifs, Paris, France, [2]Université Laval, Centre de recherche de l'Institut universitaire en santé mentale de Québec, Beauport, Canada, [3]Centre de Recherche Saint-Antoine, INSERM - UPMC, UMR S 938, Paris, France, [4]IBENS, Institut de Biologie de L'ENS, Paris, France, [5]Univ. Paris Diderot Paris7, Paris, France

Calcium/calmodulin-dependent protein Kinase II alpha (CamKIIa) is a major component of the postsynaptic density and is essential for different forms of synaptic and structural plasticity.
The olfactory bulb (OB) is a region of intense plasticity which is constantly adapting to a changing environment and where new neurons are continuously added throughout life. In this structure, CamKIIa is expressed in GABAergic interneurons residing in the granule cell layer (GCL). BrdU pulse-

chase experiments indicate that only half of granule cells (GCs) express CamKIIa independently of their age. Interestingly, analysis of CamKII-Cre;Rosa::LoxSTOPLox-TomatoRed transgenic mice, in which the expression of the reporter is driven by CamKIIa expression, showed that tomato-red GCs are also positive for CaMKIIa immunoreactivity. This indicates a constant expression of CamKIIa in GCs, confirming that CaMKIIa+ and CamKIIa- populations are distinct and not the result of a shifting expression. CamKIIa+ neoGCs start expressing CamKIIa upon synaptic integration in the OB. Our preliminary results on lentiviral injected neoGCs at different ages indicate that the two classes of GCs do not differ in term of dendritic morphology or distribution in the GCL. However, analyses of expression of immediate early genes cFos and zif268 in basal conditions or after acute odor stimulation show that GCs activation is mostly restricted to CamKIIa+ cells indicating a lower activation threshold of this population. In line with this, preliminary electrophysiological analysis of AAV-CamKIIa::GFP and control AAV-GFP labeled GCs indicate that CamKIIa+ GCs display decreased amplitude and frequency of miniature inhibitory post synaptic current (mIPSC) as compared to CamKIIa- GCs. This suggests that CamKII+ GCs harbor less (or less conductive) GABA receptors and that they receive less GABAergic inputs. We are currently doing additional experiments, including analysis of mEPSCs, comparison of the number of GABAergic synapses impinging on neoGCs using Gephyrin-mCherry knocked-in mice and number of PSD95-GFP positive puncta in retrovirally labelled neoGCs. This should allow us to understand the as yet unexplored differential functions of CamKIIa+ and CamKII- GCs in the OB.

P1.026

Long-lasting enhancement of intrinsic neuronal excitability in PV+ basket cells maintains excitatory-inhibitory balance in hippocampal circuits

Gasselin, C. (Marseille)[1,2], Campanac, E. (Marseille)[1,2], Baude, A. (Marseille)[2,3,4], Rama, S. (Marseille)[1,2], Ankri, N. (Marseille)[1,2], Debanne, D. (Marseille)[1,2]
[1]Fac de Médecine - Secteur Nord - Univ. de la Méditerranée, INSERM U.1072 - UNIS, Plasticité de l'excitabilité neuronale, Marseille, France, [2]Aix-Marseille Université, Marseille, France, [3]CNRS - CRN2M UMR 6231, Marseille, France, [4]INMED INSERM U901, Marseille, France

Excitation-inhibition balance is important for maintaining neuronal activity within physiological bounds and shaping spiking activity in space and time. Classically, activity-dependent plasticity of inhibitory circuits in the hippocampus is thought to be mainly achieved through persistent modifications of excitatory synaptic drive to inhibitory interneurons.
We challenge this view by showing that hippocampal parvalbumin-positive basket cells (PV-BCs), a subset of GABAergic interneurons in the CA1 region, express long-term (>30 min) potentiation of intrinsic neuronal excitability (LTP-IEPV-BC) upon brief repetitive stimulation of the Schaffer collaterals. LTP-IEPV-BC was blocked by MPEP, a selective antagonist of metabotropic glutamate receptor subtype 5 (mGluR5), or by dendrotoxin-I, a blocker of Kv1 channels, indicating that LTP-IEPV-BC is induced by synaptic activation of mGluR5 and mediated by the down-regulation of Kv1 channel activity. The spike threshold was found to be depolarized by 4 mV after induction of LTP-IEPV-BC. LTP-IEPV-BC promotes spiking activity at the gamma frequency (~35 Hz) and facilitates recruitment of PV-BCs to balance synaptic and intrinsic excitation in pyramidal neurons. LTP-IE was specifically expressed in PV-BCs. In fact, a second class of PV-negative interneurons recorded in the pyramidal cell layer and displaying regular spiking did not expressed LTP-IE. In conclusion, activity-dependent modulation of intrinsic neuronal excitability in PV-BCs maintains excitatory-inhibitory balance and therefore constitutes a form of homeostatic plasticity in CA1 microcircuits. *Supported by ANR, INSERM, MENRT & FRM.*

P1.027
Expression and function of BAD-LAMP in GABAergic synapses

Tiveron, M.-C. (Marseille)[1], Beurrier, C. (Marseille)[1], Koehl, M. (Bordeaux)[2], Ceni, C. (Marseille)[1], Gatti, E. (Marseille)[3], Kerkerian, L. (Marseille)[1], Abrous, N. (Bordeaux)[2], Pierre, P. (Marseille)[3], Cremer, H. (Marseille)[1]
[1]IBDM/ CNRS UMR7288/ Aix-Marseille Université, Marseille, France, [2]INSERM U862, Neurocentre Magendie, Bordeaux, France, [3]CIML/ INSERM U1104- CNRS UMR 7280- Aix-Marseille Université, Marseille, France

The Brain And Dendritic cell (BAD) - Lysosome Associated Membrane Protein (LAMP; LAMP5) is a brain specific glycoprotein in mice; in humans it shows also expression in dendritic cells. BAD-LAMP gene is expressed in the granular cells of the Olfactory bulb (OB) and in striatal projection neurons. However, in both cases the BAD-LAMP protein is confined to the target regions of the transcript positive neuron populations, namely the external plexiform layer of the OB, and the Globus pallidus (GP) and Substantia nigra pars reticulata (SNr) in case of the striatum. Using immunohistochemistry, immunoelectron microscopy and isolated synaptic vesicles, we found that BAD-LAMP is associated with GABAergic synaptic vesicles, in agreement with findings for its *C. elegans* orthologue unc-46. We generated a BAD-LAMP null mutant mouse line. BAD-LAMP deficient mice are viable and fertile and do not show any gross anatomical abnormalities. Testing behavior in the open field, the novelty induced activity test or the emergence test revealed no difference between wild type and knock-out mice. However, mutant mice were significantly less anxious in the elevated plus maze. At the electrophysiological level, mini IPSCs recorded in patched pallial cells revealed no differences in the frequency or amplitude between WT and KO mice. However, in the mutant pair pulse stimulation of striatal fibers demonstrated a lower probability of GABA release in the GP. In the light of these electrophysiological differences, and considering that BAD-LAMP is expressed in the striatal projections to the SNr and the GP that represent the direct and indirect pathways of the locomotor regulatory network, we are currently investigating the effects of BAD-LAMP mutation in situations where the equilibrium between both pathways is unbalanced. First, we will analyze locomotor activity after injection of cocaine, a stimulator of the direct pathway. Second, we will induce catalepsy by haloperidol injection, which stimulates the indirect pathway.

P1.028
FMRP and structural plasticity of new neurons in the olfactory bulb

Daroles, L. (Paris)[1], Gribaudo, S. (Paris)[1], Didier, A. (Lyon)[2], Trembleau, A. (Paris)[1], Caillé, I. (Paris)[1]
[1]Université Pierre et Marie Curie, UMR7102 Neurobiologie des Processus Adaptatifs, Paris, France, [2]Centre de Recherche en Neurosciences de Lyon (CRNL). Inserm U1028-CNRS 5292- UCBL1, 69, Lyon, France

The fragile X mental retardation protein (FMRP) is the protein whose absence, due to silencing of the Fmr1 gene leads to the Fragile X syndrome (FXS) in humans, the first cause of hereditary mental retardation. Since a growing body of evidence points to FMRP as a major regulator of neuronal structural plasticity in response to environmental changes, we are testing this hypothesis in the context of adult neurogenesis. We have recently shown that FMR1 mutated neo-neurons in the olfactory bulb are incapable of reducing the length of their dendritic tree upon olfactory deprivation as do wild type neo-neurons (Scotto-Lomassese and al., 2011). We are now extending this study of the defect of neo-neurons structural plasticity in inducible knocked-out mice (Nestin::CreERT2 X FMR1flox/flox). Control or mutated mice have been subjected to an olfactory perceptual learning paradigm, which requires adult neurogenesis (Moreno, 2009). In this task, the mice learn how to discriminate very close odorants (limonene+ and -) by repeated exposure to them. Interestingly, the mutated mice can not learn the task. Morphological analysis of wild type neurons shows that learning induces a lengthening of their dendritic arborisation, similarly to what has been shown in neo-neurons of the hippocampus (Tronel,2010). We are now analyzing the consequences of FMRP absence on these learning induced morphological modifications of neo-neurons. Moreover we are trying to rescue the learning deficits of

the mutant mice by injecting MPEP, an antagonist of mGluR receptors, currently tested in clinical trials (Berry-Kravis, Knox et al. 2011).

P1.029

Electrophysiological and molecular development of rat substantia nigra pars compacta dopaminergic neurons

Dufour, M. (Marseille)[1], Woodhouse, A. (Hobart)[2], Amendola, J. (Marseille)[1], Goaillard, J.-M. (Marseille)[1]
[1]INSERM, INSERM UMR1072, Marseille, France, [2]University of Tasmania, Wicking Dementia Research and Education Centre, Hobart, Australia

Substantia nigra pars compacta (SNc) dopaminergic neurons are involved in movement control, sleep, reward and learning, while the dysfunction and degeneration of these neurons has been implicated in various disorders and diseases including Parkinson's Disease. However, a comprehensive analysis of SNc dopaminergic neuronal development, including electrical properties and the complement of ion channel subunits expressed would be of interest to determine the changes in the expression of ion channels that occur, which may render SNc dopaminergic neurons in the adult more susceptible to degeneration.
To determine the neurochemical profile of dopaminergic SNc neurons we have performed a thorough developmental (P6, P21 and P40) immunohistochemical analysis of the expression of ion channel subunits. We also used patch-clamp recording from P3 to P21 rats to assess developmental changes in the firing properties. We found that the regularity of spontaneous pacemaking increased across postnatal development. When action potentials from P5-8 (n=64) and P16-22 (n=161) SNc dopaminergic neurons were compared the peak amplitude, rise and decay slopes significantly increased, while the spike half-width significantly decreased. Using immunolabelling (n=3 for P6, P21, P40) we identified ion channel subunits that contribute to the somatodendritic delayed rectifier (Kv1.3, Kv2.1, Kv3.2, Kv3.3), A-type (Kv4.3) and calcium-activated SK (SK1, SK2, SK3) potassium currents, I_H (mainly HCN2, HCN4) and the L- (Cav1.2,Cav1.3) and T-type (mainly Cav3.1, Cav3.3) calcium currents in SNc dopaminergic neurons. Across postnatal development the major changes were an increase in the immunolabelling intensity and the dendritic range of HCN, T-type calcium channels, Kv4.3, delayed rectifier and SK channels. This study comprehensively characterises the firing activity and ion channel subunits expressed by SNc dopaminergic neurons across development.

P1.030

Regulatory mechanisms of tau phosphorylation during embryonic and post-embryonic mouse brain development

Poitras, I. (Québec)[1], Bretteville, A. (Québec)[1], Papon, M.-A. (Québec)[1], Marcouiller, F. (Québec)[1], Morin, F. (Québec)[1], Baillargeon, J. (Québec)[1], Planel, E. (Québec)[1]
[1]Université Laval, CRCHU de Québec, Québec, Canada

Background: Tau is a microtubule-associated protein that is abundant in the central nervous system and expressed mainly in axons. The most characterized biological function of tau is to stabilize microtubules and promote their polymerization through the binding of its microtubule-binding domain. This primary function is negatively regulated by phosphorylation as phosphorylated tau species are less efficient to bind microtubules. Tau hyperphosphorylation has been reported to be increased during embryonic and post-embryonic development. However definite mechanisms underlying such hyperphosphorylation have not been fully elucidated.
Objective: To study mechanisms of tau phosphorylation regulation during mouse brain development

Methods: We monitored the levels of both tau phosphorylation and its regulators by biochemistry techniques during mouse brain development from embryonic stages E18 to post-natal stages P60.
Results/conclusions: Tau phosphorylation levels were elevated from E18 to P14 stages, followed by a rapid drop between P14 to P16 stages to reach adult levels. Interestingly, the analysis of the main tau kinases and phosphatases revealed that phosphatases rather than kinases expression/activation pattern likely correlate with the shift in tau phosphorylation during mouse brain development. Moreover, we discovered a new physiological mechanism contributing significantly to tau phosphorylation during mouse brain development.

P1.031

A role for VGLUT1 in synaptic vesicle mobility

Siksou, L. (Paris)[1], Silm, K. (Paris)[2], Biesemann, C. (Göttingen)[3], Nehring, R.B. (Houston)[4], Wojcik, S.M. (Göttingen)[3], Triller, A. (Paris)[5], El Mestikawy, S. (Paris)[2], Marty, S. (Paris)[5], Herzog, E. (Bordeaux)[6]
[1]IBENS, Paris, France, [2]INSERM U952, CNRS UMR 7224, Univ. Pierre et Marie Curie, Paris, France, [3]Max Planck Institute of Experimental Medicine, Dept. of Molecular Neurobiology, Göttingen, Germany, [4]Baylor College of Medicine, Dept. of Molecular and Human Genetics, Houston, United States, [5]IBENS, Institut de Biologie de L'ENS, Paris, France, [6]Univ. Bordeaux, Interdisciplinary Institute for Neuroscience, Bordeaux, France

The integral membrane proteins, Vesicular Glutamate Transporters (VGLUTs), use the proton gradient generated by the v-ATPase to ensure the chloride dependent filling of synaptic vesicles (SVs) with glutamate at excitatory synapses. Previous works have hypothesized a role for VGLUTs in the SVs neogenesis as, in the VGLUT KO, the number of SVs at synapses is decreased and they appear flattened. To test this idea, we reinvestigated the VGLUT1 KO phenotype in detail.
We used electron tomography combined with high-pressure freezing to analyze the VGLUT1 KO synapses' ultrastructure in a close to native-state. This method has revealed that VGLUT1KO SVs are originally spherical and become flattened due to the osmotic pressure exerted by the aldehyde fixative solution. Further test with Bafilomycin A1 showed that blockade of VGLUT1 functioning exerts the same effect on SVs as VGLUT1 removal.
While the number of SVs is reduced in VGLUT1 KO boutons, the SV cycle and the SV protein distribution in subcellular fractionation are not altered. This suggests that SVs are produced normally but do not cluster as much in axon terminals of VGLUT1 KO mice. Time-lapse movies on $Syb2^{EGFP}$ expressing neurons allow us to show that more SVs traffic occurs in VGLUT1 KO axons than in WT. This increased superpool in VGLUT1 KO strongly suggests that VGLUT1 regulates the mobility of SVs.
Altogether, our results demonstrate that beyond glutamate loading, VGLUT1 plays a role in enhancing SVs tonicity and stabilizing SVs at presynaptic terminals.

P1.032

Unique dynamics of germinal zones and precursor diversity during primate corticogenesis

Bétizeau, M. (Bron)[1,2], Cortay, V. (Bron)[1,2], Patti, D. (Bron)[1,2], Bellemin-Ménard, A. (Bron)[1,2], Pfeiffer, M. (Zurich)[3], Pfister, S. (Zurich)[3], Douglas, R. (Zurich)[3], Kennedy, H. (Bron)[1,2], Dehay, C. (Bron)[1,2]
[1]Institut Cellule Souche et Cerveau, INSERM U.846, Bron, France, [2]Université de Lyon, Lyon1, Lyon, France, [3]Institute of Neuroinformatics, University/ETH Zurich, Zurich, Switzerland

During evolution the primate neocortex underwent a large expansion characterised by the formation of new cortical areas and a specific enlargement of the supragranular layers involved in the inter-areal connections and linked to higher cognitive abilities. Previous studies showed that the primate germinal zones display unique features (Smart et al., 2002; Lukaszewicz et al. 2005; Hansen et al. 2010; Fietz et al., 2010). Contrarily to rodents where the ventricular zone (VZ) is the predominant proliferative compartment, the primate specific outer subventricular zone (OSVZ) is the primary site of proliferation during the generation of supragranular layer neurons. We investigate the specific properties of cortical germinal zones and precursors that enable the increased production of supragranular neurons in primates.
Combining in situ marker expression, quantitative confocal microscopy and time-lapse microscopy recordings on organotypic slices after GFP viral transduction, we have performed a detailed analysis of cortical precursors morphology and proliferative behavior at different stages of corticogenesis.
We report unique features of primate cortical precursors regarding cell identity, cell morphology, cell-cycle kinetics and self-renewal abilities. The morphology/markers analysis revealed a higher complexity of the straightforward rule observed in rodents where Pax6 and Tbr2 expressions are associated with radial and non-polarized morphologies respectively. 2-photon real time imaging also uncovered a high diversity of morphologies, correlated with proliferative behaviors and lineage relationships. These recordings enabled to reconstruct progenitor lineage trees both in the VZ and in the OSVZ and gain insight into the dynamics of the two precursor pools.
To conclude, our study highlights primate specific germinal zones dynamics during corticogenesis associated with a morphological and behavioral diversity of cortical precursors.
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P1.033
Evidence for a phagocytic activity of neural stem cells

Ginisty, A. (Poitiers)[1], Gély, A. (Poitiers)[1], Abamraane, L. (Poitiers)[1], Yefimova, M. (Poitiers)[1], Morel, F. (Poitiers)[2], Arnault, P. (Poitiers)[1], Benzakour, O. (Poitiers)[1], Coronas, V. (Poitiers)[1]
[1]Institut de Physiologie et Biologie Cellulaires, Université de Poitiers, CNRS FRE 3511, Poitiers, France, [2]LITEC, Poitiers, France

In adult mammalian brain, neural stem cells contribute to brain tissue homeostasis through production of new neurons and glial cells. The major reservoir of brain neural stem cells is the subventricular zone (SVZ) bordering the lateral ventricle. During the process of cell production, numbers of the newly produced cells die and must be cleared.
We demonstrate using multiple approaches, that SVZ cells in vitro, internalize simplified apoptotic targets consisting in fluorescent carboxylate-modified 1 µm beads. The phenotyping of the cells indicated that in addition to doublecortin expressing neuroblasts, neural stem-like cells expressing GFAP, Sox2, nestin or LeXSSEA and displaying high ALDH activity possess a phagocytic activity. Furthermore, we show that this phagocytic activity also concerns physiological substrates as SVZ cells engulf apoptotic cell fragments. To examine relevance of this mechanism in vivo, we injected fluorescent carboxylate-modified 1 µm beads or fragments of apoptotic cells in brain lateral ventricles and found that cells expressing the Sox2 related stem cell epitope internalize the phagocytic substrates. We then examined possible regulatory mechanisms over this activity and found that the vitamin dependent protein, protein S, regulates SVZ cells phagocytic activity.
Together, our data establish for the first time that, in addition to neuroblasts, SVZ neural stem-like cells contribute to apoptotic cell clearance, suggesting that SVZ cells may contribute to tissue homeostasis not only by producing cells but also by removing cellular debris. The present demonstration of a phagocytic activity of neural stem cells opens new perspectives in stem cell biology and brain physiopathology.

P1.034

Role of astrocytic function in hippocampal adult neurogenesis

Sultan, S. (Lausanne)[1], Moss, J. (Lausanne)[1], Toni, N. (Lausanne)[1]
[1]*University of Lausanne, DNF, Lausanne, Switzerland*

Neural stem cells are present in the dentate gyrus (DG) of the adult hippocampus and produce new functional neurons which participate to the hippocampal function, including learning and memory. This process, named adult neurogenesis, is divided in different steps; adult neural stem cells proliferate to give birth to neuroblasts which migrate in the granule cell layer, differentiate into mature granule neurons and fully integrate into the hippocampal network. Adult neurogenesis is strongly regulated by neuronal activity and neurotransmitters. Since astrocytes are important cellular constituents of the neurogenic niche and release a variety of factors that participate to synaptic transmission, we hypothesized that astrocytic functions could contribute to the integration, maturation and synaptic integration of new generated neurons. To this aim we used a retrovirus targeting progenitor cells to express red fluorescent protein (RFP) in their progenies in transgenic mice in which astrocytic vesicular release was experimentally controlled by the tetracycline transactivation. At different time points after viral injection, we have measured the dendritic maturation of adult-born neurons using a Scholl Analysis and the spine density. Our observations show that in the transgenic mice, newborn neurons are less mature than in wild type mice. Moreover, spine density but also spine maturation, in transgenic astrocytes territories is lower than in non-transgenic astrocytes territories. This observation supports the view that astrocytic vesicular release is necessary for the regulation of adult hippocampal neurogenesis.

P1.035

Unraveling the mechanisms of action of pre and postnatal ethanol exposure on hippocampus synaptic long-term depression

Pierrefiche, O. (Amiens)[1], Silvestre de Ferron, B. (Amiens)[2], Naassila, M. (Amiens)[2]
[1]*Univ. Picardie Jules Verne, UFR de Pharmacie GRAP ERI 24, Groupe de Recherche sur l'Alcool et Pharmacod, Amiens, France,* [2]*INSERM ERI-24 Groupe de Recherche sur l'Alcool et les Pharmacodépendances, Faculté de Pharmacie, Amiens, France*

Chronic ethanol exposure during gestation may induce foetal alcohol syndrome (FAS) that is characterized by life-long learning and memory impairments. Synaptic plasticity within the hippocampus take part in memory processes and some authors reported that long-term potentiation (LTP) of excitatory synaptic transmission is lower in animal model of FAS. However, whether ethanol exposure during gestation also affect long-term depression (LTD), another form of plasticity involved in memory processes, is unknown.
We studied in rats the effects of pre- and postnatal ethanol exposure on synaptic LTD in hippocampus slice from the offspring. Adolescent rats (45-60 days old) exposed to ethanol during gestation and lactation periods were decapitated under deep anaesthesia and slices from hippocampus were prepared in oxygenated aCSF. Population spike recordings in the somatic CA1 area were performed at 31°C. LTD was evoked by a single pulse low frequency stimulation (LFS, 1Hz, 10min) applied to the Shaffer's collaterals with bipolar electrodes. Population spike amplitude was measured for 120 min after LTD induction. Western-blot analysis of NMDA receptor subunits expression was performed in isolated CA1 area.
In control slices, LFS did not induce LTD as expected at this age while in ethanol slices LFS induced LTD. This LFS-induced LTD was NMDA- and more precisely GluR2B subunit dependant. We further demonstrated that LFS-induced LTD in ethanol slices was of synaptic origin and western-blot analysis revealed an over expression of GluR2B subunit specifically located in the synaptic compartment. Interestingly, blocking $GABA_A$ receptors partially blocked this LFS-induced LTD whereas in control slice, it allows now the induction of an NMDA-dependant LTD.
These results demonstrate for the first time that ethanol exposure during brain development disturbs the excitatory/inhibitory balance within the hippocampus with i) an accumulation of GluN2B in synaptic

cleft facilitating LTD induction and ii) a reversed role for $GABA_A$ receptors. These results may account for the cognitive deficits observed in FAS in addition to lower LTP.

P1.036
Two modes of analog-digital facilitation coexist at hippocampal synapses

Zbili, M. (Marseille)[1], Bialowas, A. (Trieste)[2], Rama, S. (Marseille)[1], Marra, V. (Marseille)[1], Fronzaroli-Molinieres, L. (Marseille)[1], Carlier, E. (Marseille)[1], Debanne, D. (Marseille)[1]
[1]Inserm UMR 1072, Marseille, France, [2]International Center for Molecular Physiology, School for Advanced Studies, Trieste, Italy

Synaptic transmission in the brain generally depends on action potentials and neuronal information is transmitted to the postsynaptic neurons as discrete amounts of neurotransmitter in an all-or-non (digital) mode. Recent studies indicate that subthreshold depolarization of the presynaptic membrane potential facilitates spike-evoked transmission in an analog-digital (AD) mode. According to this view, the shape of the presynaptic action potential (pAP) can be highly modulated by analog shift in the somatic membrane potential. So far, synaptic facilitation has been reported through the voltage-dependent broadening of the pAP and the possible modulation of the amplitude of the pAP has not yet been assessed.
We show here that synaptic transmission at excitatory connections formed by CA3 pyramidal neurons can be enhanced by presynaptic voltage in two ways. Transmission is facilitated when the presynaptic AP is either preceded by a long (>5 sec) presynaptic depolarization or by a brief (0.2 sec) presynaptic hyperpolarization. The depolarization-induced AD facilitation (d-ADF; +35%) results from the inactivation of presynaptic Kv1 channels that control spike width. It is blocked by dendrotoxin-K (a specific inhibitor of Kv1.1 channels). Calcium imaging in the axon shows that AP-evoked calcium signals are enhanced if the pAP is triggered after a sustained depolarization. The hyperpolarization-induced AD facilitation (h-ADF, +20%) is due to the recovery from inactivation of Nav channels controlling presynaptic spike amplitude. The spike-evoked calcium signals measured in the axon are also enhanced when the presynaptic spike is preceded by a transient hyperpolarization. Both d-ADF and h-ADF can be reproduced in a computational model of CA3 neuron using the software *Neuron*. These two coexisting modes of AD facilitation can be observed in the same connection under physiological conditions.
We conclude that the informational content of the presynaptic spike is highly modulated by the context in which it is triggered. Our study also suggests that excitatory transmission is optimized in the hippocampus if the presynaptic spike is triggered by a specific sequence of long depolarization and brief hyperpolarization.
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P1.037
A model of cytomegalovirus infection in the developing rat brain

Cloarec, R. (Marseille)[1], Massacrier, A. (Marseille)[1], Buhler, E. (Marseille)[1,2], Watrin, F. (Marseille)[1], Stamminger, T. (Erlangen)[3], Streblow, D. (Portland)[4], Bruneau, N. (Marseille)[1], Szepetowski, P. (Marseille)[1]
[1]Mediterranean Institute of Neurobiology (INMED), INSERM UMR_S901, Aix-Marseille University, Marseille, France, [2]INMED Post-Genomic Platform (PPGI), Marseille, France, [3]Institute for Clinical and Molecular Virology, Erlangen University, Erlangen, Germany, [4]Vaccine & Gene Therapy Institute, Oregon University, Portland, United States

Cytomegalovirus (CMV) is a member of the herpes virus family. Congenital CMV infections have an incidence of about 1% of all live births and represent the most frequent cause of neurodevelopmental disorders in human. 5-10% of infected neonates show severe neurological defects such as microcephaly whereas another 10% will subsequently develop brain disorders such as intellectual deficiency, sensorineural hearing loss, visual defects, or epileptic seizures. Hence congenital CMV infections represent a major health care problem. Various animal models of congenital or neonatal CMV infections of the CNS have been reported but overall the pathophysiology remains poorly known. In order to have a better knowledge on the mechanisms, the time course and the consequences of CMV infection in the developing brain, we have designed a model of CMV infection by *in utero* intraventricular injections at embryonic day 15 (E15) of recombinant rat CMV expressing GFP. Ongoing analyzes are being performed at different developmental time-points (E15.5, E17, E20, P1). The determination of the various anatomical sites of rat CMV infection within the brain, such as the periventricular areas or the choroid plexus, indicated strong overlaps with the usual brain abnormalities seen in human. Identification of the different infected cell types is currently being done by immunohistochemistry experiments. Overall our study should provide new insights on the possible mechanisms underlying the pathophysiology and the outcome of CMV infections in the developing brain.
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P1.038

Role of sumoylation in the control of spine morphology

Khayachi, A. (Valbonne)[1], Loriol, C. (Valbonne)[2], Cassé, F. (Valbonne)[2], Poupon, G. (Valbonne)[2], Gwizdek, C. (Valbonne)[2], Martin, S. (Valbonne)[2]
[1]Ipmc - Cnrs Umr 7275, Valbonne, France, [2]Ipmc - Cnrs Umr 7275, 06, Valbonne, France

Post-translational modifications play essential roles in many aspects of cellular functions and in the maintenance of cell integrity. Sumoylation is a post-translational modification that consists in the covalent but reversible conjugation of the Small Ubiquitin like MOdifier protein SUMO to specific lysine residues of target proteins. Sumoylation is a well-characterized regulator of nuclear functions but also emerges as a key factor for numerous extranuclear processes. Interestingly, it was demonstrated that sumoylation plays key roles in neuronal development and synaptic plasticity. We recently identified a protein shown to be important for the regulation of neuronal morphology that is a target for sumoylation *in vivo.* To unravel the role of this protein sumoylation on spine architecture we used site-directed mutagenesis to engineer a non-sumoylatable target protein. Using viral transduction and confocal microscopy, we assessed the neuronal morphology and synaptic architecture from primary cultured neurons expressing the WT or non-sumoylatable form of the protein. Our data indicate that overexpression of the non-sumoylatable protein in neurons is able to disrupt neuronal morphology indicating that it acts as a dominant negative on the endogenously expressed target protein. Here we demonstrate that sumoylation is important for the control of spine morphology.

P1.039

Baclofen and other $GABA_B$ receptor agents are allosteric modulators of the CXCL12 chemokine receptor CXCR4

Guyon, A. (Valbonne)[1], Kussrow, A. (Nashville)[2], Olmsted, I.R. (Nashville)[2], Bornhop, D.J. (Nashville)[2], Nahin, J.-L. (Valbonne)[1]

[1]*Institut de Pharmacologie Moléculaire et Cellulaire, CNRS UMR 7275 - UNSA, Valbonne, France,*
[2]*Vanderbilt University, Department of Chemistry, Nashville, United States*

CXCR4, a receptor for the chemokine CXCL12 (stromal-cell derived factor (SDF)-1α), is a G protein-coupled receptor (GPCR), expressed in the immune and central nervous systems and integrally involved in various neurological disorders. The γ-Aminobutyric acid type B ($GABA_B$) receptor is also a GPCR that mediates metabotropic action of the inhibitory neurotransmitter γ-aminobutyric acid (GABA) and is located on neurons and immune cells as well. Using diverse approaches, we report novel interaction between $GABA_B$ receptor agents and CXCR4 and demonstrate allosteric binding of these agents to CXCR4. First, both $GABA_B$ antagonists and agonists block CXCL12-elicited chemotaxis in human breast cancer cells. Second, $GABA_B$ antagonist blocks the potentiation by CXCL12 of high threshold Ca^{+2} channels in neurons. Third, electrophysiology in Xenopus oocytes and HEK293 cells in which we co-expressed CXCR4 and the GIRK channel showed that $GABA_B$ antagonist and agonist modified CXCL12-evoked activation of GIRK channels. To investigate whether $GABA_B$ ligands bind to CXCR4, we expressed this receptor in heterologous systems lacking $GABA_B$ receptors and performed competition binding experiments. Our FRET experiments suggest that $GABA_B$ ligands do not bind CXCR4 at the CXCL12 binding pocket suggesting allosteric modulation, in accordance with our electrophysiology experiments. Finally, using backscattering interferometry (BSI) and lipoparticles containing only the CXCR4 receptor, we quantified the binding affinity for the $GABA_B$ ligands, confirming a direct interaction with the CXCR4 receptor. The effect of GABAergic agents on CXCR4 suggests new therapeutic potentials for neurological and immune diseases.

P1.040
Cortical pyramidal cells are a major source of vasodilatory prostanoids

Lacroix, A. (Paris)[1], Plaisier, F. (Paris)[1], Karagiannis, A. (Paris)[1], Toussay, X. (Montréal)[1,2], Chausson, P. (Paris)[1], Jarlier, F. (Paris)[1], Burgess, S.A. (New York)[3], Mc Caslin, A.F.H. (New York)[3], Hillman, E.M.C. (New York)[3], Hamel, E. (Montréal)[2], Cauli, B. (Paris)[1]
[1]*CNRS UMR 7102, Neurobiology of Adaptative Processes, Paris VI University, Paris, France,*
[2]*Cerebrovascular Research, Montreal Neurological Institute, McGill University, Montréal, Canada,*
[3]*Biomedical Engineering, Columbia University, New York, United States*

The tight coupling between neuronal activity and cerebral blood flow is a complex physiological process. In the cerebral cortex this neurovascular coupling is achieved by a delicate interplay between neurons, astrocytes and arterioles. Among the various messengers involved in the hyperemic response to whiskers stimulation, the vasodilatory prostanoids, prostaglandin E2 (PGE_2) and/or prostacyclin (PGI_2), derived from the activity of cyclo-oxygenase 2 (COX-2), account for half of the response. However their cellular origin and nature are still under debate.
Using immunocytochemistry we found that COX-2 is constitutively expressed by 1/3 of pyramidal cells but not by astrocytes. To evaluate the putative nature of the vasodilatory prostanoids we determined, using patch-clamp recordings and single-cell RT-PCR, the expression profile of the PGE_2 and PGI_2 synthesizing enzymes in identified perivascular astrocytes and neurons of the rat barrel cortex. Sulforhodamine 101-labelled perivascular astrocytes were characterized by their passive membrane properties and expression of S100 beta and/or GFAP. Glutamatergic neurons and GABAergic interneurons were identified by their firing properties and respectively by the differential expression of the vesicular glutamate transporter, vGluT1, and the GABA synthesizing enzymes GAD65/67. In contrast to the terminal PGE_2 synthases, the terminal PGI_2 synthase was infrequently detected. This indicates that PGE_2 is the main vasodilatory prostanoids produced by these three parenchymal cell types. We also found that pyramidal cells are the main cell type equipped for PGE_2 synthesis, notably expressing the rate-limiting enzyme COX-2.
To evaluate the relative contribution of COX isoforms to neurovascular coupling we monitored in acute slices using videomicroscopy the vascular movements evoked by NMDA. We found that NMDA-induced vasodilations were reduced by COX-2, but not by COX-1 inhibition. Patch-clamp stimulations of single perivascular pyramidal cells were sufficient to elicit local vascular movements on neighboring arterioles.

These observations showed that pyramidal cells are the major source of COX-2-derived PGE_2. These results indicate that pyramidal cells are more important in neurovascular coupling than previously thought.

P1.041

Surface trafficking of the astroglial glutamate transporter GLT-1

Murphy Royal, C. (Bordeaux)[1,2,3], Dupuis, J. (Bordeaux)[2,3], Pinson, B. (Bordeaux)[3,4], Baufreton, J. (Bordeaux)[3,5], Groc, L. (Bordeaux)[2,3], Oliet, S. (Bordeaux)[1,3]
[1]Neurocentre Magendie - INSERM U.862, Interactions Glie-Neurones, Bordeaux, France, [2]Institut Interdisciplinaire de Neurosciences, CNRS UMR 5297, Bordeaux, France, [3]Université Bordeaux 2, Bordeaux, France, [4]Institut de Biochimie et Génétique Cellulaires, UMR 5095, Bordeaux, France, [5]Institut des Maladies Neurodegeneratives, UMR 5293, Bordeaux, France

Glutamate is the major excitatory neurotransmitter in the brain. Its concentration in the synaptic area is highly regulated in order to maintain point-to-point transmission and to prevent excitotoxicity associated with chronic activation of glutamate receptors. As there is no endogenous extracellular enzyme to degrade glutamate, the clearance of glutamate is ensured by transporters located on the surface of astrocytes. Among these surface transporters GLT-1 ensures almost 90% of glutamate uptake at synapses.
It is widely accepted that many receptors and transporters on the surface of neurons and glia display surface trafficking properties and are not simply static proteins. The basic surface trafficking properties of glutamate receptors at the synapse have been studied in depth and have allowed us to understand many important trafficking events that occur during synaptic plasticity. The action of GLT-1 is fundamentally important in synaptic communication. Surface trafficking of this transporter may play a pivotal role in the spatial and temporal properties of glutamate diffusion at the synapse. However, it is still unknown whether GLT-1 is dynamically regulated on the surface of astrocytes. Thus we set out to uncover the surface trafficking of GLT-1 in astrocytes by using a combination of high-resolution imaging, such as single particle (Quantum Dot [QD]) tracking, molecular biology and electrophysiological approaches.
Here we have demonstrated that GLT-1 is indeed subject to surface trafficking on astrocytes. Using pharmacological approaches we have shown that this diffusion is subject to regulation by the activity of the transporter itself as well as neuronal activity. Furthermore, we have demonstrated that the speed of GLT-1 diffusion is greatly reduced at excitatory synapses. This leads us to the conclusion that GLT-1 surface diffusion is highly regulated at the synapse where it can effectively carry out its role as the principal glutamate transporter at the synapse. Finally, using electrophysiology we have demonstrated that GLT-1 surface trafficking has an implicit role in regulating basal neuronal activity through glutamate uptake at the synapse.

P1.042

Characterization of a novel mouse model to study the zinc modulation of Cav3.2 T-type calcium channels

Voisin, T. (Montpellier)[1], Bourinet, E. (Montpellier)[1], Lory, P. (Montpellier)[1]
[1]Institut de Génomique Fonctionelle, Montpellier, France

T-type Cav3.2 channels are voltage-gated calcium channels that play an important role in controlling neuronal excitability, particularly in dorsal root ganglia (DRG) neurons where they are involved in pain signaling. Cav3.2 channels are also expressed in thalamic and hippocampal neurons and likely

participate to epileptic seizures. How Cav3.2 channels are regulated in neurons is therefore important to understand. Interestingly, studies on recombinant Cav3 channels have revealed that Cav3.2 channels are preferentially inhibited by low concentrations of zinc (IC50 ≈ 0,8 µM), and that the mutation of a single histidine residue at position 191 severely attenuated zinc sensitivity of Cav3.2. In order to study the zinc action on Cav3.2 channels *in vivo*, which is hypothesized to impact the activity of DRG neurons, we have generated a knock-in mouse carrying this mutation (H191Q). Electrophysiological studies were done on a specific type of DRG neurons, D-hair cells. We found that these neurons from the H191Q-Cav3.2 mice have lost their sensitivity to zinc. To investigate further the role of this modulation *in vivo*, we conducted behavioral studies. These H191Q-Cav3.2 mice showed no alteration of their locomotor phenotype. Preliminary studies on this animal model to test the pain phenotype are ongoing. We looked at the thermal pain perception with a ramp test on a hot plate, where the H191Q-Cav3.2 mice reacted in the same way as the control mice. The first tests we did to evaluate mechanical pain perception (Randall-Selitto test, Von Frey test) were not able to detect any significant differences between the KI mice and their littermates. Additional experiments are needed to characterize further the pain phenotype of the H191Q-Cav3.2 mice.

P1.043

A transcriptomic and genetic approach to identify molecular mechanisms underlying wakefulness

Seugnet, L. (Lyon)[1], Anaclet, C. (Lyon)[1], Perier, M. (Lyon)[1], Akaoka, H. (Lyon)[1], Lin, J.S. (Lyon)[1]
[1]*Equipe Physiologie intégrée du système d'éveil, Centre de Recherche en Neurosciences de Lyon, INSERM U1028 / CNRS UMR 5292 / UCBL1, Lyon, France*

Using KO mouse models lacking either histamine (HA, Hdc KO) or orexins (Ox KO), we have previously shown that HA and Ox act distinctly and synergistically in terms of wake control. HA is mainly responsible for the qualitative cognitive aspect, its deficit leading to somnolence, whereas orexins are more involved in behavioural and locomotive aspects, their defect causing narcoleptic phenotypes. Importantly, inactivating both HA and Ox results in more defects than the elimination of either system alone, revealing synergistic interactions. The significant wake deficits of the Hdc and Ox double KO (HO KO) mouse, notably somnolence, hypersomnia and EEG deficiency, make it a particularly appropriate model for the identification of new molecular and cellular pathways involved in wake control. To achieve this goal, we have used whole genome profiling to compare cortical gene expression between HO KO mice and their wild type littermates. We found over 200 genes differentially expressed between the two genotypes and notably
1) an increase in cholinergic muscarinic receptors, confirming our own unpublished data suggesting a compensatory over activation of the cholinergic system in HA deficient models.
2) an over-expression of the 5HT-5B receptor, which is thought to function as an auto-receptor co-expressed with the serotonin transporter
3) an up-regulation of the non-coding truncated *hdc* gene, suggesting the existence of a negative feed-back loop regulating the expression of *hdc*, potentially mediated by histamine H3 receptor activity.
The vast majority of the other differentially expressed genes in HO KO mice have not been previously linked to sleep and waking. Because a thorough screening of these genes is challenging in mammalian systems, we use the *Drosophila* model which is well suited for this experimental approach. Sleep in *Drosophila* exhibits many key similarities with mammalian sleep, and can be monitored in thousands of individual animals by automated locomotion detection systems. *Drosophila* genes can be activated or invalidated in a stage and tissue specific way using the Gal4-UAS system. Initial data obtained in this functional screen will be presented.

P1.044

Lack of evidence for vesicular glutamate transporter expression in mouse astrocytes

Herzog, E. (Bordeaux)[1], Li, D. (Paris)[2], Hérault, K. (Paris)[2], Silm, K. (Paris)[3], Evrard, A. (Paris)[2], Wocik, S.M. (Göttingen)[4], Oheim, M. (Paris)[2], Ropert, N. (Paris)[2]
[1]Université de Bordeaux, Institut Interdisciplinaire de Neuroscience, CNRS UMR 5297, Bordeaux, France, [2]INSERM U603, CNRS UMR 8154, Université Paris Descartes, Laboratoire de Neurophysiologie et Nouvelles Microscopies, Paris, France, [3]INSERM U952; CNRS UMR7224; Université Pierre et Marie Curie, Physiopathologie des Maladies du Système Nerveux Central, Paris, France, [4]Max-Planck-Institut für Experimentelle Medizin, Molecular Neurobiology, Göttingen, Germany

The concept of a tripartite synapse including a presynaptic terminal, a postsynaptic spine, and an astrocytic process that responds to neuronal activity by fast gliotransmitter release, confers to the electrically silent astrocytes an active role in information processing. However, the mechanisms of gliotransmitter release are still highly controversial. The reported expression of all three vesicular glutamate transporters (VGLUT1 3) by astrocytes suggests that astrocytes, like neurons, may release glutamate by exocytosis. However, the demonstration of astrocytic VGLUT expression is largely based on immunostaining, and the possibility of nonspecific labeling needs to be systematically addressed. We therefore examined the expression of VGLUT1-3 in astrocytes, both in culture and *in situ*. We used single-vesicle imaging by total internal reflection fluorescence microscopy in live cultured astrocytes and Western blots, as well as confocal microscopy at the cellular level in cortical, hippocampal, and cerebellar brain slices, combined with quantitative image analysis. Control experiments were systematically performed using wild-type, VGLUT1-3 knock-out, VGLUT1Venus knockin, and VGLUT2-EGFP transgenic mice. We quantified the degree of overlap between VGLUT1-3 and neuronal or astrocytic markers, both in an object-based manner using fluorescence line profiles, and in a pixel-based manner using dual-color scatter plots followed by the calculation of Pearson's correlation coefficient over all pixels with intensities significantly different from background. Our data provide no evidence in favor of the expression of any of the three VGLUTs by gray matter protoplasmic astrocytes in the primary somatosensory cortex, the thalamic ventrobasal nucleus, the hippocampus, and the cerebellum.

P1.045

Physiological involvement of presynaptic L-type voltage dependent calcium channels in GABA release of cerebellar molecular layer interneurons

Rey, S. (Paris)[1], Jalil, A. (Paris)[1], Maton, G. (Paris)[1], Satake, S. (Okazaki)[2], Collin, T. (Paris)[1]
[1]Université Paris Descartes, UMR 8118, Laboratoire de Physiologie Cérébrale, Paris, France, [2]National Institute for Physiological Sciences (NIPS), Departement of Information Physiology, Okazaki, Japan

Fast release of neurotransmitter is elicited by depolarization of the presynaptic compartment and Ca^{2+} entry into the terminal through voltage dependent Ca^{2+} channels (VDCCs). In cerebellar molecular interneurons (MLIs), only P/Q and N -types high threshold VDCCs have been shown to play a pivotal role in the release of GABA (Forti et al., 2000; Stephens et al., 2001). Although pharmacologically identified at the membrane of these neurons, L-types VDCCs have not been extensively studied. Two types of L- types VDCCs, Cav1.2 and Cav1.3 are expressed in the central nervous system. They are currently thought to mediate Ca^{2+}-induced Ca^{2+} release (CICR) processes at MLIs somata through a conformational association with ryanodine receptors (Chavas et al., 2004) and involvement in neurotransmitter release has been ruled out either by electrophysiology and Ca^{2+} imaging (Forti et al., 2000; Stephens et al., 2001). Our electrophysiological data confirm that P/Q (Cav2.1) , N (Cav2.2) and L-type (Cav1.2/3) VDCCs are functionally expressed in MLIs. Moreover, mRNA encoding Cav2.1, Cav2.2, Cav1.2 and Cav1.3 have been detected in MLI by single-cell RT-PCR. The use of various concentrations of Isradipine (Dihydropyridine (DHP) antagonist) suggests the presence of both L-type VDCCs Cav1.2 and Cav1.3 which are also positively modulated in the presence of BayK8644 (DHP agonist). Interestingly, BayK8644 (1-10 µM) dramatically increases mIPSCs frequency indicating that

L-type VDCCs could be present in presynaptic sites although this has been ruled out by previous studies. Action potential-evoked Ca^{2+} transients recorded in axonal varicosities exhibit a larger amplitude and slower decay kinetics in the presence of BayK8644. Using a GABAergic presynaptic marker (VGAT), we show a colocalisation between Cav1.2, Cav1.3 and VGAT. Moreover, some electrophysiological experiments show a potential coupling between L-type VDCCs and ryanodine receptors in MLI by a Calcium-Induced-Calcium-Release (CICR) or/and a physical coupling. Taken together, our data indicate demonstrate that L-type VDCCs are functionally expressed in the presynaptic compartment and are potentially involved in the release of GABA.

P1.046

The role of mitochondrial CB1 receptors in the hippocampus

Desprez, T. (Bordeaux)[1], Wadleigh, A. (Bordeaux)[1], Soria-Gomez, E. (Bordeaux)[1], Bellocchio, L. (Bordeaux)[1], Delamarre, A. (Bordeaux)[1], Hebert-Chatelain, E. (Bordeaux)[1], Puente, N. (Bilbao)[2], Grandes, P. (Bilbao)[2], Massa, F. (Bordeaux)[1], Benard, G. (Bordeaux)[3], Marsicano, G. (Bordeaux)[1]
[1]INSERM U.862, Neurocentre Magendie, Endocannabinoids and Neuroadaptation, Bordeaux, France, [2]Facultad de Medicina y Odontologia, Universidad del Pais Vasco, Neuroanatomy, Bilbao, Spain, [3]Université de Bordeaux, Maladies Rares: Génétique et Métabolisme (MRGM), EA 4576, Bordeaux, France

Cannabinoid Type-1 (CB1) receptor plays a crucial role in learning and memory by regulating neuronal activity and synaptic plasticity in the hippocampus. In this brain region, our laboratory has found that CB1 is also present on the mitochondrial outer membrane (MOM). Hereby, CB1 on mitochondria (mt-CB1) reduces mitochondrial activity and participates to depolarization induced suppression of inhibition (DSI), a form of hippocampal neuro-plasticity (Bénard et al., 2012). To selectively manipulate mtCB1 from cell membrane CB1 we create two models, a pharmacologic and a genetic. The former one consists in using CB1 agonist which can either cross or not the plasma membrane. The latter approach was to delete the first 22 amino acid of the CB1 to suppress the expression of CB1 on mitochondria. Here we found that:
1. mtCB1 are specifically controlling mitochondria metabolism by inhibiting 20% of mitochondrial respiration and
2. Hippocampal mtCB1 might be necessary for CB1 stimulation induced impairment of memory consolidation.
Further experiments are in process to examine the specific mechanism of mtCB1-mediated the inhibition of mitochondrial respiration and the impact of mtCB1 on neurotransmission and plasticity synaptic.

P1.047

Plexin-A1, a novel receptor of commissural projections for Slit family members

Delloye-Bourgeois, C. (Villeurbanne)[1], Jacquier, A. (Villeurbanne)[1], Charoy, C. (Villeurbanne)[1], Reynaud, F. (Villeurbanne)[1], Thoinet, K. (Villeurbanne)[1], Kindbeiter, K. (Villeurbanne)[1], Falk, J. (Villeurbanne)[1], Castellani, V. (Villeurbanne)[1]
[1]Université Claude Bernard Lyon 1, CGphiMC, Equipe Neuro-Developpement et Signalisation, Villeurbanne, France

We recently reported that PlexinA1, the signaling receptor for the midline repellent Semaphorin3B, plays an important role during commissural axon guidance. Its regulated processing controls transition from insensitive pre-crossing to responsive post-crossing growth cone state. Our previous analysis of

commissural axons trajectories in PlexinA1-deficient embryos revealed a variety of crossing and post-crossing defects, consistent with its functional implication in the Sema3B signaling (Nawabi *et al.*, 2010). We now report that a significant proportion of commissural axons also turned back in the FP, in the context of PlexinA1- but not Sema3B-deficiency. Similar re-crossing behaviors were observed after manipulations of the PlexinA signaling pathway in the chick neural tube. These phenotypes raised the intriguing possibility that PlexinA1 could share some functional links with the Robo/Slit signaling, whose alterations in mice induce midline re-crossing of commissural axons (Long *et al.*, 2004). We explored this hypothesis using various approaches and provide evidence that PlexinA1 binds Slit family members *in vitro* and *in vivo*, independently of Robo receptors. Notably, embryos lacking Robo1/2 receptors were reported to present remnant crossing commissural projections without any FP recrossing, leading the authors to postulate the existence of a yet unidentified Slit receptor (Jaworski *et al.*, 2010). Our data thus suggest that PlexinA1 might be this missing Slit receptor. Altogether, our studies identify a sequential and integrative molecular program in which PlexinA1 is first processed to silence commissural axon sensitivity to midline repellents allowing FP crossing, then restored at the cell surface to participate in the repulsive signaling triggering FP exit and preventing midline re-crossing.

P1.048

Opa-1 and Drp-1: deregulation of both mitochondria-shaping proteins contributes to apoptosis in an *in vitro* and *in vivo* manganese-induced parkinsonism models

Alaimo, A. (Buenos Aires)[1], Gorojod, R.M. (Buenos Aires)[1], Saravia, F.E. (Buenos Aires)[2], Kotler, M.L. (Buenos Aires)[1]
[1]*Faculty of Exact and Natural Sciences, University of Buenos Aires. IQUIBICEN-CONICET, Laboratory of Apoptosis in Nervous System/Nano-Oncology. Department of Biological Chemistry, Buenos Aires, Argentina,* [2]*Faculty of Exact and Natural Sciences, University of Buenos Aires.IBYME-CONICET, Laboratory of Neurobiology. Departamento de Química Biologica, Buenos Aires, Argentina*

Mitochondria are dynamic organelles which are organized as an interconnected network that fluctuates between two opposite processes that control their morphology: fission and fusion. These last events are regulated by a specific battery of mitochondria-shaping proteins (e.g. Opa-1 and Drp-1) whose changes in their expression or localization alter mitochondrial morphology and may promote apoptosis. Increasing evidence correlates mitochondrial dynamics alteration with the occurrence of neurodegenerative diseases. Therefore, we focused on this topic in manganese (Mn)-induced Parkinsonism termed Manganism. This neurodegenerative pathology is produced by excessive Mn accumulation, preferentially, in the basal ganglia and particularly, in the mitochondria of astrocytes. We have previously demonstrated the apoptotic signalling pathways triggered in rat C6 astrocyte-like cells treated with 750µM $MnCl_2$ (24h). In the present report, using the fluorescent probe MitoTracker Red CMXRos, we observed and quantified an increased mitochondrial network fission ($p< 0.001$) and $\Delta\Psi m$ dissipation ($p< 0.001$) in Mn-treated C6 cells. By western blot analysis and immunocytochemistry we showed that Mn induced a decreased expression of the fusion protein Opa-1 ($p< 0.05$) and its release into the cytosol, as well as an increased protein expression of fission Drp-1 ($p< 0.001$) coupled with its translocation to mitochondria. The role of both proteins in the Mn-induced apoptosis was studied using Drp-1 inhibitors (Cyclosporine A and Mdivi-1) and overexpressing Opa-1 levels. The results indicated that CsA, Mdivi-1, WT Opa-1 and Q297V Opa-1 (gain of function mutant) prevented cell death in a 25, 30, 20 and 25%, respectively (MTT assay, $p< 0.01$). Likewise, we quantified a 35, 30, 20 and 20% ($p< 0.001$) of cells with intact mitochondrial networks and $\Delta\Psi m$ ($p< 0.001$) and a 40, 40, 35 and 30% ($p< 0.001$) of prevention in the appearance of apoptotic nuclei detected with the fluorescent probe Hoechst 33258. In conclusion, these results showed that an imbalance in the mitochondrial dynamics which leads to an increased fission had a pivotal role in the apoptotic cell death induced by Mn in astroglial cells.

P1.049

Glutamate-stimulated water fluxes in cultured cortical neurons: involvement of Na^+/Ca^{2+} exchanger and ionic co-transporters in neuronal volume regulation

Jourdain, P. (Lausanne)[1], Pavillon, N. (Lausanne)[2], Boss, D. (Lausanne)[1], Depeursinge, C. (Lausanne)[2], Magistretti, P.J. (Lausanne)[1,3], Marquet, P. (Lausanne)[3]
[1]Ecole Polytechnique Fédérale de Lausanne, EPFL SV BMI LNDC, Lausanne, Switzerland, [2]Ecole Polytechnique Fédérale de Lausanne, EPFL STI-DO, Lausanne, Switzerland, [3]DP-CHUV, Centre de Neurosciences Psychiatriques, Lausanne, Switzerland

To prevent damage derived from cerebral cell swelling or shrinkage, it is crucial to tightly regulate the water transport. To better understand mechanisms of cell volume regulation, we set out to study in cultured cortical neurons the water fluxes coupled to the $[Na^+]_i$ and $[Ca^{2+}]_i$ changes during a glutamatergic stimulation, using a multimodality approach combining epifluorescence and Digital Holographic Microscopy, providing quantitative phase signal sensitive to transmembrane water movements. After a brief application of glutamate (30µM, 30s), we recorded 2 main types of phase signal: biphasic and reversible decrease responses. These optical signals could be decomposed in 2 distinguished periods: Phase decrease stage (P1) associated with a water influx and Phase recovery stage (P2) linked to the water efflux. The simultaneous recordings of phase signals, $[Na^+]_i$ and $[Ca^{2+}]_i$ changes showed that P1 was concomitant to a $[Na^+]_i$ and $[Ca^{2+}]_i$ peaks indicating that the trans-membrane water entry is initially correlated to a strong cation influx through the glutamate receptor ion channel. In Ca^{2+} free medium, phase signals and $[Na^+]_i$ changes persisted while the $[Ca^{2+}]_i$ peaks disappeared demonstrating than water fluxes are mainly driven by the Na^+ movements. With furosemide (100µM) a blocker of ionic co-transporters, the amplitude of phase signals was significantly decreased and effects on Na^+ and Ca^{2+} signals were weak, confirming the major contribution of ionic co-transporters to water fluxes. A specific blocker of the Na^+/Ca^{2+} exchanger (NCX), CB-DMB (5µM) drastically decreased the $[Na^+]_i$ peaks indicating the main role of NCX in Na^+ influx. These results indicate that, if the water and Na+ movements are concomitant, their “routes” are distinct. Finally, with CB-DMB, neuronal cells continued to display a strong decrease of phase signals (pathological signal) and the $[Ca^{2+}]_i$ could be maintained to a high concentration suggesting that ionic co-transporters, in particular NKCC1, a protein sensitive to the $[Ca^{2+}]_i$ changes, transported actively water in the cell (until osmolysis). These data demonstrate that $[Ca^{2+}]_i$ changes play a modulatory role in water fluxes and confirm that NCX ensures the functional relationship between the triggering signal and the final effector.

P1.050

Stimulation of the endocannabinoid system prevents methamphetamine-induced neurotoxicity in mice

Thiriet, N. (Poitiers)[1], Nader, J. (Poitiers)[1], Rapino, C. (Teramo)[2], Gennequin, B. (Poitiers)[1], Francheteau, M. (Poitiers)[1], Makryiannis, A. (Boston)[3], Duranti, A. (Urbino)[4], Maccarrone, M. (Rome)[5], Solinas, M. (Poitiers)[1]
[1]Univ. de Poitiers, INSERM U1084 LNEC, Pôle Biologie Santé - Building B36, Poitiers, France, [2]Dipartimento di Scienze Biomediche, Università degli Studi di Teramo, Teramo, Italy, [3]Center for Drug Discovery, Department of Pharmaceutical Sciences, and Department of Chemistry and Chemical Biology, Northeastern University, Boston, United States, [4]Dipartimento di Scienze Biomolecolari, Università degli Studi di Urbino “Carlo Bo”, Urbino, Italy, [5]Center of Integrated Research, Campus Bio-Medico University of Rome, Rome, Italy

The endocannabinoid system (ECS) is a neuromodulatory system, involving two main ligands, namely *N*-arachidonoylethanolamine (anandamide) and 2-arachidonoylglycerol (2-AG). Endocannabinoids (ECs) are synthesized from lipid precursors, and have the unique property to be released “on demand” by stimulated cells. Evidences suggest that they could act as neuroprotective agents under different pathological situations. Methamphetamine (METH) administration induces in the striatum significant dopamine neuron terminals loss similar to what is found in other neurodegenerative processes**.** We undertook this study to investigate whether the activation of the ECS could prevent the neurotoxicity

induced by METH. We stimulated the ECS 40 min before the administration of a toxic dose of METH (30 mg/kg i.p.) by administering
1) the exogenous cannabinoid tetrahydrocannabinol (THC, 3 mg/kg) or
2) inhibitors of degradation enzymes for anandamide (URB597, 1 mg/kg) or 2-AG (JZL184, 16 mg/kg).
We observed that all these treatments blunted the decreases of TH levels induced by METH in the striatum. The use of EC receptor specific antagonists revealed a main involvement of the CB2, but not the CB1, receptor subtype in this protective effect. To test a possible action of the ECs on the inflammatory reaction triggered by METH, we measured the TNF alpha accumulation 1 day after METH injection. We observed that this accumulation was also reduced when the ECS was activated 40 min before the METH injection. Taken together, these results suggest that the ECS can indeed act as a neuroprotective factor against METH-induced neurotoxicity and provide support for the role of CB2 receptors in the regulation of the neuroinflammation reactions that participate in these neurotoxic processes. These results also suggest that the stimulation of the ECS could represent an interesting therapeutic approach in the case of exogenous or endogenous insults to the brain.

P1.051

Modulation of metabotropic glutamate receptors by chloride: new insights for glutamatergic synapse regulation?

Tora, A. (Montpellier)[1], Rovira, X. (Montpellier)[1], Doyon, N. (Québec)[2], Brabet, I. (Montpellier)[1], De Koninck, Y. (Québec)[2], Pin, J.P. (Montpellier)[1], Acher, F. (Paris)[3], Goudet, C. (Montpellier)[1]
[1]Institut de Génomique Fonctionnelle, CNRS UMR 5203-INSERM U661, Dépt. de pharmacologie moléculaire, Montpellier, France, [2]Institut Universitaire de santé mentale de Québec, Unité de neurosciences cellulaires et moléculaires, Québec, Canada, [3]Laboratoire de Chimie & Biochimie Pharmacologiques et Toxicologiques, CNRS UMR 8601, Paris, France

Metabotropic glutamate receptors (mGluRs) are G protein-coupled receptors (GPCRs), mainly expressed in the central nervous system. According to their location in the synaptic cleft, they modulate glutamatergic transmission, leading to increases in neuronal excitability, potentiation of excitatory post synaptic currents or on the opposite, to feedback mechanisms of glutamate release. Their involvement in many neurological disorders such as chronic pain, epilepsy, schizophrenia and Parkinson disease, explains the rise in the interest for the development of pharmaceutical agents targeting these receptors.
As class C GPCRs, mGluRs display a large extracellular domain, called Venus Flytrap domain (VFT), which contains the orthosteric ligand binding site. A cystein rich domain connects the VFT to the seven transmembrane spanning helices, where allosteric compounds bind and modulate intracellular signaling pathways.
Similarly, we have demonstrated that small extracellular compounds, i.e. chloride ions, can behave like allosteric modulators, but binding to the VFT and thus may modulate mGluRs functional responses in a concentration dependent manner.
Thanks to computational modeling and directed mutagenesis we have identified critical residues involved in the chloride sensitivity in both mGlu4 and mGlu2. Our data suggest that sensitivity towards chloride is not the same for all mGluRs, leading to a fine tune in the activation of these receptors and a potential switch-off mechanism preventing excessive or weak glutamatergic transmission.

P1.052

Tonic activated striatal DA release by lesions of the nigro-striatal DA pathway

Leviel, V. (Bron)[1], Dzahini, K. (Bron)[1], Wilson, C. (Bron)[1], Vézoly, J. (Bron)[1], Costes, N. (Lyon)[2], Dentresangle, C. (Lyon)[3], Hassoun, W. (Lyon)[3], Procyk, E. (Bron)[1]
[1]INSERM U846, SBRI, Bron, France, [2]CERMEP, Lyon, France, [3]CNRS, UMR 5542, Lyon, France

Plastic alterations of the Dopamine (DA) neurotransmission are produced by very partial lesions of the nigro-striatal DA pathway which might simulate mechanisms underlying the initial asymptomatic phases of Parkinson´s disease (PD).
1-After restricted and lateral nigral injections of 6-OHDA, a DA-cell toxin, a lateral denervated part of the ipsi lateral caudate nucleus and a medial spared region with no denervation can be delineated. Extracellular striatal DA and glutamate (GLU) were measured using microdialysis collection and HPLC-ED analyses. Tonic increases were observed, moderate in the lateral striatum and drastic in the medial part. This could be antagonized by a chronic treatment with neuroprotective agents including Riluzol, Amantadine and Memantine three substances also known to counteract GLU neurotransmission. The phasic DA overflow induced by electrical stimulation of nigro-striatal path remained unmodified in both regions.
2- On striatal synaptosomes prepared from rats injected three weeks before with 6-OHDA, the [3H]DA uptake velocity was found to be increased due to an increased affinity (reduced Km) of the DA transporter (DAT). The number of carrying sites was simultaneously reduced. This effect could be antagonized by a chronic treatment with MK801 (NMDA receptor blocker).
3-The binding potential (BP) of [^{11}C]-PE2i, a specific ligand for DAT, was measured longitudinally by Positon Emission Tomography (PET) imaging in four cynomolgus monkeys. It was found to be enhanced in the whole striatum (confirmed by SPM analysis) soon after the onset of a MPTP treatment, and before the onset of any significant motor symptoms. This showed a persistent DAT externalization at the beginning of MPTP treatment.
The DAT is now admitted to be responsible for the tonic striatal DA homeostasis through a DA reverse transport of the amine. The present observations suggest that initial and very partial nigro-striatal DA pathway lesion results in the activation, in the striatum of an hypertonic basal DA release. This homeostatic regulation appears independent from the proper activity of the nigro-striatal DA pathway submitted to an indirect control by GLU neurotransmission, locally in the region of projection.

P1.053
Multifaceted roles of lactate in the cerebral cortex

Cauli, B. (Paris)[1], Karagiannis, A. (Paris)[1], Lacroix, A. (Paris)[1], Plaisier, F. (Paris)[1], Belliveau, L. (Paris)[1], Hepp, R. (Paris)[1], Gallopin, T. (Paris)[2], Geoffroy, H. (Paris)[2], David, C. (Göttingen)[3], Jarlier, F. (Paris)[1], L'Heureux, B. (New York)[4], Lambolez, B. (Paris)[1], Staiger, J.F. (Göttingen)[3], Seino, S. (Kobe)[5], Hillman, E.M.C. (New York)[4], Roeper, J. (Frankfurt)[6]
[1]CNRS UMR 7102, Neurobiology of Adaptive Processes, Paris VI University, Paris, France, [2]Neurobiology, ESPCI ParisTech, CNRS UMR 7637, Paris, France, [3]Neuroanatomy, Georg August University, Göttingen, Germany, [4]Biomedical Engineering, Columbia University, New York, United States, [5]Physiology and Cell Biology, Kobe University, Kobe, Japan, [6]Institute of Neurophysiology, Goethe-University, Frankfurt, Germany

Tight coupling between neuronal activity and energy supply is fundamental for normal brain function. In the cerebral cortex, energy substrates fluxes evoked by neuronal activity lead to a transient lactate surge while glucose levels remain constant. However, the relative contribution of these energy substrates to ATP production is controversial and the functional consequences of an increased energy supply on neuronal activity are largely unexplored.
Using intracellular imaging of ATP with genetically encoded FRET sensors we observed that the most of the ATP produced by cortical neurons is derived from oxidative phosphorylation and not from glycolysis. By means of intrinsic fluorescence imaging of NADH we showed that cortical neurons have the capacity to take up and oxidize lactate, two prerequisites to lactate oxidative metabolism.
To evaluate the influence of energy substrates on neuronal activity we pay attention on ATP-sensitive K^+ (K-ATP) channels, which couple intracellular ATP/ADP ratio to membrane excitability. Monitoring the response of neuronal K-ATP channels to changes in extracellular energy supply offers

opportunities to assess the relative contribution of these energy substrates to ATP synthesis and to decipher their potential impact on neuronal activity.
By combining patch-clamp recordings, single-cell RT-PCR and pharmacology in rodent acute cortical slices, we found that both glutamatergic and GABAergic neurons express K-ATP channels composed of Kir6.2 and SUR1 subunits. Consistently, we observed that cortical neurons have the capacity to sense intracellular ATP levels by modulating their K-ATP channels. Using perforated-patch recordings to preserve intracellular metabolism, we revealed that an increased energy supply via lactate, but not via glucose, exacerbates neuronal activity. Using pharmacological manipulations and Kir6.2$^{-/-}$ mice we also demonstrated that lactate-sensing involves a facilitated transport of lactate and a subsequent closure of K-ATP channels.
Our investigations disclosed that cortical neurons act as lactate sensors but not as glucose sensors. They also indicate that lactate, most likely supplied by astrocytes, is a major oxidative substrate for neurons and support the astrocyte-to-neuron lactate shuttle hypothesis.

P1.054

A proteomic and functional analysis reveals that 5-HT6 receptors modulate neuronal differentiation by recruitment of Cyclin-dependant kinase 5

Duhr, F. (Montpellier)[1], Séveno, M. (Montpellier)[2], Mannoury la Cour, C. (Croissy sur Seine)[3], Dupuis, D. (Croissy sur Seine)[3], Millan, M.J. (Croissy sur Seine)[3], Bockaert, J. (Montpellier)[1], Marin, P. (Montpellier)[1,2], Chaumont-Dubel, S. (Montpellier)[1,2]
[1]Institut de Génomique Fonctionnelle, Dpt. de Neurobiologie, Montpellier, France, [2]Institut de Génomique Fonctionnelle, Plateforme de Protéomique Fonctionnelle, Montpellier, France, [3]Institut de Recherche Servier, Dpt. de Psychopharmacologie, Croissy sur Seine, France

The serotonin 5-HT$_6$ receptor is expressed in CNS regions involved in the pathogenesis of disorders like schizophrenia. 5-HT$_6$ receptors are a promising target for treatment of the accompanying cognitive deficits, since their blockade consistently enhances mnemonic performance in rodents. Paradoxically, still little is known about 5-HT$_6$ receptor-associated signalling pathways, an issue we have addressed by a proteomic approach. Previously, we demonstrated physical interaction of 5-HT$_6$ receptors with several members of the mTOR pathway and that mTOR recruitment by prefrontal 5-HT$_6$ receptors perturbs cognition in schizophrenia. Here we show that 5-HT$_6$ receptors interact with Cyclin-dependent kinase (Cdk)5, a protein which controls actin cytoskeleton dynamics and modulates neurodevelopmental processes.
Expressing the 5-HT$_6$ receptor in neuroblastoma NG108-15 cells triggered morphological and functional changes associated with neuronal differentiation. These effects were not further enhanced by exposure to an agonist (WAY181187, 1 µM) and were prevented when cells were treated with SB258585 (10 µM), a selective 5-HT$_6$ antagonist. Expression of a dominant-negative Cdk5 or treating cells with roscovitine (a pharmacological inhibitor of Cdk5) likewise inhibited NG108-15 cells differentiation induced by 5-HT$_6$ receptor expression. SB258585 also impaired the association of 5-HT$_6$ receptor with Cdk5 in NG108-15 cells, as determined by co-immunoprecipitation, suggesting that this interaction was necessary for induction of differentiation. Treating striatal neurons with SB258585 decreased neurite length, as determined after 24 hrs. Further supporting a role of endogenously expressed receptors in differentiation of striatal neurons, silencing 5-HT$_6$ receptor expression in cultured neurons significantly reduced neurite length.
The present data show that 5-HT$_6$ receptors promote neuronal differentiation and reveal a critical role for Cdk5 in this process. These novel insights into molecular substrates underlying neurodevelopmental effects of 5-HT$_6$ receptors are of potential importance to the pathophysiology of early-onset CNS conditions like autism-spectrum disorder and schizophrenia.

P1.055

Involvement of reactive oxygen species in glucose sensitivity of hypothalamic neuron

Chrétien, C. (Dijon)[1], Fenech, C. (Dijon)[1], Grall, S. (Dijon)[1], Pénicaud, L. (Dijon)[1], Leloup, C. (Dijon)[1], Fioramonti, X. (Dijon)[1]
[1]*Centre des Sciences du Goût et de l'Alimentation, UMR 6265 CNRS, 1324 INRA-uB, Dijon, France*

Glucose homeostasis is under control of central and peripheral regulatory mechanisms. The ability of the brain to sense changes in blood glucose levels is mainly accomplished within the ventromedial hypothalamus (VMH). The VMH presents specialized glucose-sensitive neurons suggested to be involved in the control of glucose homeostasis. Among them, high glucose-excited (HGE) neurons directly increase their electrical activity in response to increased glucose level. Despite the involvement of channels presenting a non-selective cationic conductance (NSCC), the molecular mechanisms involved in HGE neuron response to increased glucose level are unknown. We recently showed that mitochondrial reactive oxygen species (mROS) are produced during hyperglycemia in the VMH and are involved in hypothalamic detection of increased blood glucose level. Interestingly, some transient receptor potential (TRP) channels which present a NSCC are redox-sensitive and directly modulated by ROS. Thus, we hypothesize that mROS are involved in HGE neuron response to increased glucose level through a ROS-TRP signaling pathway. We used Fura-2 calcium imaging as a surrogate of neuronal electrical activity on freshly dissociated rat VMH cells in response to increased extracellular glucose level from 2.5 to 10 mM, mimicking hyperglycemia *in vitro*. We observed that 5 % of VMH cells were characterized as HGE neurons as they present a transient and reversible increase in free intracellular calcium concentration in response to increased glucose level. The number of HGE neurons decreased by 60% in presence of an antioxidant cocktail (Control: 5.00 ± 1.01 % *vs* Antioxidants (gluthation 0.1 mM + trolox 0.2 mM): 1.96 ± 1.60 %, $p < 0.05$). We also investigated the presence of redox-sensitive TRP channels by RT-qPCR and found that TRPC2, 3, 4 and TRPM2, 5, 7 channels are expressed in the VMH. We currently aim to confirm the involvement and expression of redox-sensitive TRP channels in HGE neurons using pharmacological approaches and single cell RT-qPCR post-calcium imaging. The hypothesis that HGE neurons produce themselves mROS in response to increased glucose level will also be tested. Altogether, these data suggest that ROS-TRP channel signaling is involved in VMH HGE glucose sensitivity.

P1.056
Ephaptic inhibition of Purkinje cells by the pinceau

Blot, A. (Paris)[1], Barbour, B. (Paris)[1]
[1]*IBENS, Cerebellum Team, CNRS UMR 8197 / INSERM U 1024 / ENS, Paris, France*

Excitatory synaptic activity in the brain is shaped and balanced by inhibition. Because inhibition cannot propagate, it must be recruited with a synaptic delay by incoming excitation. This configuration potentially exists in the cerebellar cortex, whose output is generated by Purkinje cells driven by long-range excitatory parallel fibre inputs, which also recruit local inhibitory basket cells. The axon initial segment of each Purkinje cell is ensheathed by basket cell axons in a structure called the pinceau, which has been suggested to mediate a form of electrical inhibition. However, no direct recordings of this presumed inhibitory action exist. Here we show that activation of the pinceau causes an extracellular positivity that inhibits Purkinje cell firing by hyperpolarising capacitively the axonal membrane. The reduction of firing rate is synchronous with the presynaptic action potential, eliminating a synaptic delay and allowing granule cells to inhibit Purkinje cells indirectly simultaneously with direct excitation. This constitutes the fastest intercellular inhibitory mechanism reported in the brain, implementing ultra-rapid feedforward and lateral inhibition.
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P1.057

Role of type 1 cannabinoid receptor (CB1) in the dynamics of hippocampal network revealed through Voltage Sensitive Dye Imaging

Colavita, M. (Bordeaux)[1,2,3], Doubeck, M. (Talence)[4], Wadleigh, A. (Bordeaux)[1], Drago, F. (Catania)[3], Iollo, A. (Talence)[4], Marsicano, G. (Bordeaux)[1,2], Massa, F. (Bordeaux)[1,2]
[1]INSERM U.862, Neurocentre Magendie 'Endocannabinoids and Neuroadaptation' Group, Bordeaux, France, [2]Université Bordeaux Ségalen, Bordeaux, France, [3]Università degli Studi di Catania, Department of Clinical and Experimental Pharmacology, Catania, Italy, [4]Institut de Mathématiques de Bordeaux, Université Bordeaux 1, INRIA, Talence, France

The functioning of the brain is guaranteed by millions of cells forming elaborate networks, whose activity underlies behaviour. A useful tool to visualize and quantify action potentials spreading across neuronal networks is Voltage-Sensitive Dye Imaging (VSDI). The core material of VSDI is the dye itself: It is an amphilic molecule which binds to the cell membranes with its hydrophobic tail. Once bound and excited with the appropriate wavelength, VSDs emit a fluorescent signal instantaneously when it detects a change in membrane potential.
The cannabinoid type-1 receptors (CB1) are widely expressed in the brain and have a strong impact on activity of both neurons and astroglial cells. The actions of CB1 receptors at single cell level could be reflected on a higher neuro-architectural level as neuronal networks. To assess this, we manipulate CB1 activity in hippocampal slices through pharmacological and advanced genetic tools in mice. Combining VSDI and sophisticated mathematical analysis of VSDI data, we report the alterations induced by these manipulations in the dynamics of network activity in hippocampus. Preliminary results demonstrate that this approach is feasible and that CB1 receptors modulate the dynamics of the hippocampal network at different levels.

P1.058

Role of CX3CR1 in supporting cell-to-cell contacts between microglia and degenerating TH+ neurons

Uhlrich, J. (Lyon)[1], Touret, M. (Lyon)[1], Virgone-Carlotta, A. (Lyon)[1], Watrin, C. (Lyon)[1], Pays, L. (Lyon)[1], Nataf, S. (Lyon)[1]
[1]INSERM U1028 -CNRS UMR 5292- Univ Lyon 1, Centre de Recherche en Neuroscience de Lyon, Neurooncology Neuroinflammation Team, Lyon, France

Numerous studies have investigated the molecular cross-talk between microglia and neurons in different models of Parkinson´s disease. A major role for neuronally-derived Fractalkine/CX3CL1 and its microglial receptor CX3CR1 has been demonstrated in the MPTP model of Parkinson´s disease. Such a molecular interaction inhibits microglial activation and prevents, at least in part, microglia-mediated neuronal cell death. However, whether CX3CL1/CX3CR1 supports cell-to-cell contacts between microglia and degenerating neurons is still unknown. We recently demonstrated different categories of neuron/microglia physical interactions in the murine 6-OHDA model of Parkinson´s disease. These included, in particular, microglial ramifications that penetrated the soma of TH+ neurons and supported microglial microphagocytosis of TH+ material. In the present work, we used Cx3cr1 KO mice ($Cx3cr1^{gfp/gfp}$ homozygous mice) and the murine 6-OHDA model to investigate the role of CX3CR1 in neuron/microglia physical interactions under neurodegenerative conditions. In addition, we mapped microglia/neuron cell-to-cell contacts in the substantia nigra of autopsy samples from Parkinson´s disease patients.

P1.059

GSA-10 and SAG exhibit distinct pharmacology at Smoothened

Hoch, L. (Gif sur Yvette)[1], Gorojankina, T. (Gif sur Yvette)[1], Faure, H. (Gif sur Yvette)[1], Roudaut, H. (Gif sur Yvette)[1], Traiffort, E. (Gif sur Yvette)[1], Girard, N. (Strasbourg)[2], Schoenfelder, A. (Strasbourg)[2], Mann, A. (Strasbourg)[2], Manetti, F. (Siena)[3], Solinas, A. (Siena)[3], Petricci, E. (Siena)[3], Taddei, M. (Siena)[3], Ruat, M. (Gif sur Yvette)[1]

[1]CNRS UPR 3294, Signal Transduction and Developmental Neuropharmacology Team, Laboratory of Neurobiology and Development, Gif sur Yvette, France, [2]CNRS UMR 7200, Therapeutic Innovation Laboratory, Strasbourg, France, [3]Univ. Siena, Department of Pharmaceutical and Applied Chemistry, Siena, Italy

Activation of the Smoothened (Smo) receptor mediates Hedgehog (Hh) signaling. Smo inhibitors such as GDC-0449 and LDE225 are candidates for the treatment of brain cancers associated with dysfunction of Hh signaling and the search for Smo antagonists is under intense study (Ruat et al, *Differentiation,* 2012). To allow the characterization of novel Smo modulators, we have developed a virtual screening strategy based on a Smo pharmacophore for antagonists which has led us to the characterization of the MRT families of potent Smo inhibitors (Manetti et al, *Mol Pharmacol*, 2010; Roudaut et al, *Mol Pharmacol*, 2011; Solinas et al, *J Med Chem*, 2012).
Now, based on the structure of SAG, a reference Smo agonist (Ng and Curran, *Nat Rev Cancer*, 2011), a similar strategy led us to the identification of GSA-10, a quinolinecarboxamide which displays Smo agonist activity in multipotent mesenchymal cell differentiation assay. Our data indicate the presence of two distinct binding sites for SAG and for GSA-10 on Smo named Smo^{SAG} and Smo^{GSA-10}, respectively. GSA-10 and SAG differ in their capacity to act at the canonical bodipy-cyclopamine binding site previously identified on Smo. Moreover, we provide a pharmacological discrimination of GSA-10- and SAG-induced responses at Smo as shown by the highly reduced sensitivity of GSA-10 to several Smo reference antagonists including GDC-0449, CUR61414 and cyclopamine. Since GDC-0449 and LDE225 display different antagonist potency at Smo^{SAG} and Smo^{GSA-10}, it will be important to investigate to what extent their effects in cancer are related to inhibition of Smo^{GSA-10} and to identify if blockade of this binding site participates in the side effects of these molecules in human (Sekulic et al, *NEJM,* 2012). Our findings demonstrate also a significant variability in Smo conformations induced by different ligands and which has clearly important implications for the development of novel and more selective therapeutic agents. Experiments are in progress to further delineate the physiological relevance of Smo^{SAG} and Smo^{GSA-10} in neural tissues during development and in the adult.

P1.060

Neuronal sub-domain dependent patterns of type-1 cannabinoid receptor signaling

Ladarre, D. (Paris)[1], Roland, A. (Paris)[1], Biedzinski, S. (Paris)[1], Lenkei, Z. (Paris)[1]
[1]Ecole Supérieure de Physique et de Chimie Industrielles, CNRS UMR 7637 - Lab. de Neurobiologie, Paris, France

Type-1 cannabinoid receptors (CB1Rs) are expressed on the axonal plasma membrane and are major presynaptic modulators of synaptic function. According to our previous results, newly-synthesized CB1Rs first transiently appear on the somatodendritic plasma membrane, from where they are rapidly removed by constitutive activation-dependent endocytosis, ultimately resulting in transcytotic delivery to the axonal plasma membrane where CB1Rs accumulate. These results have shown subdomain-dependent steady-state patterns of CB1R internalization in neurons, so here we asked how sub-neuronal localization impacts on cAMP/PKA signaling, which is the major signaling pathway of this $G_{i/o}$-protein coupled receptor in non-polarized cells. We used highly resolutive imaging of cultured rat hippocampal neurons and FRET-based sensors of cAMP and PKA activation. Application of agonist WIN55-212,2 led to pertussis toxin dependent, rapid and significant somatodendritic decrease of basal cAMP/PKA activity. Application of the antagonist/inverse agonist AM251 resulted in a marked rapid increase of cAMP/PKA activity both in soma and dendrites, which is blocked by inhibitors of the production of 2-AG, the major endocannabinoid. These results indicate that somatodendritic CB1Rs are activable by exogenous agonists and that a high cell-autonomous endocannabinoid tone leads to significant constitutive activation. Compared to dendrites, the application of agonist WIN55-212,2 led to a significantly stronger decrease of basal axonal PKA activity in the axon. Moreover, the application of the antagonist/inverse agonist AM251 had no effect in this compartment, showing that accumulation of CB1Rs on axonal membrane can be related to a decreased constitutive activation of the receptor. Thus, our results suggest that transient somatodendritic CB1Rs are constitutively activated by endocannabinoids leading to constitutive endocytosis and a constitutive somatodendritic inhibition of

cAMP/PKA signaling, whereas CB1Rs on the axonal membrane are not constitutively activated by cell-autonomous endocannabinoids, leading to their accumulation and to high signal-to-noise ratio signaling in presence of exogenous activation.

P1.061

Widespread catecholaminergic activation of the cAMP/PKA pathway in the rodent neocortex

Nomura, S. (Paris)[1], Hepp, R. (Paris)[1], Lambolez, B. (Paris)[1]
[1]*Université Pierre & Marie Curie - UPMC, CNRS UMR7102 Neurobiologie des processus adaptatifs, Paris, France*

Dopamine (DA) and Noradrenaline (NA) can increase neuronal excitability and modulate synaptic transmission via their effects on the cAMP/PKA signaling pathway. In the rodent cerebral cortex, NA is released by fibers originating from neurons of the locus coeruleus (LC) which innervate the entire neocortex. In contrast DA originates from neurons of the ventral tegmental area (VTA) projecting mainly to the medial prefrontal area and the anterior cingulate, rhinal and entorhinal cortices. Only scares innervation has been reported in other cortical areas such as the parietal and occipital cortices. According to binding assay and immunohistochemistry, DA receptors are nonetheless expressed in the entire cortex. The aim of this project is to characterize cAMP/PKA signals elicited by bath applied and endogenously released catecholamines in the neocortex using 2-photon imaging of genetically-encoded fluorescent PKA sensors expressed in the neurons by viral transfer. The cortical distribution of NA/DA-induced PKA signals in the rat cortex has been first examined by bath application of NA (10µM), β-adrenoceptor agonist Isoproterenol (1µM), DA (10µM) and the D1/D5 receptor agonist SKF 38393(1 µM). Activation of the PKA pathway in response to these drugs were observed along the entire rostro-caudal axis (i.e. in frontal, parietal and occipital cortices) in both layer 2/3 and layer 5 pyramidal neurons. The amplitude of the PKA responses were respectively 60% and 40% for NA and DA when compared to the maximal activation obtained with the cyclase activator forskolin. Responses to NA and DA were inhibited by their respective antagonists (β1 adrenoceptor antagonist CGP20712, 100nM; D1/D5 antagonist SCH 23390, 1 µM). NA or DA responses were potentiated by 30 to 40 % when Gi coupled receptors were blocked by yohimbine (1 µM, alpha2 adrenoceptors) or haloperidol (1 µM, D2-like receptors). These results show a widespread distribution of functional NA and DA receptors in the neocortex that extends far beyond the reported territory of VTA fiber innervation.

P1.062

Multi-sensing bioprobes for *in vivo* brain glucose and lactate monitoring

Meiller, A. (Lyon)[1], Vasylieva, N. (Lyon)[2,3], Sabac, A. (Lyon)[3], Barbier, D. (Lyon)[3], Marinesco, S. (Lyon)[1,2]
[1]*Université Lyon 1 - CRNL INSERM U1028 CNRS UMR5292, Plateforme NeuroChem, Lyon, France,* [2]*Université Lyon1 - CRNL INSERM U1028 CNRS UMR5292, Physiologie Intégrée du Système d'éveil, Lyon, France,* [3]*Institut de Nanotechnologie de Lyon, CNRS UMR-5270, INSA de Lyon, Lyon, France*

Simultaneous monitoring of glucose and lactate levels in the living brain is essential to assess cerebral energy metabolism and could become a diagnostic tool in acute human brain injury. Implantable enzymatic biosensors are widely used for *in vivo* monitoring applications; in particular in neuroscience research to detect neurotransmitters and metabolites with outstanding temporal resolution. These biosensors are often fabricated employing a platinum wire encased in a glass-capillary. Nevertheless, monitoring several biological markers requires the implantation of several electrodes leading to extended brain damage and alteration in physiological brain functioning.

Here, we report the successful fabrication of a silicon/polymer based multisensing probe integrating three-platinium microelectrodes for simultaneous glucose and lactate monitoring with reference to a control electrode. The development of such micro-device is a complex multistage process. Microprobes are batch fabricated using microelectromechanical system technology (MEMS). Absence of electrical cross-talk current between adjacent electrodes was verified after microprobe fabrication and absence of chemical cross-talk after enzyme deposition. Then, each biosensor was characterized for activity, stability at 37°C and linearity of response for glucose and lactate in concentration ranges found *in vivo*. Finally, the multisensing probes were implanted in the cortex of anesthetized rats and the changes in brain glucose concentration were reliably detected following insulin administration and glucose injection, confirming the concept of independent simultaneous glucose and lactate monitoring *in vivo*.
This new monitoring tool can be applied for more complex neurochemical studies, aiming at understanding the metabolic regulation of energy substrates in the brain.

P1.063

***In vivo* properties of excitatory synaptic connections between layer 2 pyramidal cells in mouse somatosensory cortex**

Poulet, J.F.A. (Berlin)[1,2], Jouhanneau, J.-S. (Berlin)[1,2]
[1]*Max-Delbrück Center for Molecular Medicine (MDC), Department of Neuroscience, Berlin, Germany,* [2]*NeuroCure, Cluster of Excellence, Neuroscience Research Center, Charité-Universitätsmedizin, Berlin, Germany*

Local, excitatory synaptic connections are critical for neocortical sensory processing and cognition and form the basis of large-scale computer models, however there are extremely limited *in vivo* data available on confirmed cortico-cortical monosynaptic connections. We used two-photon microscopy to make visually targeted recordings from 2-3 neighbouring (< 100um) pyramidal neurons in the somatosensory cortex of the urethane anaesthetised juvenile (P18-21) mouse to characterise basic properties of excitatory monosynaptic connections *in vivo*. We restricted our analysis to the hyperpolarized cortical “down” states where it was possible to see small amplitude unitary EPSPs triggered by single presynaptic action potentials. We found that excitatory synaptic connections between layer 2 pyramidal neurons *in vivo* are sparse and most are small amplitude, resembling excitatory connections found in previous brain slice studies.

P1.064

Discovery of GSA-10, a novel positive modulator of Smoothened

Faure, H. (Gif sur Yvette)[1], Gorojankina, T. (Gif sur Yvette)[1], Hoch, L. (Gif sur Yvette)[1], Traiffort, E. (Gif sur Yvette)[1], Schoenfelder, A. (Strasbourg)[2], Mann, A. (Strasbourg)[2], Manetti, F. (Siena)[3], Solinas, A. (Siena)[3], Petricci, E. (Siena)[3], Taddei, M. (Siena)[3], Ruat, M. (Gif sur Yvette)[1]
[1]*CNRS UPR-3294, Laboratory of Neurobiology and Development, INAF, Signal Transduction and Developmental Neuropharmacology Team, Gif sur Yvette, France,* [2]*CNRS UMR 7200, Therapeutic Innovation Laboratory, Strasbourg, France,* [3]*University of Siena, Department of Pharmaceutical and Applied Chemistry, Siena, Italy*

The Hedgehog (Hh) signaling pathway regulates stem cell maintenance and repair in adult tissues. Hh proteins modulate electrical activities of mature neurons and stimulation of the Hh pathway has shown therapeutic efficacy in models of Parkinson disease, diabetic neuropathy, demyelination and myocardial ischemia suggesting that small-molecule agonists of the Hh pathway may have therapeutic

interest (Ruat et al, *Differentiation*, 2012; Ferent et al, *J. Neurosci*, 2013). Smoothened (Smo), a seven-transmembrane protein belonging to the G protein-coupled receptor superfamily, is the major transduction component of this pathway. High-throughput screening of chemical libraries has led to the identification of the Smo reference agonists SAG, a chlorobenzothiophene and purmorphamine, a purine derivative as well as related molecules. These molecules have been used for modulating embryonic stem cells and adult neural precursor cells but none has reached clinical trials (Tran et al, *Syst Biol Med*, 2013).
Here, we have generated and validated a pharmacophoric model for Smo agonists and used this model for the virtual screening of a library of commercially available compounds. Among the 20 top scoring ligands, we have identified and characterized a novel quinolinecarboxamide derivative (GSA-10) as a Smo agonist. Using pharmacological, biochemical and molecular approaches, we provide compelling evidence that GSA-10 acts at Smo to promote the differentiation of multipotent mesenchymal progenitor cells into osteoblasts. However, this molecule does not display the hallmarks of reference Smo agonists. Remarkably, GSA-10 does not recognize the classical bodipy-cyclopamine binding site, induces neither Gli-dependent reporter gene transcription nor cerebellar granule cell proliferation, and it does not regulate the subcellular localization of Smo at the primary cilium. Moreover, we observed that cholera toxin and forskolin, two known activators of adenylate cyclase, are positive and negative regulators of GSA-10 and SAG-mediated cell differentiation, respectively. Thus, GSA-10 belongs to a novel class of Smo agonists and should be helpful for dissecting Hh mechanism of action with important implications in physiology and in therapeutic.

P1.065

Induction of cell death associated with oxidative stress in human neuronal cells (SK-N-BE) treated with C22:0 or saturated very long chain fatty acids (C24:0; C26:0)

Zarrouk, A. (Dijon)[1,2], Nury, T. (Dijon)[1], Athias, A. (Dijon)[3], Hammami, M. (Monastir)[2], Lizard, G. (Dijon)[1]
[1]Faculté des Sciences Gabriel - Université de Bourgogne / EA7270, Dijon, France, [2]Université de Monastir, Faculté de Médecine, UR' Nutrition Humaine et Désordres Métaboliques', Monastir, Tunisia, [3]Université de Bourgogne / CHU de Dijon-Hôpital du Bocage, Plateau technique de lipidomique, Dijon, France

In Alzheimer´s disease (AD), some lipid alterations point towards peroxisomal dysfunctions. An accumulation of C22:0 and saturated very long chain fatty acids (VLCFAs: C24:0 and C26:0), substrates for peroxisomal β-oxidation, has been found in cortical lesions of AD patients. Human neuronal cells (SK-N-BE) were treated with C22:0, C24:0 or C26:0 (0.1 - 20 µM; 48 h). The impact of these fatty acids on cell death induction was evaluated by cell counting in the presence of trypan blue, MTT and crystal violet tests, flow cytometric analysis (quantification of dead cells and of cells in SubG1 after staining with propidium iodide (PI)), measurement of LDH activity in the culture medium, fluorescence microscopy after staining with Hoechst 33342 in order to distinguish between viable, apoptotic and necrotic cells, and transmission electron microscopy to identify the impacts of fatty acids at the ultrastructural level. Oxidative stress was estimated by flow cytometric analyses with different fluorescent probes (H_2DCFDA, DHE, DHR123, and DAF), by fluorescence microscopy in the presence of monochlorobimane for GSH quantification, and by gas chromatography coupled with mass spectrometry to evaluate lipid peroxidation. With C22:0, C24:0 and C26:0 used at 10 and 20 µM, an induction of cell death was observed. It was characterized by higher percentages of dead cells (trypan blue positive cells, PI positive cells), lower MTT and crystal violet values. No sign of apoptosis was observed with Hoechst staining (no increase of cells with condensed and/or fragmented nuclei) and by analysis of SubG1 cells (no increase of the percentage of cells in SubG1). Noteworthy, an overproduction of reactive oxygen species, including superoxide anions and H_2O_2, was also revealed, no or slight overproduction of NO was found, and a decrease of GSH associated with an increased lipid peroxidation was detected. Thus, C22:0, C24:0, and C26:0 are able to induce neuronal damages (cell death associated with oxidative stress) which could contribute to the development of AD. Consequently, increased levels of these fatty acids in cortical lesions of AD patients might favor the development of this disease.

P1.066

Contralesional suppressive rTMS enhances language recovery in non-fluent aphasic patients—a sham-controlled, double-blinded study

Tsai, P.-Y. (Taipei)[1], Wang, C.-P. (Taipei)[2]
[1]Taipei Veterans General Hospital, Dep. PMR, Taipei, Taiwan, Republic of China, [2]Taipei Veterans General Hospital, Dep Radiology, Taipei, Taiwan, Republic of China

Background: While neuromodulation through inhibitory repetitive transcranial magnetic stimulation (rTMS) administrated to the right pars triangularis (PTr) has shown promise for language recovery in a relatively small number of stroke subjects, the effectiveness of this method remains unclear. To convincingly demonstrate the reliability of this approach, additional investigations in larger cohorts of patients could be crucial.
Materials and methods: In our sham-controlled, double-blinded parallel study, thirty-one stroke patients were randomly allocated to Group A (n=20), which underwent 10-session of 1Hz rTMS over right PTr and Group B (n=11), which received sham 1Hz stimulation. We performed Picture Naming Test and Concise Chinese Aphasia Test (CCAT) at the baseline, post rTMS intervention and at 3-month follow-up.
Results: Following rTMS, Group A showed greater improvement than Group B in CCAT score ($P< 0.001$) and subcategories: conversation ($P=0.039$), expression ($P=0.012$) and repetition ($P=0.042$). Group A also manifested higher object naming accuracy ($P=0.012$) and shorter naming RT ($P=0.005$) over Group B. This effect persisted for at least 3 months ($P=0.007$) related to the baseline levels.
Conclusions: Improvements in language production and naming performance were identified with immediate and long-term effect. The results confirm the role of right PTr and the efficacy of inhibitory rTMS in relatively large cohorts of patients.

P1.067

Spadin, a sortilin-derived peptide, a new concept in the antidepressant drug design

Veyssiere, J. (Valbonne)[1], Moha ou Maati, H. (Valbonne)[2], Lucas, G. (Lyon)[3], Widman, C. (Valbonne)[2], Gandin, C. (Valbonne)[2], Heurteaux, C. (Valbonne)[2], Borsotto, M. (Valbonne)[2]
[1]Institut de Pharmacologie Moléculaire et Cellulaire, CNRS UMR 6097 - UNSA, Valbonne, France, [2]Institut de Pharmacologie Moléculaire et Cellulaire, CNRS, Valbonne, France, [3]Centre de Recherche en Neuroscience de Lyon, Lyon, France

Antidepressant treatments require several weeks of administration before observed effects but remain inadequate for many patients. Improving the treatment of depression is challenging. The TREK-1 potassium channel has been identified as a new target in depression and it has been hypothesized that TREK-1 antagonists might be effective antidepressants. We identified spadin a peptide derived from the maturation of the neurotensin receptor 3 (NTSR3 /Sortilin) that specifically blocks TREK-1 channel.
Spadin efficacy was studied through five different animal behavioral models, namely the Porsolt forced swim, the tail suspension, the conditioned suppression of motility, the learned helplessness and the novelty-suppressed feeding test. Spadin blocks the TREK-1 activity in COS-7 transfected cells and CA3 hippocampal neurons. These effects are absent in TREK-$1^{-/-}$ mice. Spadin does not affect the activity of four other K2P channels. Spadin increases the efficacy of serotoninergic neurotransmission. Similarly to that observed in TREK-$1^{-/-}$ mice, spadin induces a resistance to depression in the five behavioral models. Spadin appears to be specific for the depression because it had no effect in three anxiety animal tests: the elevated plus maze, the stair case and the white-dark box. More importantly, a spadin intravenous 4-day treatment induced a strong antidepressant effect and also enhanced

hippocampal phosphorylation of CREB protein and neurogenesis, considered to be key markers of antidepressant action after chronic treatment with selective serotonin reuptake inhibitors. Spadin does not affect other functions of TREK-1 like pain or epilepsy.
Our data together with the Alpha Screen dosing method, indicated that spadin could be used as an antidepressant molecule and as a biomarker for depression disease. Spadin can be considered as a putative endogenous antidepressant of new generation with a rapid onset of action.

P1.068

Migraine patients show attention orienting dysfunction, but preserved sensory processes

Morlet, D. (Bron)[1], Caclin, A. (Bron)[1], Fischer, C. (Bron)[1,2], Brudon, F. (Lyon)[3], Demarquay, G. (Lyon)[4]
[1]INSERM U1028, CNRS UMR5292, Université Lyon 1, Centre de Recherche en Neurosciences de Lyon, Dynamique Cérébrale et Cognition, Bron, France, [2]Hospices Civils de Lyon, Hôpital Neurologique, Neurologie fonctionnelle et épileptologie, Lyon, France, [3]Clinique du Tonkin, Lyon, France, [4]Hospices Civils de Lyon, Hôpital de la Croix-Rousse, Lyon, France

Migraine pathophysiology is still incompletely understood, in particular regarding the hypersensitivity to sensory stimulations. Changes in cortical excitability occurring between migraine attacks have been suggested. Previous electrophysiological studies have shown abnormal responses to attended warning stimuli but also to passively endured stimuli. They emphasized attention-orienting exacerbation and/or habituation deficit in migraine without clearly disentangling basic sensory processing dysfunction and abnormal attention orienting processes.
Our objectives were to investigate long-term and short-term habituation of auditory event-related potentials (ERPs) using a classic habituation paradigm and to explore automatic change detection (i.e., mismatch and attention orienting processes) during a migraine cycle.
Auditory ERPs were recorded in 22 female patients suffering from menstrually-related migraine and in 20 age-matched control subjects, in 3 sessions: in the middle of the menstrual cycle, before and during menses. In 12 patients, a migraine attack occurred during one of the peri-menses sessions. In each session, 200 trains of tone-bursts were presented in a passive listening condition, with an average of 10 stimuli per train, including two duration deviants.
In all sessions, migraine patients exhibited a drastically larger orienting component of N1 than matched controls in response to the first stimuli of the trains[1]. They also showed an increased residual orienting component in response to all stimuli inside the trains. The first stimuli elicited a larger P3a in the interictal session for migraineurs, with normalization during attacks.
In all sessions, duration deviance elicited a normal mismatch negativity (MMN) in migraine patients but a prolonged N2b. Migraineurs also presented a different modulation of P3a amplitude to deviants along the menstrual cycle, with a tendency to normalization during migraine attacks.
None of the studied ERP components showed a default of short-term or long-term habituation.
Our observations suggest normal auditory processing up to attention triggering but enhanced activation of attention-related frontal networks in migraineurs.
[1] Demarquay et al. Clin Neurophy 2011;122(9):1755-1763.

P1.069

Exposure to an alternative reward does not reduce cocaine seeking behavior

Nicolas, C. (Poitiers)[1], Lafay-Chebassier, C. (Poitiers)[1,2], Solinas, M. (Poitiers)[1]
[1]Université de Poitiers, INSERM U-1084 LNEC, Poitiers, France, [2]CHU Poitiers, Département de Pharmacologie Clinique, Poitiers, France

Environmental enrichment (EE) is comprised of several elements such as social interactions, novelty and sensory and physical activity, which have been described to be rewarding in rodents. Because both EE and access to alternative rewards decrease addiction-related behaviors, it could be hypothesized that EE produces its positive effects on addiction by acting as an alternative reward. Here we investigated whereas chronic access to a natural reward during periods of withdrawal under conditions similar to those previously used for EE, could reduce drug craving.
For this, we let rats to self-administer cocaine during 10 experimental sessions (0.15mg/injection, 6h/day) while yoked-saline rats received only saline injections. At the end of the self-administration training, rats were subjected to a 30-day withdrawal period during which they had access a) to water and sucrose (10%w/v) continuously (SC); b) to water and sucrose (10%w/v) intermittently (SI) or c) water only (WAT). At the end of the withdrawal period, rats were tested for cocaine seeking behavior during a single 6h session.
We found that, during abstinence, SC and SI rats avidly consumed sucrose, drinking three times more than WAT rats and completely ignoring water bottles. Sucrose consumption and preference were constant over the entire period of withdrawal and did not differ between cocaine and saline-yoked rats. After the 30-day withdrawal period, cocaine exposed rats showed high levels of drug-seeking behavior but no differences were found among SC, SI and WAT rats.
Altogether, these results show that exposure to a natural reward such as sucrose during withdrawal periods does not affect cocaine seeking behavior. These results suggest that exposure to an alternative reward that is temporally and physically distinct from drug-related environments is not sufficient to reduce the risks of relapse. Moreover, they suggest that elements of enrichment other than alternative reinforcement play a more important role in reducing drug seeking or, at least, that alternative reinforcement must be combined with other elements of enrichment to be effective in decreasing drug seeking.

P1.070

Comprehensive pathological analysis in MPTP-treated macaques reveals widespread synucleopathy and tauopathy

Canron, M.-H. (Bordeaux)[1], Fernagut, P.-O. (Bordeaux)[1], Li, Q. (Beijing)[2], Vital, A. (Bordeaux)[1], Bezard, E. (Bordeaux)[1]
[1]Université Bordeaux Segalen, IMN UMR CNRS 5293, Bordeaux, France, [2]Institute of Laboratory Animal Sciences, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing, China

While the MPTP macaque model of PD is considered the gold-standard for validating symptomatic treatments, its relevance to the parkinsonian neurodegeneration is questioned. A few reports have investigated the regulation and/or pattern of alpha-synuclein expression after MPTP exposure but have focused on the substantia nigra, neglected other markers of neurodegeneration and were not controlled for dopamine replacement therapy. We thus investigated the expression pattern of tau and alpha-synuclein in untreated and levodopa-treated chronic MPTP-lesioned macaques.
A comprehensive neuropathological study was performed on four groups of macaque monkeys: controls (normal), drug-naive MPTP-lesioned (parkinsonian), levodopa-treated MPTP-lesioned (dyskinetic) and aged controls. Immunohistochemistry for alpha-synuclein, tau and glial fibrillary acidic protein (GFAP) were performed on neo-cortex (frontal, temporal, occipital), hippocampus, caudate-putamen, putamen-pallidum, thalamus, substantia nigra, pons, medulla oblongata and cerebellum (dentate nucleus). Our results reveal a widespread immunoreactivity for both alpha-synuclein and tau in several brain regions in untreated MPTP-lesioned macaques. These include temporal cortex, hippocampus, putamen, substantia nigra, and dentate nucleus. GFAP staining was marked in all investigated regions except in the cerebellum. Interestingly, while the tauopathy was not corrected by the levodopa treatment, the immunostaining for alpha-synuclein was clearly reduced in the pallidum, pons, medulla oblongata and cerebellum but not in the substantia nigra of levodopa-treated MPTP-lesioned animals. This study extends the validity of the MPTP-lesioned macaque model of PD to pathological findings involving tau and alpha-synuclein accumulation during the course of the disease. In addition, it suggests that while alpha-synuclein increased expression is levodopa-insensitive in the

substantia nigra, it is sensitive to dopamine replacement therapy in other brain regions. Therefore, these data offer a window for studying dopamine/alpha-synuclein interactions and their consequences upon widespread neurodegeneration and following chronic dopamine replacement therapy.

P1.071

Deciding in a changing world: a study in Asperger syndrome

Robic, S. (Lyon)[1,2], Sonié, S. (Lyon)[1,2,3], Joffily, M. (Lyon-St-Etienne)[4], Fonlupt, P. (Lyon)[1,2], Henaff, M.-A. (Lyon)[1,2], Coricelli, G. (Paris)[5], Mattout, J. (Lyon)[1,2], Schmitz, C. (Lyon)[1,2]
[1]INSERM, U1028; CNRS, UMR5292; Lyon Neuroscience Research Center, Brain Dynamics and Cognition Team, Lyon, France, [2]University Lyon 1, Lyon, France, [3]Centre Hospitalier Le Vinatier, Bron, France, [4]Department of GATE-LSE, Lyon-St-Etienne, France, [5]Laboratoire de Neurosciences Cognitives, INSERM U960, Ecole Normale Supérieure, Paris, France

Resistance to change, a well-known symptom of autism, suggests crucial differences in predicting upcoming events in this population, especially in a social -therefore fickle- world. It is not clear whether persons with autism present with a primary difficulty to process the unpredictable nature of any kind of information, or if the difficulties they encounter in a social environment are caused by impaired social processing or/and by the unpredictability associated with the social world. We conducted an fMRI study, aiming at investigating the processing of unstable social or non-social information in persons with High Functioning Autism (HFA) and Asperger Syndrome (AS) and in paired controls.
We scanned 12 HFA/AS participants and 14 controls during a decision task performed either in a social environment or in a non-social one. In each kind of environment (social and non-social), 2 conditions were proposed: a stable (constant probabilities) and an unstable one (switching probabilities).
HFA/AS subjects showed lower performance (in terms of winning trial number) than control subjects in all conditions. Their difficulties were amplified in the unstable conditions, especially in the social environment. We found a strong correlation between autistic quotient (AQ, see Baron-Cohen et al., 2001) and performance, in all social conditions (both stable and unstable) as well as in the unstable non-social condition.
The fMRI contrast “unstable > stable” revealed an increased activity in the right frontal gyrus and in the bilateral parietal superior region in controls, and not in HFA/AS subjects. These regions have been shown to be involved in attentional processes, especially in the voluntary orienting of attention. Moreover, the percent of bold signal change in both regions was negatively correlated to AQ.
These results are in line with the difficulties observed in persons with autism to selectively direct attention to relevant information and to disengage or shift their attention. Their difficulty to process the unpredictable nature of information was amplified when the source of information was social in nature. We conclude that, both the social nature of the environment and its unpredictability cause difficulties to people with autism.

P1.072

Interplay between flexibility and emotion in ASD: a behavioural and fMRI study

Gomot, M. (Tours)[1], Cléry, H. (Tours)[1], Bonnet-Brilhault, F. (Tours)[1], Fonlupt, P. (Bron)[2]
[1]UMR 930-Inserm Université Francois Rabelais de Tours, Centre de Pédopsychiatrie - CHU Bretonneau, Tours, France, [2]INSERM U1028, Centre de Recherche en Neuroscience de Lyon, Bron, France

Patients with Autism Spectrum Disorder (ASD) display difficulties in changing strategy during daily activities or adapting their perspective, especially during social interactions. However despite number of evidence of deficits in both socio-emotional processing and cognitive flexibility in ASD, the

interactive effects of difficulties in these two domains remain unexplored. This study thus aims at investigating emotion-cognition interaction by assessing behavioural and brain correlates of cognitive flexibility when applied to emotional stimuli.
Thirteen adults with ASD and 15 controls participated in an event-related fMRI paradigm using an emotional version of the Wisconsin Card Sorting Task (WCST). The cards presented were surrounded by a coloured frame and represented emotional faces. Participants have to match cards on one of three possible dimensions according to a non spoken rule: frame colour, face identity or facial emotion.
Behavioural results revealed that compared to controls, patients succeeded in fewer categories, committed more perseverative errors when switching to Emotion matching, and displayed longer RT for Emotion and Identity conditions. fMRI results showed activity in the neural network typically recruited during WCST in both groups, including the prefrontal cortex, the inferior parietal lobule, the anterior cingulate cortex (ACC), and the inferior temporal gyrus. However switching to a new rule led to larger brain activity in ASD than controls in the ACC, striatum, cerebellum and the frontal and orbito-frontal regions, together with lower activity in the temporal poles. Results are discussed according to the sorting rule.
Findings are consistent with the difficulties in processing socio-emotional stimuli in ASD and suggest that cognitive flexibility abilities are strongly modulated by the nature of the information to be processed.

P1.073

Effect of anesthesia and surgery on tau pathogenesis

Dickler, M. (Québec)[1], Morin, F. (Québec)[1], Petry, F. (Québec)[1], Planel, E. (Québec)[1]
[1]*Centre Hospitalier de l'Université Laval, Neurosciences, RC-9800, Aile P, Québec, Canada*

Alzheimer's disease (AD) is the leading form of dementia. The neuropathological hallmarks of AD include senile plaques of β-amyloid (Aβ) peptides and neurofibrillary tangles (NFT) resulting from hyperphosphorylated tau protein. The large majority of cases (~99%) of AD is late onset and sporadic in origin. The causes of AD are considered multifactorial, with external factors interacting with biological or genetic susceptibilities to accelerate the manifestation of the disease. Anesthesia/surgery might be such factor, as there are several reports suggesting an increased incidence of AD after anesthesia/surgery.
In patients, and for obvious ethical reasons, it is not possible to dissociate anesthesia and surgery. Our previous results have already showed that different types of anesthetics have an effect on the level of tau phosphorylation either directly or indirectly by inducing hypothermia. Here we aim to isolate effects of surgery on tau pathology and try to identify whether the mechanism underlying this plausible effect involves peripheral and central inflammation.
We have collected data in young and old wild type mice as well as in young hTau mice, a mutant model of AD pathology overexpressing non-mutant human tau on a murine tau KO background. The mice were separated in two groups, one undergoing simple anesthesia and the other anesthesia and a minor abdominal surgery. While, overall our preliminary results are inconclusive on the isolated effect of surgery *per se*, we expect to see in a group of old hTau mice undergoing surgery, enhanced tau phosphorylation as well as an augmentation of brain inflammatory markers.
While there has been clinical interest on the consequences of surgery and anesthesia on cognitive decline, their biochemical consequences on AD neuropathogenic pathways have only begun to be studied very recently. Overall, more studies have to take place in order to further understand the cognitive consequences that could have a simple surgery in an elderly patient on the long term as well if there is a risk for developing AD.

P1.074

Neuronal death in cerebellar organotypic cultures from prion protein-deficient mice

Bailly, Y. (Strasbourg)[1], Guillemain, A. (Strasbourg)[1], Ragagnin, A. (Strasbourg)[1], Grant, N. (Strasbourg)[1]
[1]*CNRS, INCI CNRS UPR3212, Strasbourg, France*

Twenty years of intensive research clearly point out the pivotal cytoprotective role played by the cellular prion protein PrP^c in neurons, but still little is known about the molecular mechanisms underlying its neuroprotective properties. One approach to respond to this question is to study the susceptibility of cerebellar Purkinje cells (PC) in *Prnp*$^{0/0}$ mutant mice. In the Zurich-1 (ZH-I) *Prnp*$^{0/0}$ mutant mouse, however, the PrP^c-deficiency does not induce spontaneous PC loss, but the ectopic expression of the neurotoxic PrP^c-like protein doppel (Dpl) in the Nagasaki (Ngsk) *Prnp*$^{0/0}$ mutant mouse results in progressive PC loss. Our previous data indicates that this Dpl-mediated neurotoxicity in Ngsk PCs involves a mitochondrial apoptotic pathway, as well as a blockade of autophagic flux. Here, we have analyzed the development and survival of PC neurons in cerebellar organotypic cultures from Ngsk and ZH-I *Prnp*$^{0/0}$ mice. Morphometric and quantitative analyses of PC showed that the growth deficits and death of PC occur with similar kinetics and amplitude in both mutants, suggesting that the absence of PrP^c is responsible for their neurodegeneration. Furthermore, increases in autophagic and apoptotic markers were detected in extracts by Western blotting, suggesting that both autophagy and apoptosis are activated in the *Prnp*$^{0/0}$ cultures.
These results imply that PrP^C has a neuroprotective role in cerebellar Purkinje cells. In addition, the cerebellar organotypic cultures should provide a suitable system for analyzing the mechanisms underlying the neurotoxic effects of PrP^C-deficiency.

P1.075

Parasagittal compartmentation of scrapie prion protein deposition and inflammation in the mouse cerebellar cortex

Ragagnin, A. (Strasbourg)[1], Pietri, M. (Paris)[2], Guillemain, A. (Strasbourg)[1], Alleaume-Butaux, A. (Paris)[2], Haeberlé, A.-M. (Strasbourg)[1], Grant, N. (Strasbourg)[1], Vidal, C. (Paris)[3], Kellermann, O. (Paris)[2], Schneider, B. (Paris)[2], Bailly, Y. (Strasbourg)[1]
[1]*CNRS, INCI CNRS UPR3212, Strasbourg, France,* [2]*Université Paris Descartes INSERM UMR S747, Paris, France,* [3]*Institut Pasteur, Biologie Cellulaire et Infection, Paris, France*

Prion diseases are neurodegenerative disorders characterized by the transition of the cellular prion protein (PrP^c) into a pathogenic conformer, PrP^{Sc}, which accumulates in the diseased brain. With spongiosis and loss of synapses and neurons, inflammation of the CNS occurs in prion diseases as revealed by reactive gliosis. Pro-inflammatory cytokines like TNF-α released by glial cells are believed to contribute to neurodegeneration. However, it remains unknown whether PrP^{Sc} accumulation in neurons would exacerbate the toxicity of inflammatory cytokines and whether the altered neuronal response to cytokines would be restricted to specific neuronal populations infected by PrP^{Sc}.
Our current work substantiates that PrP^C conversion into accumulating PrP^{Sc} in cultured neurons trigger the up-regulation of TNF-α receptor type 1 (TNFR1) at the plasma membrane, which hypersensitizes infected neurons to TNF-α toxicity. TNFR1 over-expression also occurs in the brains of mice infected by the 22L strain of the scrapie PrP^{Sc} at the clinical stage of the disease. Intriguingly, in the cerebellar cortex of 22L-infected mice, TNFR1 displays a parasagittal pattern of alternating bands of weak and strong immunoreactivity (IR) similar to that of PrP^{Sc22L} deposition. However, TNFR1 IR increases in bands where there is less accumulation of PrP^{Sc22L}, which apparently contrasts with our *in vitro* observations. Bands with the strongest TNFR1 IR are enriched with surviving Purkinje cells (PCs), while bands with the most intense PrP^{Sc22L} IR are deprived of PCs. This suggests that PrP^{Sc22L} precipitates where neuronal loss occurs and that PCs are likely TNFR1-expressing neurons. In addition, intense astrogliosis is shown in bands with strong PrP^{Sc22L} IR, indicating that astrocytes proliferate in response to neuronal loss and that they are not sites of TNFR1 expression. Finally, in the cerebellar cortex of 22L scrapie-infected mice expressing eGFP under the control of the promoter of the PC-specific glutamate transporter gene *eaat4*, the banding patterns of PrP^{Sc22L} IR and eGFP are

not topographically correlated. Further investigations are required to understand this differential sensitivity of PCs to prion toxicity across the parasagittal compartments of the cerebellar cortex.

P1.076

L-Dopa acquires psychostimulant-like properties after nigral dopaminergic loss

Engeln, M. (Bordeaux)[1], Fasano, S. (Milano)[2], Ahmed, S.H. (Bordeaux)[1], Cador, M. (Bordeaux)[1], Baekelandt, V. (Leuven)[3], Bezard, E. (Bordeaux)[1], Fernagut, P.-O. (Bordeaux)[1]
[1]Université Bordeaux Segalen, Bordeaux, France, [2]Institute of Experimental Neurology, Division of Neuroscience, San Raffaele Research Institute, Milano, Italy, [3]Laboratory for Neurobiology and Gene Therapy, Katolik University, Leuven, Belgium

Symptomatic treatment of Parkinson's disease (PD) with dopamine replacement therapy (DRT) is associated with important limits. In addition to its inability for alleviating non-motor symptoms, chronic DRT leads to a number of motor and non-motor side-effects. The latter include the dopamine dysregulation syndrome (DDS) occurring in 3-4% of PD patients taking levodopa and/or apomorphine. DDS is characterized by compulsive DRT-seeking and hoarding, self-medication, and withdrawal syndrome. Growing evidences for an activation of the reward pathways by different types of DRT have been provided and compulsive DRT use shares features with psychostimulant addiction.
This study therefore investigated whether L-Dopa possesses psychostimulant-like properties by evaluating its rewarding properties using conditioned place preference (CPP) and its ability to affect a nondrug reward in a sweet-taste preference paradigm. The influence of an altered dopaminergic function causing motor deficits (-24% of adjusted steps) on the emergence of compulsive DRT use was assessed using partial (-30.7%) bilateral lesions of the Substantia Nigra *pars compacta* (SNc) induced by viral-mediated overexpression of alpha-synuclein.
Our results demonstrate rewarding properties of L-Dopa at therapeutically relevant doses. Interestingly, such rewarding effect was observed only in lesioned animals, indicating that midbrain neurodegeneration is critical for the expression of this behavior. While lesion alone had no effect on the animal's discrimination capacities or appetite for sweet taste, L-Dopa decreased sweetened-water consumption in lesioned rats suggesting that, like psychostimulants, L-Dopa affects the palatability of a nondrug reward. Altogether these observations suggest that, when combined to a SNc lesion, L-Dopa acquires psychostimulant properties that might contribute to the subsequent development of DDS described in some PD patients.

P1.077

Protective effects of caffeine in a transgenic model of Alzheimer´s disease-like Tau pathology

Blum, D. (Lille)[1], Laurent, C. (Lille)[1], Derisbourg, M. (Lille)[1], Eddarkaoui, S. (Lille)[1], Demeyer, D. (Lille)[1], Schneider, M. (Bonn)[2], Müller, C. (Bonn)[2], Hamdane, M. (Lille)[1], Buée, L. (Lille)[1]
[1]Inserm U837, 'Alzheimer & Tauopathies', Lille, France, [2]Pharmaceutical Institute, University of Bonn, Bonn, Germany

Alzheimer's disease (AD) is characterized by extracellular amyloid deposits and intraneuronal neurofibrillary tangles, made of aggregated hyper- and abnormally phosphorylated Tau proteins. The latter, referred to as “Tau pathology”, contribute to synaptic impairments leading to memory deficits in AD. Previous epidemiological studies revealed that habitual chronic caffeine consumption reduces the risk to develop AD. Further, recent experimental studies suggested that chronic caffeine treatment mitigates spatial memory alterations and central / plasma Ab λοαδ in APP transgenic mouse models. However, whether caffeine exerts effects towards Tau pathology remains unknown so far.
In the present study, we investigated impact of chronic caffeine consumption in a transgenic model of AD-like Tau pathology, the THY-Tau22 strain. THY-Tau22 mice have been shown to exhibit

progressive hippocampal Tau pathology paralleling memory deficits and neuro-inflammatory processes. Caffeine (0.3g/L) was chronically administrated through drinking water to Tau mice and littermate controls from 3 to 12 months of age. At completion of the treatment, caffeine and its metabolites were readily detected in the brain and plasma of treated animals. Using Morris Water Maze, we observed that caffeine treatment improved spatial memory in THY-Tau22 mice. That was associated with reduced Tau phosphorylation and Tau proteolysis. Further, we observed that caffeine treatment significantly mitigated hippocampal neuro-inflammation as shown by the reduction of several pro-inflammatory markers previously found overexpressed in THY-Tau22 hippocampus. Finally, other marker variations suggested that beneficial effect exerted by caffeine in THY-Tau22 mice is also related to its anti-oxidant properties. Altogether, the present data are the first reporting that caffeine exerts beneficial effect in a transgenic mouse model of AD-like Tau pathology. Caffeine may thus likely be a potential treatment for AD.
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P1.078

Target-specific vulnerability of excitatory synapses leads to deficits in associative memory in a model of intellectual disorder

Houbaert, X. (Bordeaux)[1], Zhang, C.-L. (Bordeaux)[1], Gambino, F. (Genève)[2], Lepleux, M. (Bordeaux)[1], Herzog, E. (Bordeaux)[1], Humeau, Y. (Bordeaux)[1]
[1]Université Bordeaux 2, IINS, Interdisciplinary Institute for Neuroscience, Equipe Yann Humeau, Bordeaux, France, [2]Centre Médical Universitaire, Département des Neurosciences Fondamentales, Genève, Switzerland

Intellectual disorders (ID) have been regularly associated with morphological and functional deficits at glutamatergic synapses in both humans and rodents. How these synaptic defects may lead to the variety of learning and memory deficits defining ID is still unknown. Here we studied the functional and behavioural consequences of *Il1rapl1* deficiency in mice, and reported that *Il1rapl1* constitutive deletion independently alters cued fear memory in mice. Combined *in vivo* and *in vitro* approaches allowed us to unveil a causal relationship between a strong Inhibitory/Excitatory (I/E) imbalance in dedicated amygdala neuronal sub-circuits and behavioural deficits. Cell-targeted recordings further demonstrated a strong impact of the mutation at excitatory synapses contacting principal cells while the same afferents contact interneurons normally. We thus propose that excitatory synapses have a heterogeneous vulnerability to *Il1rapl1* gene constitutive mutation, and that target-specific functional alteration of excitatory synapses in neuronal circuits is sufficient to generate permanent cognitive deficits.

P1.079

Beneficial effects of adenosine A_{2A} receptor inactivation in a transgenic model of Alzheimer´s disease-like Tau pathology

Laurent, C. (Lille)[1], Burnouf, S. (Lille)[1], Ferry, B. (Lyon)[2], Marciniak, E. (Lille)[1], Eddarkaoui, S. (Lille)[1], Parrot, S. (Lyon)[2], Demeyer, D. (Lille)[1], Ledent, C. (Brussels)[3], Muller, C.E. (Bonn)[4], Hamdane, M. (Lille)[1], Sergeant, N. (Lille)[1], Humez, S. (Lille)[1], Lopes, L. (Lisbonne)[5], Buee, L. (Lille)[1], Blum, D. (Lille)[1]
[1]Inserm U837, 'Alzheimer & Tauopathies', Lille, France, [2]Inserm 1028, Lyon, France, [3]ULB, Brussels, Belgium, [4]Pharmaceutical Institute, Bonn, Germany, [5]Instituto de Medicina Molecular, Lisbonne, Portugal

Alzheimer's disease (AD) is characterized by extracellular amyloid deposits and intraneuronal neurofibrillary tangles, made of aggregated hyper- and abnormally phosphorylated Tau proteins. The latter, referred to as “Tau pathology”, contributes to synaptic impairments leading to memory deficits in AD patients. Previous epidemiological studies revealed that habitual chronic caffeine consumption reduces the risk to develop AD. In line, we recently demonstrated that caffeine exhibits beneficial properties in the THY-Tau22 model of AD-like Tau pathology (see poster of D. Blum this meeting). One of the molecular targets of caffeine is represented by adenosine A_{2A} receptors on which it is acting as a non-selective antagonist. Although impact of A_{2A} receptor blockade in AD is largely unknown, we assume that the neuroprotective properties of caffeine are mediated by its action on A_{2A} receptors. The present study is aimed at evaluating the effects of A_{2A} receptors deletion on the physiopathological development of AD symptoms in a transgenic model of AD-like Tau pathology (THY-Tau22) exhibiting progressive hippocampal Tau pathology coinciding with memory deficits and neuro-inflammatory processes.
To this aim, A_{2A} receptors were genetically deleted in THY-Tau22 mice by two successive crossings with $A_{2A}^{+/-}$ mice. At the age of 6-7 months, spatial memory was tested using Morris Water Maze and Y-Maze tasks. Results showed that loss of A_{2A} receptors prevents spatial memory alterations found in THY-Tau22 mice. These effects were associated with a normalization of the Glutamate/GABA ratio obtained with *in vivo* hippocampal microdyalisis. Further, we observed that A_{2A} receptor deletion led to reduced hippocampal Tau phosphorylation. Finally, we found that THY-Tau22 $A_{2A}^{-/-}$ mice exhibited reduced hippocampal neuro-inflammation as shown by the reduction of several pro-inflammatory markers previously overexpressed in THY-Tau22 hippocampus. Impact of A_{2A} receptor deletion upon synaptic plasticity is currently under investigation. Altogether, the present data are the first reporting that A_{2A} blockade exerts beneficial effect in a transgenic mouse model of Alzheimer's disease and support the therapeutic potential of A_{2A} receptor targeting in this disorder.

P1.080

The human TREK-1-HEK cell line, a powerful tool for antidepressant screening

Moha, H. (Valbonne)[1], Veyssiere, J. (Valbonne)[1], Devader, C. (Valbonne)[1], Widmann, C. (Valbonne)[1], Gandin, C. (Valbonne)[1], Mazella, J. (Valbonne)[1], Heurteaux, C. (Valbonne)[1], Borsotto, M. (Valbonne)[1]
[1]*Institut de Pharmacologie Moléculaire et Cellulaire, CNRS UMR7275, Valbonne, France*

Introduction The two pore-domain potassium channel TREK-1 has been identified as a new target in depression. The hypothesis that TREK-1 blockers might be effective antidepressants was demonstrated with spadin.
Methods: cDNA encoding the human-TREK-1 gene (kcnk2) was stably transfected in HEK293 cells. Positive clone were isolated using the Dulbecco's modified Eagle's medium supplemented with 10% heat inactivated fetal bovine serum containing 0.5 mg/mL of Geneticin (G418) in an atmosphere of 95% air/5% CO2. This G418 containing medium was used for all subsequent cultures. Functional expression was validated by performing electrophysiological measurements. Iodinated-spadin binding activity was also determined and the protective effect of TREK-1 expression against ischemia was measured.
Results: The human TREK-1 stably transfected into HEK293 cells conserved all its regulation properties. Currents generated by h-TREK-1/HEK cells are blocked by spadin or fluoxetine, activated by polyunsaturated fatty acids such as arachidonic acid, alpha linoleic acid, docosahexaenoic acid, or by riluzole. These currents have also conserved their ability to be increased by the membrane stretch or internal acidification. Using equilibrium binding experiments we demonstrated that spadin displayed a high affinity for the TREK-1 channels. By using the oxygen-glucose deprivation protocol we confirmed the protective effect of TREK-1 channels against hypoxia.
Discussion: Our data clearly indicated that the h-TREK-1/HEK clone displayed all the characteristics of the native h-TREK-1 channels. Consequently, this clone can be used to screen antidepressant molecules. Additionally, the h-TREK-1/HEK clone can also be used to screen molecule against other diseases where TREK-1 channels are involved such as pain, epilepsy or ischemia.

P1.081

Parallel increases in [^{3}H]Muscimol and [^{3}H]Flumazenil binding in the dorsolateral prefrontal cortex in schizophrenia are linked to $GABA_A$ receptor α4 and γ2S mRNA subunit levels respectively

Verdurand, M. (Bron)[1,2,3], Fillman, S. (Sydney)[2,4,5], Shannon Weickert, C. (Sydney)[2,4,5], Zavitsanou, K. (Sydney)[2,4,5]

[1]Université Claude Bernard Lyon 1, Centre de Recherche en Neurosciences de Lyon, Equipe BIORAN - GHE - Bât CERMEP, Bron, France, [2]Schizophrenia Research Institute, Sydney, Australia, [3]ANSTO LifeSciences, Australian Nuclear Science and Technology Organisation, Sydney, Australia, [4]Schizophrenia Research Lab, Sydney, Australia, [5]School of Psychiatry, University New South Wales, Sydney, Australia

Background: The gamma-aminobutyric acid type A receptor ($GABA_AR$) is an ion channel that mediates neuronal inhibition in the brain and is comprised of 2α, 2β and 1γ/δ or ε subunits. Each $GABA_AR$ contains two GABA binding sites (α-β interface) and one benzodiazepine (BDZ) binding site (α-γ interface). Evidence suggests that deficits in GABAergic neurotransmission in the dorsolateral prefrontal cortex (DLPFC) may contribute to the cognitive impairments in patients with schizophrenia (SCZ). The present study used a large cohort of 37 SCZ cases and their 37 matched controls (CON) to test whether changes in the binding sites of $GABA_AR$ exist in the DLPFC in SCZ and how they are related to variations in specific $GABA_AR$ receptor subunit mRNAs.
Methods: The GABA and BDZ binding sites were assessed by autoradiography with the agonist [^{3}H]Muscimol and the antagonist [^{3}H]Flumazenil respectively, on both the superficial (I, II, and III) and deep layers (IV, V, VI) of the DLPFC. Quantitative RT-PCR was used to measure mRNA subunits expression level (β1, β2, β3, γ1, γ2, γ2S for short and γ2L for long isoform, γ3 and δ) in the DLPFC. Measurements of mRNA expression and binding were analysed by correlation and regression analysis.
Results: Significant increases in both [^{3}H]Muscimol ($F(1,142)=5.98$, $p=0.016$) and [^{3}H]Flumazenil ($F(1,140)=6.5$, $p=0.012$) binding were observed in SCZ compared to CON in both the superficial and deep layers of the DLPFC. Within the SCZ group, both binding measures were not affected by the total, mean or final recorded APD dose or BDZ treatment before death. The expression level of mRNA subunits didn't show any significant variations in SCZ compared to CON. Interestingly, regression analysis revealed that in SCZ, [^{3}H]Muscimol binding variance was mainly explained by α4 and [^{3}H]Flumazenil binding variance was most related to γ2S subunit mRNA levels.
Conclusion: Our results showed an up-regulation of both GABA and BDZ binding to the $GABA_AR$ in the DLPFC in SCZ, independent from previous APD/BDZ treatment. Most importantly, our data suggests that these increases in binding may be linked to a “shift” in α4 and γ2S subunit mRNA levels in the disease state and may have implications for the therapeutic strategies that target specific receptor subunits.

P1.082

Hippocampal proliferative effect of protein kinase C inhibitors is not required for their antimanic-like activity in the sleep deprivation model

Abrial, E. (Lyon)[1], Scarna, H. (Lyon)[1,2], Lucas, G. (Lyon)[1,2], Haddjeri, N. (Lyon)[1,2], Lambás-Señas, L. (Lyon)[1,2]

[1]Institut Cellule Souche et Cerveau, Neuropsychopharmacologie des troubles affectifs, Lyon, France, [2]INSERM, Lyon, France

Bipolar disorder is a devastating long-term disease characterized by alternate episodes of depression and mania. In recent years, protein kinase C (PKC) has emerged as a potential molecular target for the treatment of bipolar disorder. Accordingly, several clinical trials have shown a rapid improvement

of manic symptoms in bipolar patients treated with tamoxifen, an antiestrogenic drug which also exhibits PKC inhibitory properties.
To better understand the molecular and cellular mechanisms underlying the putative antimanic effects of PKC inhibition, we aimed to examine the effects of two PKC inhibitors on the behavioral alterations displayed by sleep deprived (SD) rats, an animal model of mania. Moreover, within the framework of novel theories suggesting that cell proliferation is required for the therapeutic effects of mood disorders treatments, we investigated whether cell proliferation is impaired in the SD model. Further, we also assessed the involvement of this factor in the behavioral response of PKC inhibitors.
Our results showed that rats subjected to 72h of REM-SD exhibited lithium-responsive hyperlocomotion and hippocampal cell proliferation deficits. An acute injection of either tamoxifen (80 mg/kg, i.p.) or chelerythrine (3 mg/kg, s.c.), a selective PKC inhibitor, attenuated the SD-induced increase of locomotor activity. In addition, both PKC inhibitors acutely rescued the cell proliferation impairments in the dentate gyrus of SD animals. Importantly, treatment with the antimitotic agent thioTEPA, which abolished cell proliferation in the hippocampus, did not affect the antimanic-like effect of tamoxifen and chelerythrine that we evidenced in the SD model.
Together, these results emphasize the therapeutic potential of PKC inhibitors, by revealing their antimanic-like and proliferative properties in an animal model of mania. Moreover, they suggest that the antimanic-like activity of PKC inhibitors does not primarily involve cell proliferation. Overall, these findings may provide new insights on the pathophysiology of bipolar disorder and on the mechanisms underlying antimanic response.

P1.083

The impact of monoamine depletions on the therapeutical efficacy of L-dopa and subthalamic high frequency stimulation in the context of Parkinson's disease

Faggiani, E. (Bordeaux)[1], Delaville, C. (Bordeaux)[1], Benazzouz, A. (Bordeaux)[1]
[1]*Université Bordeaux Segalen IMN, CNRS UMR 5293, Bordeaux, France*

Introduction: Parkinson's disease (PD) is a neurodegenerative disorder characterized by the loss of dopamine (DA) neurons in the pars compacta of substantia nigra. PD is also characterized by a loss of noradrenaline (NA) cells in the locus coeruleus and serotonin (5-HT) cells in the dorsal raphe. Besides motor symptoms, non-motor symptoms, such as depression and anxiety, are also seen in PD patients. Motor symptoms are generally treated by levodopa or in advanced stages with high frequency stimulation (HFS) of the subthalamic nucleus (STN) alone or combined with levodopa. However, the origin of the loss of levodopa efficacy in severe PD patients is not clearly determined. The present study aimed to characterize the consequences of dopamine, noradrenaline and serotonin alone or combined on the efficacy of antiparkinsonian treatments (levodopa and/or STN HFS) on the motor and non-motor deficits.
Approach: This study was carried out on rodents: a sham group and four groups with different monoamine depletions.
DA depletion was performed by stereotaxic bilateral injection of 6-hydroxydopamine (6-OHDA) into the medial forebrain bundle. An intra-peritoneal (i.p.) injection of N-(2-chloroethyl)-N-ethyl-2-bromobenzylamine (DSP-4) was given to induce a noradrenergic deficiency and parachlorophenylalanine (PCPA) for the depletion of serotonin.
Two stimulating electrodes were implanted bilaterally into the STN and levodopa was given at a dose of 12 mg/kg.
Motor behaviour was assessed in an open-field, anxiety in the elevated plus-maze and “depressive-like” behaviour was studied using the forced swim test.
Results: Our results show that DA and/or NA depletion induced motor deficits. STN HFS can reverse the motor deficit induced selectively by DA depletion, but was without any effect when NA and/or 5-HT were depleted. Anxiety behaviour, which is DA dependent, was improved by levodopa.
Depressive-like behaviour was potentiated with the depletion of the three monoamines and can be reversed by the two antiparkinsonian treatments.
Conclusion: The present study provides evidence on the key role played by the three monoamines depletions in the pathophysiology and therapy of Parkinson's disease.

P1.084

Role of cation chloride cotransporter KCC3 on Cl^- handling of spinal motoneurons. Relevance to the Andermann syndrome

Tremblier, B. (Montpellier)[1], Salsac, C. (Montpellier)[1], Raoul, C. (Montpellier)[1], Scamps, F. (Montpellier)[1]
[1]*Institut de Neurosciences de Montpellier, Montpellier, France*

The cation-Cl^- cotransporters of the *Slc12a* gene family participate to volume regulation. Among the five members, the Cl^- accumulating NKCC1 and Cl^- extruding KCC2 and KCC3 control neuronal Cl^- balance. In humans, KCC3 mutations lead to agenesis of the corpus callosum with peripheral motor and sensory neuropathy (ACCPN or Anderman syndrome). The peripheral neuropathy of the disease is replicated in mice with a targeted disruption of KCC3 gene. KCC3 loss of function leads to axon swelling followed by progressive neurodegeneration of peripheral fibres in adult mice. It is assumed that axon loss and myelin degeneration likely underlie motor and posture abnormalities. However, defects in motoneuron Cl^- handling have never been addressed.
We first investigated transcripts expression and contribution of cation-Cl^- cotransporters to the Cl^- handling of motoneurons purified from wild-type and $KCC3^{-/-}$ E12.5 embryos and maintained in culture from 1 to 7 days. Quantitative RT-PCR performed on immunopurified motoneurons showed that NKCC1 was predominantly expressed at 1 DIV. After 7DIV, KCC2 transcript expression increased, becoming predominant over NKCC1. The expression of KCC3 remains constant during this period. Analysis of transcript expression in $KCC3^{-/-}$ motoneurons did not reveal major developmental modifications. Gramicidin-perforated patch-clamp recordings of $GABA_A$ chloride current reversal potential, E_{GABA-A} in wild-type neurons revealed that Cl^- accumulated in embryonic motoneurons at 1 DIV, and was significantly decreased at 7 DIV which is consistent with the increased KCC2 transcript expression. Recordings of E_{GABA-A} in $KCC3^{-/-}$ motoneurons show no significant differences with wild-type motoneurons. A similar pattern of cation-Cl^- cotransporter transcripts expression was obtained from $KCC3^{-/-}$ motoneurons compared to wild-type. In conclusion, our data show that the chloride shift, observed during maturation of the central nervous system, is reproducible *in vitro*. Therefore, our *in vitro* model provides a solid means to analyze the role and regulation of cation-Cl^- cotransporters in motoneuron during development and in adulthood. Our experimental evidence suggests that Anderman syndrome does not involve defects in spinal motoneuron Cl^- handling.

P1.085

1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine does not impact upon central nervous system availability of drugs

Thiollier, T. (Marcy l'Etoile)[1,2,3], Wu, C. (Beijing)[4], Contamin, H. (Marcy l'Etoile)[1], Li, Q. (Manchester)[5,6], Zhang, J. (Beijing)[4], Bezard, E. (Bordeaux)[2,3]
[1]*Cynbiose, Marcy l'Etoile, France,* [2]*CNRS, Institut des Maladies Neurodégénératives, UMR 5293, Bordeaux, France,* [3]*Univ. de Bordeaux, Institut des Maladies Neurodégénératives, UMR 5293, Bordeaux, France,* [4]*Institute of Materia Medica, Chinese Academy of Medical Sciences, Beijing, China,* [5]*Motac Neuroscience Ltd, Manchester, United Kingdom,* [6]*Institute of Laboratory Animal Sciences, China Academy of Medical Sciences, Beijing, China*

Brain bioavailability of drugs developed for managing central nervous system diseases is classically documented based upon cerebrospinal fluid (CSF) sampling collected in normal animals. That disease

states might affect brain availability of drugs is almost never considered at this stage although several conditions are associated with blood brain barrier (BBB) damage.
Building upon our expertise in Parkinson's disease translational research, the present study addressed this gap comparing plasma and CSF bioavailability of L-DOPA, carbamazepine, quinidine, lovastatin and simvastatin, in normal and MPTP-treated macaque monkeys, the gold standard model of Parkinson's disease. The drugs were selected based upon their differential transport across the BBB. Surprisingly, no major difference was observed between the two experimental conditions, suggesting that brain availability of drugs, at least in this model, is preserved and that therefore pharmacokinetic/pharmacodynamic experiments of Parkinson's disease drugs might be performed in normal animals.

P1.086

Impact of iron deprivation on the dopaminergic system in *Cynomolgus* monkey: toward a Willis-Ekbom disease model?

Hyacinthe, C. (Bordeaux)[1], De Deurwaerdere, P. (Bordeaux)[1], Li, Q. (Bejing)[2], Bezard, E. (Bordeaux)[1], Ghorayeb, I. (Bordeaux)[1]
[1]Université Bordeaux Ségalen, Bordeaux, France, [2]Institute of Lab Animal Sciences, China Academy of Medical Sciences, Bejing, China

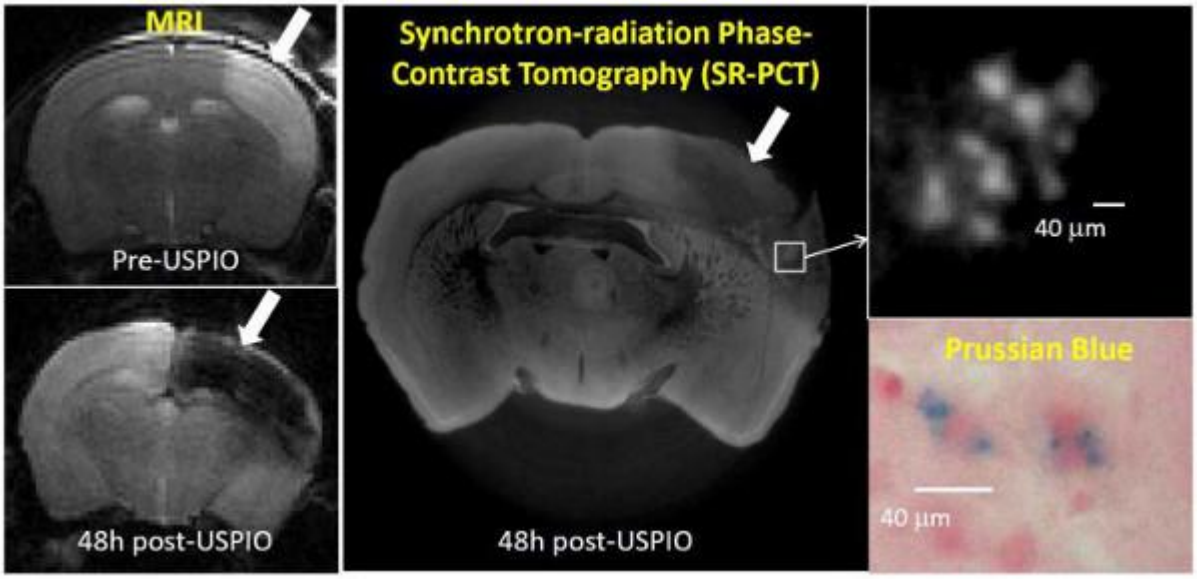
Dysregulation of iron metabolism has largely been involved in neurodegenerative diseases. Iron overload in the central nervous system has been linked to Parkinson's disease (PD). By contrast, iron deficiency has been involved in Willis-Ekbom disease (WED), a condition mainly characterized by an urge to move the legs, associated with increasing desagreable sensations in the evening and/or at night. In WED, iron and ferritin deficiencies may be found in both serum and cerebrospinal fluid (CSF). Both, PD and WED are treated with dopaminergic treatments suggesting an interaction between iron and the dopaminergic system. Here, we investigated the impact of light, mild or severe iron depletion on the dopaminergic system and on general locomotor activity in three groups of *Cynomolgus* monkeys that respectively underwent four, six and eight blood withdrawal (BW) sessions.
Iron, ferritin and transferrin concentrations were measured in both serum and CSF, before and during BW sessions in order to distinguish three iron depleted (ID) groups. Behavioural effects of the iron depletion were characterized by actimetry recordings before and after the induction of iron depletion. An increase in the locomotive behaviour was reported in the severe and mild ID groups but not in the light ID group, suggesting that under a certain threshold, iron level in serum impacts on locomotive behaviour. HPLC on *in-vivo* microdialysis revealed that dopamine and 3,4-Dihydroxyphenylacetic acid (DOPAC) were increased in striatum of the severe ID group compared to the control group. However HPLC dosages did not reveal any significant changes in serotonin and its metabolite levels. Post-mortem striatum, prefrontal cortex and spinal cord are currently being analyzed using HPLC and may highlight further changes in monoamine and their metabolite levels.
To our knowledge, this is the first study to establish a relationship between iron depletion, locomotive behaviour and dopaminergic modifications in *Cynomolgus* monkey. Providing further brain and spinal cord histological analyses for iron and dopamine markers in iron-depleted monkeys will help to unravel the pathophysiology of WED. This may pave the way toward a potential non-human primate model of the disease.

P1.087

Synchrotron radiation micro-computed tomography as a new method to detect iron oxide nanoparticles in the brain

Chauveau, F. (Lyon)[1,2], Marinescu, M. (Lyon)[2], Langer, M. (Lyon)[2,3], Durand, A. (Lyon)[2], Olivier, C. (Lyon)[2,3], Chabrol, A. (Lyon)[2], Rositi, H. (Lyon)[2], Cho, T.-H. (Lyon)[2], Nighoghossian, N. (Lyon)[2], Berthezène, Y. (Lyon)[2], Peyrin, F. (Lyon)[2,3], Wiart, M. (Lyon)[2]
[1]Université de Lyon, Centre de Recherche en Neurosciences de Lyon (CRNL); CNRS, UMR 5292; INSERM, U1028; Université Lyon 1, Lyon, France, [2]Université de Lyon, Centre de Recherche en Acquisition et Traitement de l'Image pour la Santé (CREATIS); CNRS, UMR 5220; INSERM, U1044; INSA-Lyon; Hospices Civils de Lyon; Université Lyon 1, Lyon, France, [3]European Synchrotron Radiation Facility (ESRF), Grenoble, France

Purpose: Ultrasmall superparamagnetic particles of iron oxide (USPIOs) are currently under assessment as magnetic resonance imaging (MRI) markers for the study of inflammatory disorders associated with elevated phagocytic activity, such as cerebral ischemia. The limitation of this approach presently lies in the difficulty of interpreting MR signal changes in terms of exact USPIO localization. The present paper introduces Synchrotron Radiation X-ray Phase Computed Tomography (SR-PCT) as a new method for brain mapping of USPIO in a mouse model of cerebral ischemia.
Materials and methods: The sensitivity of the technique was assessed by performing SR-PCT and MRI back-to-back in 10 mice stereotaxically injected with a range of USPIO concentrations. Eight mice with cerebral ischemia were then intravenously injected with USPIOs and imaged back-to-back with MRI and SR-PCT (Fig. 1).
Results: SR-PCT proved sensitive enough to detect iron in nanomolar quantities. In stroke-induced animals, SR-PCT showed hyperintense areas in the regions of MR signal loss. Some of these hyperintense areas consisted of bright spots about 40-µm in diameter, compatible with USPIO-labeled macrophages. Immunohistochemistry demonstrated the presence of iron-labeled cells, most probably macrophages, at the same location. The SR-PCT pipeline, moreover, identified brain anatomy as clearly as histology, without the need for sectioning or staining, with an examination time of 44 minutes per brain at an isotropic spatial resolution of 8 µm.
Conclusion: Phase-contrast X-ray computed tomography may become an invaluable complement to USPIO-enhanced MRI to study post-ischemic inflammation in intact brain, with unequaled neuroanatomic analysis.



[Figure 1.]

Figure 1. Mouse brain with an ischemic lesion (arrow), imaged with MRI and SR-PCT.

P1.088

Antiangiogenic therapy with bevacizumab in gliomas: an *in vitro* study in a three-dimensional matrix

Simon, T. (Rouen)[1], Coquerel, B. (Rouen)[1], Le Cerf, D. (Mont Saint Aignan)[2], Vannier, J.-P. (Rouen)[1]
[1]*Université de Rouen, Laboratoire Micro-Environnement et Renouvellement Cellulaire Intégré, Rouen, France,* [2]*Université de Rouen, Laboratoire Polymères Biopolymères Surfaces UMR 6270, Mont Saint Aignan, France*

High grade gliomas are one of the most aggressive solid tumors. The brain Extra-Cellular Matrix and the neoangiogenesis phenomenon are essentials to the glioma tumorigenesis. VEGF-A is a key pro-angiogenic cytokine during neoanigoenesis. In fact, our preliminary studies have shown that VEGF-A, secreted by malignant cells, is also involved in the glioma aggressiveness in an autocrine way. Several anti-angiogenic treatments have been developed, such as bevacizumab (or Avastin, Roche

®), a monoclonal antibody against VEGF-A. This therapy has a significant anti-tumoral activity in some cancers. However, despite some effects observed in controlled clinical studies, a resistance is usually reported in gliomas. This resistance could be due to a direct effect of bevacizumab on glioma cells. The aim of our project is to study the glioma cells behavior in response to a VEGF-A inhibition by bevacizumab, in an avascular tridimensional environment.
We compared the behavior of three human glioma cell lines in a tridimensional cell culture model, a Hyaluronic Acid hydrogel mimicking some aspects of the brain ECM, versus the usual bidimensional cell culture. Thereby, glioma cell aggressiveness was evaluated by measuring the invasion status in the hydrogel and the proliferation and migration status in the bidimensional model. An increase in the number of the colonies inside the HA hydrogels, with bevacizumab, has been noted; furthermore, the same treatment has a pro-migratory and proliferative effects on glioma cells in bidimensional cell culture. Changes in the genetic and protein expression profiles have been studied. Using PCR, we evidenced an alteration in the expression of the VEGF-A receptors mRNA, in response to bevacizumab treatment. Data from cytokine arrays showed that adding bevacizumab directly on glioma cells can modify the proteomic synthesis of cytokines involved in neo-angiogenesis. Therefore, we will study the intracellular mechanisms involved in the direct effect of bevacizumab on glioma cells. Moreover, to counteract this glioma resistance to bevacizumab, some combinations with current chemotherapies (as irinotecan) will be tested. This will allow us to imagine a new therapeutic approach to enhance bevacizumab efficiency in the treatment of gliomas.

P1.089

Pattern of cocaine use influences drug seeking and neuronal activity in a relevant network

Fiancette, J.-F. (Bordeaux)[1], Martín-García, E. (Bordeaux)[1], Renault, P. (Bordeaux)[1], Courtin, J. (Bordeaux)[1], Levet, F. (Bordeaux)[1], Herry, C. (Bordeaux)[1], Deroche-Gamonet, V. (Bordeaux)[1]
[1]*INSERM, Bordeaux, France*

Clinical and preclinical data suggest that vulnerability to develop a fast pattern of cocaine use might contribute to pathological cocaine seeking and transition to addiction. The causal relationship between fast pattern of use and increased cocaine seeking remained hypothetical, however. We therefore investigated whether manipulation of the pattern of use could influence cocaine seeking and neuronal activity in a key brain network.
We first established conditions allowing controlling the pattern of cocaine self-administration in the rat. We then evaluated the impact of fast and slow imposed patterns of use on cocaine seeking, as measured by cocaine-induced reinstatement. Finally, using c-Fos expression as a marker of neuronal activity, we evaluated whether fast and slow patterns of use differentially influenced activity within the prelimbic and infralimbic prefrontal cortex, the basolateral nucleus of the amygdala and the nucleus accumbens core and shell. Imposing a fast pattern of use increased both cocaine seeking and c-Fos expression in the five structures of interest. Qualitative differences were also observed, with fast and slow patterns of use differently impacting the functional recruitment of the neuronal network. Optogenetic experiments are in progress to test direct causal relationships between pattern of use, alterations in this neuronal network and cocaine seeking.
The individual vulnerability to develop a fast pattern of cocaine use could be a risk factor for increased cocaine seeking and addiction. Evidencing the mechanisms underlying this individual vulnerability would be a critical step for our understanding of the addiction process and our ability to prevent transition to addiction.

P1.090

Transcriptome changes during brain aging and Alzheimer´s disease-like pathology in the cortex of the lemurian primate *Microcebus murinus*: comparison of genechips and sequencing data

Devau, G. (Montpellier)[1], Jean, P.-A. (Montpellier)[1], Pichon, F. (Montpellier)[1], Janin-Nidelet, S. (Montpellier)[2], Dubois, E. (Montpellier)[2], Verdier, J.-M. (Montpellier)[1]
[1]*Inserm U710 - Université Montpellier 2 - EPHE, Université Montpellier 2, Montpellier, France,*
[2]*GenomiX, Montpellier, France*

Aging is the major risk factor for neurodegenerative disease such as Alzheimer´s disease (AD). However, the molecular events and their regulation occurring during aging still remain unclear.The aim

of this study was to identify genes with expression changes in relation with age or AD in the cortex of *Microcebus murinus*, a lemurian primate, which some of them develop, as they age, the pathognomonic lesions of AD: ß-amyloid plaques and cortical atrophy (AD-like animals). We have investigated these transcriptomic changes on temporal cortex samples using human Affymetrix microarrays (HGU113 Plus2). Over 14,911 transcripts detected, 152 were identified with significant changes in their expression in relation with age or with AD, and 47 revealed very discriminative (Abdel Rassoul et al, 2010).
These data were however incomplete because they only took into account the transcripts preserved during evolution. We have therefore pursued our investigation by high throughput sequencing (Illumina, Solexa HiSeq 2000 platform, Genomix) of 8 cortex samples (3 young adults, 3 healthy aged and 2 AD-like). A library was built for each sample, about 38 millions of short reads (50 nucleotides) was produced for each one. A preliminary analysis was performed using the Illumina's sequencing analysis software (CASAVA 1.8.2) for assembling RNA-seq to a super set of all transcripts resulting from Ensembl, known and pseudo gene predictions (micMur 1.67). About 13,000 genes have been identified for determining differential expression. The comparison of the data of the 3 groups allowed us to detect 2,258 genes differentially expressed (edgeR software package). We compared these genes with the genes detected by microarrays. We found that 80 on the previously 152 identified genes were sorted by the two approaches. This result validated microarray analysis with human genechips. Furthermore, microarray analysis and high throughput sequencing showed complementary informations. Indeed, the additional genes identified by mRNA sequencing will allow us to complete and strengthen the nature of the pathways, which essentially belong to cellular assembly and organization, and cell-to-cell signalling. Further analysis will allow us a better differentiation of the aging process from AD in the cortex.

P1.091

Bee venom alleviates motor deficits and restores functional balance in basal ganglia circuits in rat models of Parkinson's disease

Maurice, N. (Marseille)[1], Melon, C. (Marseille)[1], Deltheil, T. (Marseille)[2], Degos, B. (Paris)[3], Mourre, C. (Marseille)[2], Amalric, M. (Marseille)[2], Kerkerian-Le Goff, L. (Marseille)[1]
[1]Aix-Marseille Université/CNRS, UMR 7288, IBDML, Marseille, France, [2]Aix-Marseille Université/CNRS, UMR 7291, Marseille, France, [3]INSERM/Collège de France, U1050, Paris, France

Bee venom has been shown to exert a neuroprotective action on dopamine neurons in animal models of Parkinson's disease (PD). Here, we investigated whether bee venom can also exert a symptomatic action in PD by counteracting pathological activities within the basal ganglia network using combined pharmacological, behavioral and electrophysiological approaches in rat PD models. In the hemi-parkinsonian lesion model (unilateral intranigral injection of 6-OHDA), acute or subchronic bee venom treatment significantly alleviates contralateral forelimb akinesia and apomorphine-induced rotations. Bee venom action mechanisms were further investigated in a pharmacological model based on neuroleptic-induced catalepsy, reminiscent of parkinsonian akinetic deficit. Bee venom acute injection reverses haloperidol-induced catalepsy, an effect mimicked by apamin, a blocker of small conductance Ca2+-activated K+ (SK) channels, and blocked by CyPPA, an activator of these channels, suggesting the involvement of SK channels in the anti-parkinsonian action of bee venom. *In vivo* electrophysiological recordings of the neuronal responses evoked by motor cortex stimulation in the basal ganglia output structure (the *substantia nigra pars reticulata*) shows that bee venom prevents or reverses the neuroleptic-induced bias in the influence exerted by the direct inhibitory and the indirect excitatory striato-nigral circuits. Bee venom may thus alleviate parkinsonian-like akinetic deficits by restoring the functional inhibitory/excitatory balance within the basal ganglia circuits. These data provide the first evidence for a beneficial action of bee venom on the pathological functioning of the basal ganglia underlying motor PD symptoms with potential relevance to the symptomatic treatment of this disease. This work is supported by Association France Parkinson, CNRS and AMU.

P1.092

Use of 2D-DIGE to identify early markers of neurofibrillary degeneration in Alzheimer's disease

Sergeant, N. (Lille)[1], Fernandez-Gomez, F.-J. (Lille)[1], Eddarkaoui, S. (Lille)[1], Obriot, H. (Lille)[1], Zommer, N. (Lille)[1], Caillierez, R. (Lille)[1], Duban-Deweer, S. (Lille)[2], Christophe, F. (Lille)[2], Derisbourg, M. (Lille)[1], Deramcourt, V. (Lille)[1], Hamdane, M. (Lille)[1], Blum, D. (Lille)[1], Buée-Scherrer, V. (Lille)[1], Buée, L. (Lille)[1]

[1]Cité hospitalière - Bât. Gérard Biserte, INSERM UMR837, Alzheimer & Tauopathies, Univ. Lille Nord de France, Lille, France, [2]Université d'Artois, IFR114/IMPRT, Lille, France

Alzheimer's disease (AD) is the most prevalent cause of dementia in the elderly. Current treatments try to mitigate the disease evolution as well as the deleterious effects. However, early diagnosis markers and therapeutic targets remain difficult to establish and preventive treatment are thus precluded. Despite many proteins are prone to be candidates as potential early AD biomarkers, none yet showed sufficient accuracy to discriminate among neurological disorders. In order to find potential biomarkers of AD, we compared the brain proteome of AD compared to control patients as well as the brain proteome of Thy-Tau22 transgenic mouse model of neurofibrillary degeneration at early developmental stage of the lesion. We have adapted and optimized a protocol to perform two dimensional fluorescence difference gel electrophoresis (2D-DIGE) of human and mice brain tissue samples. Brain tissue protein samples were extracted, purified and labeled with cyanines. Firstly, proteins were separated according to their isoelectric point and secondly according to their molecular weights. 2D-DIGE proteomes of AD, Thy-Tau22 and controls were compared using SameSpots and spots significantly different between AD and controls but also found different between Thy-Tau22 and control mice were characterized. Several proteins were identified and differences were further confirmed using 2D gel electrophoresis followed by immunoblotting. Immunohistochemistry was also used to determine the association or not with AD neuropathological lesions of biomarkers. The results and protein identified will be presented. Most of the differences observed were related to post-translational modifications rather than changes in expression levels suggesting that 2D-DIGE is not only useful to identify quantitative modifications in protein expression. Further characterization is necessary to ascertain the post-translational modifications responsible for the differences observed by 2D-DIGE. In the line of these results, our data suggest post-translational modifications should be considered to increase the accuracy for the use biomarkers. Moreover, deciphering the mechanisms responsible for these post-translational changes may provide pharmacological targets for the current AD therapy.

P1.093

Hedgehog signaling blockade *in vivo* results in oligodendrocyte progenitor impairment during adult remyelination

Traiffort, E. (Gif sur Yvette)[1], Ferent, J. (Gif sur Yvette)[1], Ruat, M. (Gif sur Yvette)[1]

[1]CNRS, UPR3294, Gif sur Yvette, France

Overactivation of the Sonic Hedgehog (Shh) pathway in a healthy brain causes a significant increase in the proliferation of oligodendrocyte progenitor cells (OPCs), the source of mature oligodendrocytes, which are responsible for the formation of myelin sheaths (Loulier et al, J Neurochem, 2006). Now, we have investigated the status and role of Shh signaling in remyelination by using lysolecithin (LPC)-induced focal demyelination in wild-type or *plp*-GFP transgenic mice. We identified the oligodendrocyte lineage cells as a source of Shh protein within the lesion, soon after the LPC injection. To investigate the effects induced by the blocking of Shh activity in the lesion *in vivo*, we used the Ad-mHip adenoviral vector bearing the gene encoding Hedgehog-interacting protein (Hip), the natural physiological antagonist of Shh (Coulombe et al, Mol Cell Neurosci, 2004; Angot et al, Stem Cells, 2008). Ad-mHip or the corresponding control adenoviral vectors were stereotaxically injected into the right lateral ventricle of mice two days before LPC injection. Animals were killed 5, 10 or 15 days post-lesion for further analysis. The dynamics of OPC differentiation was analyzed with specific markers and stainings. While no increase in cell death was observed as demonstrated by TUNEL analysis, Hip-

mediated blocking of Shh pathway induced a complete arrest of new oligodendrocyte production. As a consequence, the demyelinated lesions, which normally repair by themselves, did not reduce. Our experiments suggest that Hip, when delivered before the LPC-induced lesion, antagonizes Shh produced during the acute phase of the lesion. They also demonstrate that the negative regulation of Shh signaling pathway plays a major role in tissue repair. Further experiments are in progress to analyze the effects of small molecules regulating the Shh pathway during remyelination.
Together with our recent findings identifying Shh as a positive regulator during demyelination (Ferent et al, J Neurosci, 2013), these investigations should give valuable insights concerning the potential use of pharmacological modulators of Shh signaling as a novel therapeutic approach for multiple sclerosis and other myelin diseases.

P1.094

Neural substrates related to levodopa-responsive and levodopa-resistant freezing of gait in Parkinson's disease: a PET study

Maillet, A. (Lyon)[1], Thobois, S. (Lyon)[2], Fraix, V. (Grenoble)[3], Redouté, J. (Bron)[4], Le Bars, D. (Bron)[4], Lavenne, F. (Bron)[4], Derost, P. (Clermont-Ferrand)[5], Durif, F. (Clermont-Ferrand)[5], Bloem, B. (Nijmegen)[6], Krack, P. (Grenoble)[3], Chabardès, S. (Grenoble)[3], Pollak, P. (Genève)[7], Debû, B. (Grenoble)[8]
[1]INSERM U836, Grenoble Institut des Neurosciences, Université Joseph Fourier, Grenoble / Institut des Sciences Cognitives, CNRS UMR 5229, Université Claude Bernard, Rhône, Lyon, France, [2]Hospices Civils de Lyon, Hôpital Neurologique Pierre Wertheimer, Service de Neurologie C / CNRS UMR 5229, Centre des Neurosciences Cognitives, Rhône, Lyon, France, [3]INSERM U836, Grenoble Institut des Neurosciences, Université Joseph Fourier / Centre Hospitalier Universitaire de Grenoble, Unité des Troubles du Mouvement, Isère, Grenoble, France, [4]CERMEP - Centre d'imagerie du Vivant, Rhône, Bron, France, [5]Hôpital Gabriel Montpied, Service de Neurologie, Puy-de-Dôme, Clermont-Ferrand, France, [6]Radboud University, Nijmegen, Netherlands, [7]Hôpitaux Universitaires de Genève, Genève, Switzerland, [8]INSERM U836, Grenoble Institut des Neurosciences, Université Joseph Fourier, Isère, Grenoble, France

Background: In Parkinson's disease (PD), gait disorders, including freezing of gait (a sudden inability to start or continue walking) are frequent and disabling symptoms that lead to severe alteration of the quality of life. Recent neuroimaging studies have shed some light on the neural correlates involved in gait impairment in PD. However, the influence of dopaminergic treatments and of pedonculopontine nucleus (PPN) stimulation - a surgical approach that has recently been proposed as a therapeutic alternative for the treatment of levodopa-resistant gait disorders - on these networks remains largely unclear.
Objectives: To explore the effects of levodopa and PPN stimulation on brain networks involved in freezing of gait respectively in PD patients suffering from levodopa-responsive (L+) and from levodopa-resistant (L-) freezing of gait.
Methods: Eight L+FoG patients, three L-FoG patients with bilateral PPN DBS and eight controls were included. All participants performed a motor imagery of gait and a control tasks during H_20^{15}-PET acquisitions. The L+FoG patients were tested both off and on medication, and the L-FoG patients off and on PPN stimulation.
Results: During motor imagery of gait, activations within the sensorimotor areas, basal ganglia (BG), and cerebellum were observed in controls. In L+FoG PD patients off medication, we observed supplementary activations within the superior parietal lobule (SPL) and pontine nuclei and reduced activation within BG. Levodopa administration restored BG loop activation and reduced parietal and brainstem overactivations. L-FoG patients off PPN stimulation showed as well overactivation of the SPL during motor imagery of gait. PPN stimulation restored activations within the BG, cerebellum, and brainstem - including pedonculopontine and cuneiform nuclei - and reduced the SPL activation.
Conclusions: Our PET data suggest that SPL and pontine nuclei activations could represent compensatory mechanisms aiming at improving gait when patients are off treatment. Both levodopa and PPN DBS tend to restore a more physiological pattern of brain activation and suppress accessory circuits' recruitments.

P1.095

Behavioral phenotyping of mouse models of cognitive disabilities

Meziane, H. (Illkirch)[1], Lalanne, V. (Illkirch)[1], Mittelhaeuser, C. (Illkirch)[1], Lux, A. (Illkirch)[1], Selloum, M. (Illkirch)[1], Birling, M.-C. (Illkirch)[2], Pavlovic, G. (Illkirch)[2], Chelly, J. (Paris)[3], Potier, M.-C. (Paris)[4], Humeau, Y. (Bordeaux)[5], Billuart, P. (Paris)[3], Sorg, T. (Illkirch)[6], Hérault, Y. (Illkirch)[7]
[1]Institut Clinique de la Souris (ICS), Phénomin, Neurosciences comportementales et cognitives, Illkirch, France, [2]Institut Clinique de la Souris (ICS), Phénomin, Ingénierie génétique, Illkirch, France, [3]Institut Cochin, Paris, France, [4]Institut du Cerveau et de la Moelle Epinière, Paris, France, [5]Institut interdisciplinaire de Neurosciences, Université Bordeaux 2, Bordeaux, France, [6]Institut Clinique de la Souris (ICS), Phénomin, Phénotypage, Illkirch, France, [7]Institut Clinique de la Souris (ICS), Phénomin, Illkirch, France

GENECODYS is a translational program aiming to identify new genes involved in cognitive dysfunctions, to elucidate molecular mechanisms underlying cognitive deficits, and to screen for potential therapeutics. In this regard, one of the objectives of GENECODYS consists of neurological and behavioral phenotypic analysis of mouse models for intellectual disability (ID), focusing particularly on evaluation of cognitive dysfunction.
Several mutant lines are being generated in the context of this program. Among genes of interest, mutations in the X-linked gene encoding the interleukin-1 receptor accessory protein-like 1 (IL1RAPL1) have been associated with nonsyndromic forms of ID, and more recently to autism. Deletion of IL1RAPL1 in mice causes a transient disinhibition of deep cerebellar nuclei neurons due to higher activity level of molecular layer interneurons (MLIs), supporting a key function during cerebellar development in establishing local excitation/inhibition balance. IL1RAPL1-KO mice were also shown to display a reduction of both dendritic spine density and excitatory synapses in the CA1 region of the hippocampus, associated with specific deficits in hippocampal long-term synaptic plasticity.
In the present study, IL1RAPL1-KO mice were submitted to behavioral tests designed to evaluate a wide range of functions or their pathologies including circadian activity, neurological reflexes and specific motor abilities, anxiety-related behavior, sensorimotor gating or learning and memory processes. Interesting behavioral traits were found in Il1Rapl1-KO mice, including hyperactivity and altered learning and memory processes, suggesting the relevance of these mutants for evaluation of cognitive dysfunctions related to ID and autism spectrum disorders, and for development of therapeutic strategies.

P1.096

High tPA release by neonate brain microvascular endothelial cells under glutamate exposure affects neuronal fate

Henry, V. (Rouen)[1], Lecointre, M. (Rouen)[1], Laudenbach, V. (Rouen)[1,2], Ali, C. (Caen)[3], Macrez, R. (Caen)[3], Julienne, A. (Caen)[3], Beresowski, V. (Lens)[4], Carmeliet, P. (Leuven)[5], Vivien, D. (Caen)[3], Marret, S. (Rouen)[1,2], Gonzalez, B. (Rouen)[1], Leroux, P. (Rouen)[1]
[1]Lab. Néovasc, Equipe Région INSERM (ERI 28), UFR de Médecine/Pharmacie Université Rouen, Rouen, France, [2]Service de pédiatrie néonatale et de réanimation, CHU de Rouen, Rouen, France, [3]Lab. SP2U, Equipe INSERM U919, Cyceron, Caen, France, [4]Lab. de Biologie Neurovasculaire Intégrée, Université d'Artois, Lens, France, [5]Lab. of Angiogenesis and Neurovascular Link, Vesalius Research Center, VIB, KU, Leuven, Belgium

Excitotoxicity is a consolidated hypothesis in neonatal brain injuries and tissue plasminogen activator (tPA) participates in the processes through proteolytic and receptor mediated effects. In brain microvascular endothelial cell (nBMEC) cultures from neonates, tPA content and release upon glutamate are higher than in adult (aBMECs) cultures. Owing to pleiotropic effects of tPA, the study was aimed at determining the putative roles of endothelial tPA in the neonatal brain parenchyma under glutamate challenge. Basal tPA release was 4.4 fold higher in nBMECs vs aBMECs and glutamate was 20 fold more potent to allow Evans blue vascular permeability in neonate microvessels indicating that, under noxious glutamate (50 µM) exposure, high amounts of endothelial tPA stores may be mobilized and may access the nervous parenchyma. Culture media from nBMECS or aBMECs challenged by excitotoxic glutamate were applied to neuron cultures at DIV11. While media from adult cells did not evoke more LDH release in neuronal cultures that under glutamate alone, media from nBMECs enhanced 2.2 fold LDH release. This effect was not observed with media from $tPA^{-/-}$ nBMECs and was inhibited by PAI-1. In Cortical slices from 10 day-old mice, tPA associated with glutamate evoked neuronal necrosis in deeper (more mature) layers, an effect reversed by NMDA receptor GluN1 amino-terminal domain antibody capable of inhibiting tPA potentiation of the receptor. In superficial layers (less mature), tPA alone inhibited apoptosis, an effect reversed by the EGF receptor antagonist AG1478. Applied to immature neurons in culture (DIV5), media from nBMEC rescued 85.1% of neurons from cell death induced by serum deprivation. In cortical slices, the anti-apoptotic effect of tPA fitted with age dependent localization of less mature neurons. These data suggest that in the immature brain, propensity of vessels to release high amounts of tPA may not only impact vascular integrity but may also influence neuronal fate, via regulation of apoptosis in immature cells and, as in adult by potentiating glutamate toxicity in mature neurons. The data suggest implication of microvessels in glutamate neurotoxicity in the development, and justify research towards vessel oriented neuroprotection strategies in neonates.

P1.097

Metabolic phenotyping of the Fragile X syndrome mouse model

Davidovic, L. (Valbonne)[1,2], Durand, N. (Valbonne)[1,2], Bardoni, B. (Valbonne)[1,2], Dumas, M.-E. (London)[3]
[1]IPMC, CNRS UMR7275, Valbonne, France, [2]Université de Nice Sophia-Antipolis, Nice, France, [3]Imperial College London, Biomolecular Medicine, Faculty of Medicine, Department of Surgery and Cancer, London, United Kingdom

Fragile X syndrome (FXS) is the first cause of inherited intellectual disability, due to the silencing of the X-linked Fragile X Mental Retardation 1 gene encoding the RNA-binding protein FMRP. While extensive studies have focused on the cellular and molecular basis of FXS, neither human fragile X patients nor the mouse model of FXS -the *Fmr1*-null mouse- have been profiled systematically at the metabolic and neurochemical level. Using proton high-resolution magic angle spinning nuclear magnetic resonance (1H HR-MAS NMR)-based metabolic profiling, we have previously identified a metabolic signature and biomarkers associated with FXS in various brain regions of *Fmr1*-deficient mice, involving notably neurotransmitters and oxidative stress response (Davidovic et al., 2011). To functionally connect Fmr1-deficiency to its metabolic biomarkers, we derived a functional interaction network based on the existing knowledge (literature and databases) and show that the FXS metabolic response is initiated by distinct mRNA targets and proteins interacting with FMRP. This opens new investigations avenues to understand the neuronal pathways perturbed in FXS.
We are now extending the metabolic phenotyping of FXS mouse model to the analysis of biofluids. Our data indicate a clear metabolic signature of FXS in urine and plasma, which strongly suggest that *Fmr1*-deficiency has systemic effects. These data pave the path to a systematic assessment of FXS treatment efficacy in its mouse model. These metabolomic and integrative strategies will contribute to develop potential drug targets and novel therapeutic interventions, which will eventually benefit FXS patients.

P1.098

Anatomical study on non-human primate that characterizes cortico-basal ganglia circuits involved in apathetic-like state with loss for food motivation, sexual manifestation and stereotyped behaviours

Sgambato-Faure, V. (Bron)[1], Worbe, Y. (Paris)[2], Epinat, J. (Bron)[1], Féger, J. (Paris)[2], Tremblay, L. (Bron)[1]
[1]*Centre de Neuroscience Cognitive, UMR 5229 CNRS Université Lyon I, Bron, France,* [2]*Centre de Recherche de l'Institut du Cerveau et de la Moelle épinière, Paris, France*

Convergent evidence from animal and human studies pointed to the ventral striatum (VS) as a major structure of interest for the neuropsychiatric disorders. Previously, we have shown that the VS could be implicated in various behavioural disorders via modulation of motivational process (Worbe et al., 2009). Namely, local injection of bicuculline ($GABA_A$ antagonist) into lateral, central and medial part of the VS induced hypoactivity (or apathetic-like state) associated with loss for food motivation, stereotyped behaviours (liking/biting fingers and grooming) potentially related to the induction of anxiety and sexual manifestations (erection) respectively. In this study we address the question of neuronal circuits that underpin these behavioural disorders. To this aim, in 10 monkeys we used the injection of axonal tracer biotin dextran amine (BDA) into the VS sites where bicuculline induced various behavioural effects. We also compared the reconstructed neuronal circuits with those described previously for the stereotyped behaviours (Worbe et al., 2012). The apathetic-like state was related to a strong neuronal cortical labelling in the dorsal (areas 24, 32, 8), lateral (area 45) prefrontal and insula cortices. The sexual manifestations were characterized by labelling in the medial and anterior prefrontal cortex (areas 13, 25, 10, 11) while the stereotyped behaviours were characterized by the strongest labelling in the orbitofrontal cortex (areas 12, 13, 14). On the basal ganglia level, the terminal fibers were found in the limbic part of both pallidal segments and substantial nigra pars reticulata for all behavioural effects. Nonetheless, there was a clear latero-medial gradient of the labelled regions in relation with the specific behavioural effect. Thus, the hypoactive state was mostly related with a labelling in the lateral subregions of the limbic territory and the sexual manifestations with the medial part. Labelling for the stereotyped behaviours was found in the junction between the areas related to the two other effects. Overall, our data pointed to the partially overlapping but distinct neuronal circuits underpinning the behavioural disorders induced from dysfunction in the VS and reflecting perturbation of different motivational domains.

P1.099

Vulnerability to opiate in maternally-deprived rats: implication of the epigenetic markers MeCP2 and HDAC2

Tesone-Coelho, C. (Strasbourg)[1], Bhatt, J. (Paris)[2], Estevez, L. (Paris)[2], Giros, B. (Paris)[2,3], Zwiller, J. (Strasbourg)[1], Daugé, V. (Paris)[2]
[1]*Univ. de Strasbourg, CNRS UMR 7364, Strasbourg, France,* [2]*INSERM U952 ; CNRS UMR7224 ; Université Pierre et Marie Curie, Paris, France,* [3]*Douglas Hospital Research Center, McGill University, Department of Psychiatry, Montréal, Canada*

When occurring during a critical period of neuronal development, maternal deprivation predisposes adult male rats to increased anxiety and opiate intake. This is accompanied by a basal hypoactivity of enkephalinergic system in the striatum. We searched for brain epigenetic mechanisms that possibly underlie the decrease in preproenkephain PPE levels in the striatum of maternally-deprived rats. For that, we analyzed the expression of the methyl-CpG-binding protein MeCP2 and the histone deacetylases HDAC2 and HDAC3, as well as the acetylation status of histone H3 and H4 in mesolimbic structures of adult maternally-deprived rats. Maternal deprivation was carried out daily for 3h, from postnatal day 1 to day 14. Three months later, the animals displayed increased MeCP2 and HDAC2, but not HDAC3 levels in caudate putamen and NAc core, but not in the ventral tegmental area. HDAC activity was also increased in the NAc of deprived animals and was accompanied by a

decrease in the acetylation content of histone H3 and H4. We next treated deprived rats for three weeks (weeks 12 to 14) with the HDAC inhibitor sodium valproate. The treatment was found to abolish the escalating behavior in the oral morphine consumption test. It was accompanied by normalized HDAC activity and normal acetylation of histone H3 and H4. The treatment also abolished the decrease in PPE mRNA levels in the NAc of deprived rats, indicating that the PPE gene was indeed under the control of HDAC activity. The data indicate that epigenetic mechanisms induced during early age through adverse environment memorize life experience, only to trigger greater vulnerability to drugs of abuse during adult life. They suggest that HDAC inhibitors may lessen vulnerability to opiate dependence, particularly in a subgroup of individuals with a history of traumatic experience during early infancy.

P1.100

Impact of a serotonergic lesion on parkinsonian symptoms and behavioral effects induced by L-DOPA treatment on MPTP-intoxicated monkeys

Beaudouin, M. (Bron)[1], Epinat, J. (Bron)[1], Neumane, S. (Bron)[1], Météreau, E. (Bron)[1], Tremblay, L. (Bron)[1], Sgambato-Faure, V. (Bron)[1]
[1]*Centre de Neuroscience Cognitive, Bron, France*

Convergent studies performed in humans have underlined an involvement of the serotonergic (5-HT) system in idiopathic Parkinson's disease (PD). Correlations were indeed evidenced between alterations of the 5-HT system and the expression of motor (tremor, dyskinesia) and non-motor (depression, fatigue) symptoms. Moreover, animal studies performed by our team evidenced an increase of striatal dopamine (DA) and serotonin (5-HT) in the striatum of parkinsonian monkey recovering from their motor symptoms, suggesting that 5-HT could participate to compensatory mechanisms. To investigate the involvement of 5-HT in the expression of parkinsonian symptoms and behavioral effects induced by L-DOPA, the main symptomatic treatment for PD, we developed a new monkey model of PD exhibiting a double DA/5-HT lesion due to sequential use of 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) followed by 3,4-methylenedioxy-N-methamphetamine (MDMA or Ecstasy). Depending on the mode of MPTP administration (acute or progressive), macaca *fascicularis* monkeys exhibited stable or transient motor symptoms and were therefore divided into stable or recovered groups. MDMA lesion did not evoke reappearance or worsening of tremor and akinesia/bradykinesia parkinsonian symptoms, suggesting that the 5-HT system does not play a compensatory role. But the 5-HT lesion counteracted the expression of rigidity when present in stable monkeys and favored it in recovered monkeys. Before MDMA, the long-term administration of L-DOPA evoked severe dyskinesia in stable monkeys and behavioural hyperactivity in recovered ones. Interestingly, both responses were drastically reduced after MDMA. Plastic changes occurring in response to L-DOPA both before and after MDMA were investigated longitudinally by PET (positron emission tomography) imaging using in particular [^{11}C]-PE2I and [^{11}C]-DASB, respective ligands of the DA and 5-HT transporters. Those results were then confronted to post-mortem analysis performed by immunohistochemistry against DeltaFosB, a marker for dyskinesia, and DA and 5-HT transporters. In conclusion, our results highlight a role of 5-HT system in L-DOPA-induced dyskinesia and behavioural hyperactivity but refute the hypothesis that the 5-HT is involved in motor compensatory mechanisms.

P1.101

Progressive parkinsonism by acute nigral excitatory amino acid transporter dysfunction in the rat

Assous, M. (Marseille)[1], Had-Aissouni, L. (Marseille)[1], Gubellini, P. (Marseille)[1], Melon, C. (Marseille)[1], Nafia, I. (Marseille)[1], Salin, P. (Marseille)[1], Kerkerian-Le Goff, L. (Marseille)[1], Kachidian, P. (Marseille)[1]

[1]*Institut de Biologie du Développement de Marseille-Luminy (IBDML), UMR7288 CNRS/Aix-Marseille Université, Marseille, France*

Parkinson's disease (PD) is characterized by the progressive degeneration of *substantia nigra* (SN) dopaminergic neurons presumably involving a multifactorial cascade of pathogenic events. Central players in PD pathogenesis, such as mitochondrial dysfunction and oxidative stress, might affect the function of excitatory amino acid transporters (EAATs), which play a major role in preventing excitotoxicity and maintaining physiological levels of GSH. Here we investigated whether dysfunction of EAATs might in turn contribute to the vicious cycles sustaining the progression of dopamine neuron degeneration. Acute nigral EAAT dysfunction was produced in rats by a single unilateral injection of the substrate inhibitor L-trans-pyrrolidine-2,4-dicarboxylate (PDC) in the SN. PDC triggered progressive loss of SN dopaminergic neurons without affecting the non-dopaminergic ones. Cell death evolved ipsilaterally with a typical caudo-rostral pattern and affected lately the contralateral uninjected side. This degenerative process associates GSH depletion and specific increase in γ-glutamyltranspeptidase activity, oxidative stress, excitotoxicity, autophagy and glial reaction. The anti-oxidant N-acetylcysteine and the NMDA receptor antagonists ifenprodil and memantine provided significant neuroprotection. Compensatory changes in dopamine function markers occurred in the SN (increased TH mRNA levels) and striatum (transient recovery of dopamine transporter expression). Late motor abnormalities (decreased spontaneous locomotor activity and forelimb akinesia) were detected when ipsilateral neuronal loss exceeded 50%. These findings show that acute dysfunction of nigral EAATs *in vivo* can drive a chronic self-propelling neurodegenerative process that recapitulates main PD pathogenic mechanisms and pathological features, thus providing a novel pharmacological PD rodent model.
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P1.102

Study of the pathophysiological implications of TREM-1 and its modulation during ischemic stroke

Darlot, F. (Vandoeuvre-les-Nancy)[1], Maskali, F. (Vandoeuvre-les-Nancy)[2], Strub, C. (Vandoeuvre-les-Nancy)[1], Gibot, S. (Vandoeuvre-les-Nancy)[1]
[1]*Université de Lorraine - Faculté de médecine de Nancy, UMR 1116, Vandoeuvre-les-Nancy, France,* [2]*Nancyclotep, Vandoeuvre-les-Nancy, France*

The theme of the research team focuses on the pathophysiological implications of TREM-1 and its modulation during inflammatory diseases.
In France, cerebrovascular accidents (CVA) are the first cause of non-traumatic handicap, the 2nd cause of dementia (after Alzheimer´s), and the third cause of death (after cancers and heart disease) in adults. The incidence of stroke is approximately 130 000 new cases per year (2 cases per 1000 inhabitants per year). The sensorimotor consequences associated have an impact and a socio-economic cost very high. It is an acute brain disease whose origin is vascular, and there are two types of stroke: hemorrhagic (20%) and ischemic (80%).
Inflammation is a key process in post-ischemic cascade observed after an ischemic stroke. Recently, a new family of receptors expressed on myeloid cells has been described: TREM family. All TREM-like receptors associate with the adapter DAP12. In this family, TREM-1 has been identified on the surface of neutrophils and mature monocytes. The function of this receptor TREM-1 is to modulate rather than initiate/activate the inflammatory response.
In this context, the objective of this research project is to highlight the role of TREM-1 in the cerebral ischemia and the consequences of its modulation. We study its modulation, by an activating antibody αTREM-1 and a peptide inhibitor LR12, and physiological consequences associated.
We use the MCAO protocol to induce experimental stroke. We study the evolution of stroke obtained by cellular and molecular approaches: immunohistochemistry, flow cytometry, WB, qRT-PCR, Elisa. We also carry out a study with PET imaging. We study TREM-1 as well as several factors associated with inflammation, plasticity, brain function...

P1.103

Brain monoamine concentrations in three genetic models of depression

Denoroy, L. (Lyon)[1], Parrot, S. (Lyon)[1], Zimmer, L. (Lyon)[1], Vaugeois, J.-M. (Rouen)[2], El Yacoubi, M. (Lyon)[1]
[1]*Centre de Recherche en Neuroscience de Lyon, Université Lyon1, Laboratoire de Neuropharmacologie, Lyon, France,* [2]*Université Rouen, EA 4651-ABTE /ToxEMAC, Rouen, France*

Three lines of “helpless” mice, called H/Rouen, H/TST and H/FST with different genetic backgrounds were developed by selective breeding from their long duration immobility in the tail suspension test (H/Rouen and H/TST) or in the forced- swim test (H/FST). In order to investigate whether brain monoaminergic systems could be involved in the mechanism of depressive behaviors exhibited by these animals, monoamines and metabolites contents were determined by HPLC with electrochemical detection in brain areas from adult H/Rouen, H/TST and H/FST female mice and compared to their respective “non-helpless” controls.
In the prefrontal cortex (PFC), noradrenaline (NA) concentration was increased (+54% $p< 0.001$) in H/Rouen mice whereas the 5-hydroxyindole acetic acid (5-HIAA) concentration and the ratio 5-HIAA/serotonin (5-HT) were decreased (-24% $p< 0.01$ and -31% $p< 0.05$). Both 5-HIAA and 5-HT concentration were decreased in H/TST mice (-28% $p< 0.05$ and -25% $p< 0.01$), while the ratio 5-HIAA/5-HT was unchanged. In contrast, 5-HIAA and 5-HIAA/5-HT ratios were increased in H/FST animals (+21% $p< 0.05$ and +30% $p< 0.05$). In the hippocampus (HIC), NA was slightly increased and 5-HIAA decreased (-28% $p< 0.01$) in H/TST mice. In the striatum (STR), dopamine (DA), dihydroxyphenylacetic acid (DOPAC) and homovanillic acid (HVA) were lower in H/TST mice (-40% $p< 0.01$; -44% $p< 0.05$; -34% $p< 0.01$); in contrast DA was increased in H/FST mice (+52% $p< 0.05$). 5-HT and 5-HIAA concentrations were decreased in both H/Rouen (-25%, $p< 0.001$ and -28% $p< 0.01$) and H/TST mice (-43% $p< 0.01$ and -39% $p< 0.001$) and 5-HIAA slightly increased in H/FST. In the hypothalamus (HYP) NA was lower in H/Rouen mice (-25% $p< 0.001$), whereas 5-HIAA and 5-HT were higher in H/FST (+38% $p< 0.05$ and +25% $p< 0.05$).
These data strongly suggest that the involvement of brain dopaminergic, noradrenergic and serotoninergic system may be different in these three genetic models of depression. In H/Rouen animals, NA metabolism may be higher in the CPF and decreased in HYP, while 5-HT metabolism is lowered in the CPF and STR. In H/TST animals, 5-HT metabolism is lowered in the CPF, HIC and STR, while DA metabolism is decreased in the STR. In contrast H/FST animals exhibit enhanced 5-HT metabolism in the CPF, STR and HYP.

P1.104

Characterization of a new monkey model of Parkinson's disease with dopamine and serotonin lesions: study associating PET, DTI and immunohistochemistry on monkeys treated with MPTP followed by MDMA

Metereau, E. (Lyon)[1], Beaudoin-Gobert, M. (Lyon)[1], Neumane, S. (Lyon)[1], Epinat, J. (Lyon)[1], Costes, N. (Lyon)[2], Lebars, D. (Lyon)[2], Ballanger, B. (Lyon)[1], Thobois, S. (Lyon)[1], Tremblay, L. (Lyon)[1], Sgambato-Faure, V. (Lyon)[1]
[1]*Centre de Neuroscience Cognitive, UMR5229-CNRS, Université Lyon1, Lyon, France,* [2]*CERMEP - Centre d'imagerie du Vivant, Lyon, France*

Idiopathic Parkinson´s disease (PD) is classically characterized by degeneration of dopaminergic (DA) neurons from the substantia nigra (SN). However alterations of serotonergic (5-HT) neurons from the raphe occur during the disease process as well. The impact of such 5-HT alteration, besides the DA one, on PD symptomatology has gained particular interest. In this context, we developed a double lesion model on monkeys by sequentially using MPTP to induce DA lesion and MDMA (Ecstasy) to alter the 5-HT system. Indeed, MDMA destroys 5-HT fibers in the normal monkey (Hatzidimitriou et al.,

1999).The impact of MPTP and MDMA was characterized with an *in vivo* brain imaging protocol using positron emission tomography (PET) with multiple tracers and diffusion tensor imaging (DTI) in combination with post-mortem immunohistochemistry. As expected after MPTP injection, we showed in PET a drastic and stable reduction of [^{18}F]-DOPA uptake in the SN and its projection areas, and by immunohistochemistry a strong loss of tyrosine hydroxylase (TH)-positive cells in the SN on post-mortem tissues. By using DTI, we further showed a decrease in fractional anisotropy (FA) of the SN, as well as a reduction of the number of tracts along the mesostriatal pathway. DTI data are in line with previous findings obtained in the MPTP-treated mice (Boska et al., 2007) and in PD patients (Chan et al., 2007) and constitute the first evidence that DTI could be used to assess the DA lesion in MPTP-treated monkeys. Regarding the effect of MDMA, we evidenced by PET using the [^{11}C]-DASB, a 5-HT transporter (SERT) ligand, a decrease of binding in all regions of the brain, and on post-mortem tissues an important reduction of SERT-positive fibers in basal ganglia regions, while raphe cells are preserved. Consistently, our preliminary DTI results show a decrease of the FA in the thalamus (target region of 5-HT neurons) but no change in the raphe. This study is pioneer to combine PET, DTI and Immunohistochemistry in order to assess neuronal integrity and plasticity in non-human primates. This multimodal approach validates that DA and 5-HT systems are altered following MPTP and MDMA administrations respectively and opens new avenues to investigate the role of 5-HT in PD symptomatology.

P1.105

Cognitive control, chronic neurophysiology and neuropharmacology in a slow MPTP-induced dopaminergic lesion in macaque monkeys prior to the onset of motor symptoms

Wilson, C. (Bron)[1,2], Stoll, F. (Bron)[1,2], Faraut, M. (Bron)[1,2], Vezoli, J. (Frankfurt am Main)[1,3], Leviel, V. (Bron)[1,2], Procyk, E. (Bron)[1,2]
[1]INSERM U846, Bron, France, [2]Univ. Claude Bernard Lyon I, Lyon, France, [3]Ernst Strüngmann Institute (ESI) for Neuroscience in Cooperation with Max Planck Society, Frankfurt am Main, Germany

We investigated the 'pre-symptomatic' phase and non-motor symptoms of Parkinson's disease (PD) with the neurotoxin MPTP in a rhesus macaque model.
Two monkeys learned the Problem Solving Task (PST; Procyk & Goldman-Rakic 2006), in which they had to search amongst several targets for a rewarded target (the search phase, SEA), and then repeat the correct response a number of times (the repetition phase, REP). A change signal then instructed them to begin a new problem by searching again. Monkeys were chronically implanted with at least 22 electrodes resting on the dura mater to provide electroencephalographic (ECoG) recordings.
We characterized electrophysiological markers, including the ERD/ERS pattern in the 20-30Hz beta oscillations over sensorimotor cortex, as well as other beta oscillations during the delay period prior to each trial. The stable beta oscillation in the delay period was modulated by the different cognitive control demands in the SEA and REP phases of the task. We also showed significant within-session modulation of the power and frequency of beta oscillations, an effect selective to the delay period of the task, demonstrating ongoing behavioural adaptation throughout a session related to measures such as time-on-task.
After this stable control period, monkeys received a slow low dose treatment of weekly injections of 0.2 mg/kg MPTP. Each week following an injection, monkeys received 2 days of rest followed by 5 days of testing with ECoG recordings. Progression of the treatment was also monitored using PET-imaging with the ligand [^{11}C]PE2I to follow levels of the dopamine transporter DAT, and scoring of motor symptoms on the Parkinsonian Monkey Rating Scale (PMRS).
Throughout the MPTP phase monkeys remained below the threshold for clinically significant motor symptoms on the PMRS, and so were in the 'pre-symptomatic' phase of the PD model. We observed maintenance of beta oscillatory power with ongoing treatment, along with the ability of this power to reflect differences between SEA and REP phases of the task. This effect can be seen in contrast to an increase in DAT binding potential in the early stage of the lesion (see poster by Leviel et al).

P1.106

5-HT$_4$ agonists: novel promising agents for AD prevention

Giannoni, P. (Montpellier)[1,2,3], Gaven, F. (Montpellier)[1,2,4], Baranger, K. (Marseille)[5], Rivera, S. (Marseille)[5], Marin, P. (Montpellier)[1,2,3], Bockaert, J. (Montpellier)[1,2,3], Claeysen, S. (Montpellier)[1,2,3]
[1]CNRS, UMR-5203, Institut de Genomique Fonctionnelle, Montpellier, France, [2]Inserm, U661, Montpellier, France, [3]Universités de Montpellier 1 & 2, UMR-5203, Montpellier, France, [4]Univesités de Montpellier 1 & 2, UMR-5203, Montpellier, France, [5]Aix-Marseille Université, NICN, UMR 7259, F-13344, Marseille, France

Objectives: 5-HT$_4$ agonists have been proved to exert procognitive effects in rodents and to induce the non-amyloidogenic processing of the amyloid precursor protein (APP), leading to an increase of soluble sAPPα (sAPPα). Therefore, 5-HT$_4$ receptors (5-HT$_4$R) could be of interest to delay AD progression. We studied the action mechanism of 5-HT$_4$R ligands and analyzed their effects on Aβ production and amyloid plaque formation.
Methods: COS-7 cells were stimulated with 5-HT$_4$R agonists and sAPPα release quantified through ELISA. Results were confirmed in vivo with i.p. injections in wild-type mice (WT). Chronic administration of 5-HT$_4$ agonists was performed in an aggressive mouse model of AD, the 5XFAD, during the prodromal phase preceding the appearance of behavioural deficits. Following treatments, amyloid plaque load and Aβ burden were measured through ELISA and thioflavin T staining. Astroglial inflammation associated to plaques was revealed through GFAP staining.
Results: 5-HT$_4$R agonists induced an increase of sAPPα release both in cell cultures and in WT mice. The chronic and prodromal administration of 5-HT$_4$R agonists to 5XFAD mice reduced the production of Aβ peptides and slowed down the formation of plaques. These effects were prevented by a co-treatment with a specific 5-HT$_4$R antagonist, that was ineffective by itself. Astroglial inflammation was also markedly reduced after 5-HT$_4$R agonist administration.
Conclusions: Chronic treatments promoting sAPPα release via the stimulation of 5-HT$_4$ receptors clearly hinder plaque formation and Aβ load while jointly attenuating inflammation processes. We conclude that 5-HT$_4$ agonists administration could represent an interesting and promising strategy for AD prevention.

P1.107

RGC proteomic analysis reveals c-myc as an effector of axon regeneration

Belin, S. (Boston)[1], Nawabi, H. (Boston)[1], Warren, P. (Boston)[1], Wang, C. (Boston)[1], Schorle, H. (Bonn)[2], He, Z. (Boston)[1], Steen, J. (Boston)[1]
[1]F.M. Kirby Neurobiology Center Children's Hospital Boston, Harvard Medical School, Boston, United States, [2]Institute for Pathology, University of Bonn, Department of Developmental Pathology, Bonn, Germany

A major reason for the devastating and permanent disabilities after injury to the central nervous system (CNS) is the inability of lesioned axons to regenerate and re-build functional circuits. Thus, there has been tremendous interest in understanding and overcoming CNS regeneration failure. While previous studies have extensively analyzed the inhibitory factors of the environment, it appears that other mechanisms, particularly decreased intrinsic ability of adult CNS neurons for axon growth contribute significantly to regeneration failure. Our previous studies showed that deletion of either tumor suppressor PTEN or SOCS3 or both genes induce a robust axonal regeneration in the central nervous system. However the intrinsic mechanisms involved in the failure of regeneration are not fully understood. To decipher the mechanisms, we performed a quantitative proteomic analysis of Retina Ganglia Cells (RGC) proteome before and after optic nerve crush. To specifically analyze the RGC proteome, we took advantage of a reporter mouse line that expresses YFP specifically in those cells. RGC were FACS sorted, lysed and digested. The resulting peptides were then labeled using isobaric

tags to allow relative quantification of protein expression between injured and control samples. Our experiment identified and quantified 1400 proteins. The experiment identified the MYC pathway in the injury response in addition of other pathways. C-MYC is a transcription factor that plays a critical role during development in regulating the expression of many genes involved in cell growth and proliferation. Our the current work using *in vitro* and *in vivo* experiments shows that the overexpression of c-MYC protects RGC from death after an optic nerve crush. Moreover, c-MYC activation induces robust axonal regeneration. Using mouse genetic models we also observed that the PTEN deletion and MYC activation have a robust synergistic effect. Our study is the first *in vivo* proteomic analysis of a pure neuronal population, which enabled us to discover new intrinsic factors such as c-myc that are involved in neuronal survival and CNS axon regeneration.

P1.108

Compulsive food eating in a animal model of Parkinson disease: model for behavioral addiction in Parkinson disease?

Dardou, D. (Clermont-Ferrand)[1], Durif, F. (Clermont-Ferrand)[2]
[1]*Université d'Auvergne, Faculté de médecine, EA7280 NpsySyDo, Clermont-Ferrand, France,*
[2]*Service de Neurologie A, Hôpital G. Montpied, Clermont-Ferrand, France*

In Parkinson´s disease (PD), beside the improvement on motor symptoms induced by dopamine replacement therapy, a growing number of studies have described the occurrence of impulse control disorders (ICD) such as: pathological gambling, compulsive shopping and compulsive eating behavior. However, it's still unclear why they appears in Parkinson´s patients and how they could be imitated in animal models. Development of food eating disorders, under the use of pramipexol (PPX) could provided an interesting way to explore such ICD in animal models. In the present study, we used rat with partial bilateral lesion of the VTA and daily injection of PPX in a model of intermittent daily feeding (IdF), a behavioral model inducing a slowly increase of sugar consumption over days and a strong addiction to it without period of food or water restrictions. Moreover, our experimental groups were evaluated in open fiield, spontaneous alternation in Y maze and on elevated plus maze, in order to ensure that they didn´t present motor or cognitives problems. Our results showed that PPX had no or little impact on motor and spatial memory capacities either in rats with VTA lesion and controls. On the contrary, we showed that PPX treatment induced a sligth decrease of the expression of anxiety-like behavior in animals with lesions. In the IdF experiment, we observed that animals with lesions of VTA and PPX treatment developed a strong and robust increase of their daily sucrose consumption since although rats with lesion of VTA and saline injection developed a modest increased of consumption. In addition, we have also investigated, using western blotting and immunochemistry, the possible change in dopamine D1, D2, D3 and mGLUR5 expression in brain link to this change in motivation to consumed sucrose in striatum and accumbens nuceus. While D1 expression not change, we noted an increase of D2, D3 receptor and mGLUR5 expression, mainly in nucleus accumbens brain regions. The present results open potential pists for investigating behavioral addiction in the context of PD and potential therapeutic targets to cure them.

P1.109

Use of maurocalcine as a vector for the delivery of antitumour agents to glioblastoma cells

Tisseyre, C. (Grenoble)[1], Perret, P. (Grenoble)[2], Broisat, A. (Grenoble)[2], Coquery, N. (Grenoble)[1], Ahmadi, M. (Grenoble)[2], Riou, L. (Grenoble)[2], Ronjat, M. (Grenoble)[1], Rémy, C. (Grenoble)[1], Ghezzi, C. (Grenoble)[2], De Waard, M. (Grenoble)[1]

[1]*INSERM U836, Grenoble Institut des Neurosciences, Université Joseph Fourier, Grenoble, France,* [2]*Inserm U1039, Laboratoire Radiopharmaceutiques Biocliniques, Université Joseph Fourier, Grenoble, France*

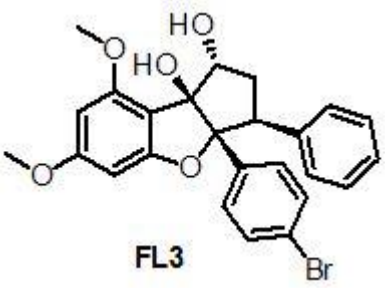
Maurocalcine (MCa) is a 33-mer peptide initially isolated from the venom of the scorpion *Scorpio Maurus Palmatus*. MCa has been characterised as the first animal toxin acting as a cell-penetrating peptide (CPP). The main CPP characteristic of MCa is its high number of positively charged residues (12 out of 33), many of them being critical for cell penetration. This peptide also interacts with membrane components involved in cell penetration, such as proteoglycans and negatively charged lipids.
Primary brain tumours are one of the ten main causes of death by cancer and the incidence of glioblastoma is approximately 5 to 10 per 100,000 (general population). Glioblastomas are highly aggressive tumours and despite multimodal treatments involving surgery, radiation, and chemotherapy, the prognosis associated with this pathology remains poor. This is partially due to the fact that those tumours tend to develop a resistance against both chemo- and radio-therapies. Previous studies have shown that coupling antitumour agents to CPP may represent a pertinent approach to overcome those resistances. Thus, using the cell penetration properties of MCa seems to be an interesting alternative for the delivery of antitumour agents within the cytoplasm of tumour cells. Here are presented the *in vitro* results of epifluorescence microscopy of MCa coupled to 5,6 carboxyfluorescein (FAM) or cyanine 5 (Cy5) and incubated on rat F98 glioma cells, illustrating the efficient cell entry of this peptide. To follow the in vivo distribution of MCa, a Tyr-MCa analogue was designed and radiolabelled with iodine 125. ^{125}I-Tyr-MCa displayed good radiochemical stability after 30 min of incubation in mouse blood *in vitro*.
The *in vivo* biodistribution of this radioactive complex was assessed after intravenous injection by SPECT imaging in both healthy mice and glioblastoma-bearing rats. The results demonstrate that besides being accumulated in the spleen, liver, and kidney, ^{125}I-Tyr-MCa accumulates preferentially in the tumour mass rather than in other regions of the brain, and is still present within the tumour at 24 hours post injection.
Altogether, these data indicate that MCa can be considered as a promising vector for the delivery of antitumour agents for the treatment of glioblastoma.

P1.110

Flavaglines are neuroprotective agents in experimental models of Parkinson disease

Bureau, C. (Paris)[1,2,3], Hild, A. (Paris)[1,2,3], Michel, P.P. (Paris)[1,2,3], Désaubry, L. (Illkirch)[4], Hirsch, E.C. (Paris)[1,2,3]
[1]*INSERM, UMR 975, Paris, France,* [2]*Université Pierre et Marie Curie - Paris 6, CR-ICM, UMR-S975, Paris, France,* [3]*CNRS, UMR 7225, Paris, France,* [4]*Therapeutic Innovation Laboratory, UMR7200, CNRS/Université de Strasbourg, Illkirch, France*

Flavaglines which constitute a family of compounds isolated originally from medicinal plants of Southeast Asia, display a number of interesting pharmacological properties. In particular, it was reported that flavaglines can operate as potent anticancer or cytoprotective agents (Ribeiro et al, J Med Chem, 2012). Here, we synthesized a series of novel flavagline derivatives and analyzed their neuroprotective potential using an *in vitro* model of neurodegeneration in Parkinson disease. More specifically, we used rat midbrain cultures and experimental conditions where the death of dopamine (DA) neurons is either spontaneous and progressive (Toulorge et al, Faseb J, 2011), or induced by application of MPP+ (Guerreiro et al, Mol Pharmacol, 2008), the active metabolite of the selective dopaminergic toxin MPTP. Structure-activity relationships carried out in both culture settings, allowed us to identify FL3 as a lead compound (see below for chemical structure). An optimal level of protection was observed in the low nanomolar range of concentrations. The incorporation of [3H]-thymidine was not affected in cultures exposed to neuroprotective concentrations of FL3, suggesting that survival promotion did not result from an anti-proliferative effect of this compound on dividing glial cells but rather from a direct effect onto DA neurons. Further characterization of this effect is ongoing.



[FL3]

P1.111

The PPN region integrates motor and non-motor information: an anatomical study in monkeys

Tandé, D. (Paris)[1], Karachi, C. (Paris)[1], Bensaid, M. (Paris)[1], André, A. (Paris)[1], Lau, B. (Paris)[1], François, C. (Paris)[1]
[1]*CRICM-UMR975 Inserm, Thérapeutique Expérimentale de la neurodégénéresce, E.Hirsch, Paris, France*

The pedunculopontine nucleus (PPN) and the cuneiform nucleus (CuN) play an important role in controlling gait and posture in mammals. However, this brainstem region also appears to be involved in other aspects of behavior such as motivation, attention and mnemonic processes. Anatomically, we have previously shown that the internal pallidum and the substantia nigra, which both integrate sensorimotor, associative and limbic information, project to different parts of the PPN and CuN. While the PPN is known to receive direct cortical inputs from motor areas, no data are available on cortical inputs from non-motor areas to the PPN and to the CuN.
Our aim is to determine first, if the PPN and/or CuN receive inputs from non-motor cortical areas, and second, if they send outputs to independent mesencephalic dopaminergic cell groups. For that, small deposits of biotine dextran amine (BDA) were injected into the PPN, CuN or infralimbic cortex of 4 macaques. As expected, BDA injection into the most anterior part of the PPN retrogradely labeled cell bodies in all motor cortical areas (A6, SMA, A8 or FEF, very few in A4). Labeled cell bodies were also seen in the anterior and ventral parts of the insular cortex. Moreover, PPN terminals were densely distributed in the ventrolateral part of dopamine A9 group. Tracer injection into the CuN revealed numerous retrogradely labeled cell bodies in the whole extent of the insular cortex and in the adjacent auditory and secondary somato-sensorial cortical areas. CuN axons were preferentially distributed in the medial A9 and A10 groups. Tracer injected into infralimbic cortex (Area 10mc) revealed numerous labeled terminals in the hypothalamus and periaqueductal area as already reported. A few labeled terminals were encountered in the most medial part of a region located between the ventral CuN and the dorsal PPN, avoiding PPN cholinergic neurons and reaching the adjacent laterodorsal tegmental nucleus. Our results suggest the existence of two separate pathways: a cortico-PPN-dopamine lateral A9 pathway that conveys motor information, and a cortico-CuN-dopamine medial A9 and A10 pathway that conveys somato-sensory information. This finding supports the hypothesis that the PPN region integrates motor and non-motor information.

P1.112

Early hippocampal synaptic plasticity and episodic like-memory deficits in a transgenic mouse model of Alzheimer disease - involvment of corticosterone

Lanté, F. (La Tronche)[1], Chafai, M. (Sophia-Antipolis)[2], Raymond, E. (Sophia-Antipolis)[2], Peirera, A.R. (Sophia-Antipolis)[2], Bethus, I. (Sophia-Antipolis)[2], Marie, H. (Sophia-Antipolis)[2]
[1]Grenoble Institut des Neurosciences U 836, La Tronche, France, [2]IPMC - CNRS/UNS UMR7275, Sophia-Antipolis, France

The etiology of Alzheimer's disease (AD) is unclear and no cure is yet available. The function of the hippocampus, a key structure responsible for memory encoding and consolidation, is affected early in AD leading to progressive irreversible memory loss.
There is strong evidence that AD onset is, at least partly, due to accumulation within the hippocampus of peptides processed from the amyloid precursor protein APP, such as amyloid-beta (Ab). Also, several studies demonstrated an abnormal elevation in the main stress hormone, cortisol (CORT in mice), in the initial phase of AD. Here, we investigated if early co-accumulation of CORT and Ab could be a main trigger driving the onset of memory deficits in AD. We used the Tg2576 mouse model, which accumulates hippocampal Ab with age and displays hippocampus-dependent memory loss. We first investigated the mechanisms underlying long term depression (LTD) and long term potentiation (LTP) in CA1 region of hippocampal slices of Tg2576 mice. We show that at 4 months of age, an age where Ab begins to accumulate, Tg2576 mice already display enhanced NMDA receptor-dependent LTD but unchanged LTP in CA1 pyramidal neurons compared to WT littermates. We also evaluated circadian plasma CORT levels by ELISA and show that while morning CORT levels are identical in 4 months old WT and Tg2576 mice, the evening level are significantly higher in Tg2576 mice. These data suggest alterations in CORT signaling. We further demonstrate that the LTD alteration observed in these Tg2576 mice is prevented by 4 days of subcutaneous injection of the glucocorticoid receptor antagonist, RU486. To better understand the impact of APP misprocessing, early synaptic deficits and increased CORT levels inTg2576 mice on episodic memory processing, the type of memory first affected in AD patients, we optimized an elaborated version of the object recognition (OR) task, which can probe for the “what, where and when” components of episodic memory. Our data show that WT mice have a good memory of the three components. By contrast, 4 months old Tg2576 mice show impairments in the spatial component “where” of this episodic-like memory. Altogether, these data identify early Ab/CORT-related alterations of hippocampal function with concomitant memory deficits.

P1.113

Inhibitors of DNA methyltransferases increase cocaine reinforcing properties in a self-administration paradigm

Fonteneau, M. (Strasbourg)[1], Romieu, P. (Strasbourg)[1], Zwiller, J. (Strasbourg)[1]
[1]CNRS, UMR 7364, Strasbourg, France

Chronic cocaine administration is known to produce long-term adaptations in brain structures that are mediated by persisting alterations of gene expression. Emerging evidence suggests that epigenetic marks, including DNA methylation and histone modifications, are essential mechanisms underlying drug-induced neuroadaptations. To test the involvement of DNA methylation on drug-reinforcing properties, we submitted rats to the intravenous cocaine self-administration paradigm. Using a fixed ratio-5 schedule, we found that repeated daily intracerebroventricular injections of the DNA methyltransferase (DNMT) inhibitors 5-aza-2'-deoxycytidine and zebularine dose-dependently increased cocaine self-administration at a cocaine dosage of 0.33 mg/kg/infusion. No such effect was noticed when DNMT inhibitors were given in a progressive ratio schedule. To further characterize the effect of DNMT inhibitors on the reinforcing properties of cocaine, we used a protocol in which the cocaine dosage gradually decreased from 0.33 to 0 mg/kg/infusion, during 30 min sessions separated by 15 min drug-free periods. We found that in this protocol, 5-aza-2'-deoxycytidine produced an upward shift of the dose-response curve for cocaine, suggesting that inhibition of DNMT actually increased the reinforcing properties of cocaine. Control experiments showed that the increase in drug-taking was not due to a general motor effect, since the inhibitor had no effect on locomotion during the habituation session or on the cocaine-induced hyperlocomotion. In summary, the results reveal that DNA demethylating agents such as 5-aza-2'-deoxycytidine or zebularine increase the reinforcing

properties of cocaine in rats without affecting the motivation for the drug. We next plan to identify a set of genes, the methylation of which is decreased by the treatment with the DNMT inhibitors. Some of them are likely candidates for underlying the neurobiological mechanisms responsible for the alteration in drug-seeking and drug-taking behavior.

P1.114

Postnatal inactivation of the prefrontal cortex induced an increase in dorsal striatal dopaminergic responses to ketamine

Louilot, A. (Strasbourg)[1], Usun, Y. (Strasbourg)[1], Eybrard, S. (Strasbourg)[1], Meyer, F. (Nijmegen)[2]
[1]Faculté de Médecine/FMTS/Université de Strasbourg, INSERM U1114, Strasbourg, France, [2]Radboud University Nijmegen, Donders Institute for Brain, Cognition and Behaviour, Nijmegen, Netherlands

Schizophrenia would result from a defective connectivity, between several integrative regions, stemming from developmental anomalies. Various abnormalities reminiscent of early brain development disturbances have been observed in the patients' left prefrontal cortex (PFC). The existence of a striatal dopaminergic (DA) dysregulation in schizophrenia is commonly acknowledged. Psychomimetic drugs such as the non-competitive NMDA/glutamate antagonist ketamine, can induce psychotic symptoms in healthy humans and exacerbate these symptoms in patients with schizophrenia. The striatal DAergic dysregulation in schizophrenia may be dependent on prefronto-striatal dysconnection involving glutamatergic NMDA receptors.
This study was designed to investigate the effects of ketamine in adult rats on locomotor activity and DA responses, in the dorsal striatum following a postnatal inactivation of the left PFC (infralimbic/prelimbic region). During the neurodevelopmental period, impulse electrical activity appears to be crucial for shaping connections once developing axons reach the target structure. Therefore, reversible functional inactivation of the left PFC was carried out by local tetrodotoxin (TTX) microinjection in rat pups at postnatal day 8 (PND8). Locomotor activity and DA variations in the dorsal striatum were recorded in parallel using in vivo voltammetry in freely moving adult rats (11 weeks). Ketamine was administred s.c. with different doses (5mg/kg; 10mg/kg; 20mg/kg); saline being the solvent.
The obtained results were the following:
1) A clear dose effect for locomotor responses and DA responses was observed for the two conditions (PBS and TTX microinjected at PND8);
2) Increases in locomotor response and DA levels in the dorsal striatum in adult animals after the administration of the highest ketamine doses (10mg/kg; 20 mg/kg) were more elevated in TTX microinjected animals compared to PBS microinjected animals.
These data suggest that animals microinjected with TTX in the left PFC at PND8 present higher reactivity to ketamine, than PBS animals. To conclude, these findings suggest that early functional impairment of PFC induced by TTX is a relevant approach for the animal modelling of the pathophysiology of schizophrenia.

P1.115

Expression of phospholipid molecular species in brain structures of juvenile and weaning rats following pilocarpine-induced *status epilepticus*

Bodennec, J. (Villeurbanne)[1], Ibeas Bih, C. (Villeurbanne)[2], Mameri, M.O. (Villeurbanne)[2], Dey, R. (Villeurbanne)[2], Bonnet, C. (Villeurbanne)[2], Georges, B. (Villeurbanne)[2], Belmeguenaï, A. (Villeurbanne)[2], Bezin, L. (Villeurbanne)[2]

[1]CRNL, INSERM U1028-CNRS UMR5292, Univ. Cl. Bernard Lyon1, Villeurbanne, France, [2]CRNL, INSERM U1028-CNRS UMR5292, Univ. Cl. Bernard Lyon1, Equipe TIGER, Villeurbanne, France

Phospholipids (PLs) are the main constituents of cellular membranes and are involved in many biological processes such as inflammation. Neuroinflammation is known to occur following *Status epilepticus* (SE) both in humans and in animal models of epilepsy. Surprisingly, and despite the crucial role played by PLs in inflammation, there are still sparse data on the putative changes that may occur in expression of these amphipatic lipids following SE in animal models. We thus determined PLs expression in different rat brain structures (Hippocampus, Amygdala, Piriform Cortex, Neocortex, and Cerebellum) 1 day and 7 days following SE induced by lithium-pilocarpine treatment either at weaning (21 days) or at juvenile stage (50 days). In juvenile rats, SE resulted in a decrease (» -40%, 7 days post-SE) in total PLs content in Amygdala, but not in other brain structures. Individual phospholipid analysis showed that the decrease in total PLs content was mainly due to hydrolysis of phosphatidylethanolamine and phosphatidylcholine, i.e., the two main brain PLs. A drop (- 40%) in sphingomyelin concentrations was also observed in Amygadala one day after SE, suggesting hydrolysis of this sphingophospholipid to the pro-apoptotic ceramide lipid messenger. Hydrolysis of phosphatidylinositols, phosphatidylserines and diphosphatidylglycerols, but not of phosphatidylcholines and phosphatidylethanolamines, were observed in the Hippocampus. We also analyzed the changes that may occur in molecular species of total and individual PLs. Fatty acid analysis showed that remodeling of phospholipid molecular species occurs in all brain regions following SE but at different extent. For instance, although concentrations of phosphatidylcholine are not changed after SE in Hippocampus, its fatty acid composition is strongly altered, with palmitic acid representing 33 % of fatty acids in controls and 20 % 7 days after SE. In conclusion, major changes in expression of PLs and their molecular species occur in all brain regions after SE. These changes differ according to the brain region and with time following SE. The potential meanings of these results are discussed. Data on the expression of PLs molecular species in young rats subjected to SE are also presented.

P1.116

Psychological stress increases short-term cognitive deficits in a rat model of moderate traumatic brain injury

Ogier, M. (Villeurbanne)[1,2], Carré, E. (Villeurbanne)[1,2], Belmeguenai, A. (Villeurbanne)[1], Mermet, N. (Villeurbanne)[1], Bouvard, S. (Villeurbanne)[1], Bonnet, C. (Villeurbanne)[1], Bezin, L. (Villeurbanne)[1]
[1]Centre de Recherche en Neurosciences de Lyon - Equipe TIGER, CNRS UMR5292, INSERM U1028, Université Claude Bernard Lyon 1, Villeurbanne, France, [2]Institut de Recherche Biomédicale des Armées, Dept de Soutien Médico-Chirurgical des Forces, Brétigny-sur-Orge, France

Traumatic brain injury (TBI) is a leading cause of death and disability worldwide. Despite extensive pre-clinical and clinical research, there is still no treatment for TBI. Intriguingly, extrinsic factors that could worsen the outcomes of TBI in humans, such as systemic insults or stress, are understudied in animal models of the pathology. Thus, it is possible that current experimental models of TBI do not reproduce the complete pathophysiology of human TBI. Here, we aimed at determining in a rat model of TBI if behavioral and cognitive outcomes can be worsened by prior exposure to psychological stress. Adult male Sprague Dawley rats were exposed to a predator odor (fox feces: 2-4-5-trimethyl-3-thiazoline, TMT) or water for 7 minutes every 2-3 days during 2 weeks. Then, a subset of rats from each exposure group underwent either Sham surgery or bilateral moderate fluid percussion injury (LFP). Thus, we studied four experimental groups: Sham, Sham-TMT, LFP and LFP-TMT rats. We measured motor activity and anxiety-like behaviors in these groups of rats at 7, 14 and 43 days post-trauma and spatial learning at 1 month post-trauma using the Water Exploration Test (WET) and the Morris Water Maze (MWM), respectively. In these rats, we also measured at 2 months post-trauma the hippocampal long-term potentiation (LTP), an index of the activity of the neural substrates of learning and memory, using *in vitro* slice electrophysiology. We detected transient signs of motor hyperactivity at 7 days post-trauma but no signs of anxiety in LFP rats compared to Sham rats. TMT exposure had no effect on motor activity and anxiety-like behaviors in Sham and LFP rats. We also detected slight learning deficits in LFP rats at 1 month post-trauma compared to Sham animals. TMT exposure had

no effect on the learning ability of Sham animals; however, it further enhanced learning deficits in LFP rats. Intriguingly, no deficit in hippocampal LTP could be measured in LFP and LFP-TMT rats at 2 months post-trauma compared to Sham and Sham-TMT rats. Hence, these data suggest either that cognitive deficits are only transient, or that spatial learning and hippocampal LTP are not functionally linked, in injured rats.

P1.117

Erythropoietin abrogates beneficial effects of environmental enrichment on learning and memory in healthy and epileptic rats

Belmeguenai, A. (Villeurbanne)[1], Mermet, N. (Villeurbanne)[1], Fares, R.P. (Villeurbanne)[1], Kouchi, H.Y. (Villeurbanne)[1], Bonnet, C. (Villeurbanne)[1], Bodennec, J. (Villeurbanne)[1], Georges, B. (Villeurbanne)[1], Bezin, L. (Villeurbanne)[1]
[1]*Université Lyon 1, CRNL, UCB Lyon1 - U1028 Inserm - UMR5292 CNRS, Villeurbanne, France*

Cognitive impairment is a condition frequently associated with pediatric epilepsy that can severely affect the quality of life of children with epilepsy and their family. Therefore, there is an urge to find therapeutical and alternative interventions aimed at preventing or reducing the decline of cognitive functions. In rats raised in conventional environment (CE), we showed that recombinant erythropoietin (Epo) treatment (1 daily i.p. injection for 5 consecutive days at 5000IU/kg) following pilocarpine-induced *status epilepticus* (Pilo-SE) at weaning prevented alteration of spatial memory and synaptic plasticity. Housing rats in an environmental enrichment (EE) alone just after Pilo-SE produced the same effects as Epo alone. However, the amplitude of LTP in rats subjected to Pilo-SE and raised in EE did not reach the magnitude measured in healthy rats raised in EE. We therefore tested the hypothesis that the combination of Epo and EE after Pilo-SE may help to reach this goal. Unexpectedly, we observed that this combination totally abolished the beneficial effects of both Epo and EE in rats subjected to Pilo-SE. In order to determine whether this catastrophic effect was specific to injured rats, we tested the effect of this combination in healthy rats. While EE had highly positive effects on LTP amplitude, Epo by itself had not effect when given to rats raised in CE. Combination of Epo treatment with EE in healthy rats produced the same effects as in rats subjected to Pilo-SE : it not only prevented the induction of LTP usually observed in rats raised in EE, but also reduced LTP amplitude to the level of rats subjected to Pilo-SE and raised in CE. All these effects have first been observed in the short term following Pilo-SE. We thus examined whether the dramatic effect of Epo combined with EE could be counterbalanced in long term by EE. We observed that long term EE almost restored LTP expression in epileptic rats, but not in healthy rats. Altogether these results demonstrate that the beneficial effect of Epo on learning and memory is highly dependent of the sensory and social stimuli during living: the worst effects were observed when Epo was administered to rats highly stimulated during housing in EE, in both healthy and pathological conditions.

P1.118

Apamin, a small potassium-calcium activated channel blocker, reverses cognitive deficits in a rat model of Parkinson disease

Mourre, C. (Marseille)[1], Chen, L. (Marseille)[1], Liberge, M. (Marseille)[1], Watanabe, I. (Marseille)[2], Amalric, M. (Marseille)[1]
[1]*UMR 7291, Laboratoire de Neurosciences Cognitives, CNRS-AMU, Marseille, France,* [2]*UMR 7260 Neurosciences Intégratives et Adaptatives, Marseille, France*

Neurodegeneration of dopaminergic (DA) midbrain neurons is the hallmark of Parkinson's disease (PD). Tonic or phasic firing activity of DA neurons is known to modulate DA release in the nigrostriatal pathway. The calcium-activated-potassium channels (SK) activity increases DA release and is crucial to the DA midbrain neuron firing by stabilizing the intrinsic pacemaker activity, and preventing burst firing in these neurons. The specific blockade of SK channels by apamin facilitates memory and alters emotional memory. Because Parkinson's disease is characterized by cognitive and emotional deficits occurring early in the disease before the motor symptoms, we tested whether apamin could reduce the cognitive deficits in a rat model of early PD. Rats received bilateral intrastriatal injection of the DA neurotoxin, 6-hydroxydopamine (6-OHDA), that produce a progressive loss of DA nigrostriatal neurons. Two weeks later, rats were injected with apamin (0.1 or 0.3 mg/kg i.p.) before being tested in the elevated-plus-maze, social recognition, object recognition and spatial memory tests. Results indicate that apamin treatment drastically attenuates 6-OHDA-induced anxiety-like deficits and short-term spatial but not non-spatial object recognition memory, suggesting that apamin may have potential as an early treatment for non-motor symptoms of PD.

P1.119

Development of two genetic mouse models of "behavioral despair" based on a bidirectional selective breeding strategy

El Yacoubi, M. (Lyon)[1], Arthaud, S. (Lyon)[1], Parmentier, R. (Lyon)[1], Martin, B. (Rennes)[2], Malleret, G. (Lyon)[1], Vaugeois, J.-M. (Rouen)[3]
[1]CRNL, CNRS UMR5292, INSERM U1028, Université Lyon1, Lyon, France, [2]INSERM, U642, Rennes, Université de Rennes 1, LTSI, Rennes, France, [3]Université de Rouen, EA 4651-ABTE /ToxEMAC, U.F.R. Médecine & Pharmacie, Rouen, France

Depression is a multifactorial illness and genetic factors play a role in its etiology. The understanding of its physiopathology relies on the availability of experimental models potentially mimicking the disease. We previously built a genetic mouse model of depression based on a selective breeding of mice displaying, or not, helplessness (a cardinal symptom of depression in human) in the tail suspension test (TST). In this model, helpless CD1 "H/Rouen" mice are thus essentially immobile in the TST, but also in the Porsolt forced swim test (FST), the two major stress paradigms aimed at screening potential antidepressants, whereas nonhelpless "NH/Rouen" show the opposite behavior i.e. very low immobility. However, it is still unclear to what extent the two tests screened the same form of helplessness or depression. Here, we replicated this study on a different genetic mix (an eight-way cross with ABP/Le , A/J, BALB/c B6, C3H, CBA/H, DBA/2, , SWxL-4, as founders) and obtained three mouse lines called H/TST (helpless), I/TST (intermediate) and NH/TST (nonhelpless) selectively bred according to their helplessness in the TST. We compared them to another mouse model selectively bred according to their helplessness in the FST called H/FST (helpless), I/FST (intermediate) and NH/FST (nonhelpless). As expected, H/TST and H/FST are helpless in both the TST and the FST as compared to their NH and I controls. However, H/TST mice displayed anhedonia, high anxiety and altered paradoxical sleep as compared to H/FST mice which do not display such impairments. Our results suggest that H/TST and H/FST mice may model different forms of depression as they can be described in human.

P1.119bis

Longitudinal changes of functional activation and connectivity in OCD patients undergoing cognitive-behavioral therapy: a fMRI study

N'Diaye, K. (Paris)[1], Morgieve, M. (Paris)[1], Tezenas, C. (Paris)[1], Haynes, W. (Paris)[1], Clair, A.-H. (Paris)[1], Granger, B. (Paris)[2], Pelissolo, A. (Paris)[3], Mallet, L. (Paris)[1]
[1]Université Pierre-et-Marie Curie, INSERM U 975, CNRS UMR 7225, Institut du Cerveau et de la Moelle Épinière (ICM), Paris, France, [2]CHU Pitié-Salpêtrière, Département de Santé Publique, de Biostatistiques et d'Information Médicale (bioSPIM), Paris, France, [3]CHU Pitié-Salpêtrière, Department of Psychiatry, Paris, France

While anomalies of task-related metabolic responses may be detected by functional MRI (fMRI), resting-state fMRI has the ability to reveal task-independent functional networks whose connectivity may also be altered in clinical conditions. Obsessive-compulsive disorder (OCD) is an anxiety disorder characterized by recurrent negative thoughts and worries as well as repetitive behaviours or mental acts. The neurobiological bases of OCD remain poorly understood so as the psychotherapeutical mechanisms involved in cognitive behavioural therapy (CBT), a well-validated treatment for OCD. Cross-sectional studies using fMRI and functional connectivity MRI (fcMRI) have recently accumulated, showing abnormal metabolic responses and distinct brain networks alterations in OCD patients compared to controls. However whether these functional networks also reflect therapeutical interventions remains untested. N=35 moderately severe OCD patients who followed a 3-month CBT participated in the study. Along with clinical assessment, fMRI during an exposure task (with neutral, generic and personalised obsession-inducing images) and fcMRI (10-min resting-state) were acquired on a 3T scanner at 4 time points throughout the therapy: before, halfway through, at the end, and 6 months after. Data were processed using the standard SPM8 pipeline and the CONN toolbox. Contrasting image types, fMRI revealed increased activity in anterior cingulate (ACC) and left and right orbitofrontal (L/R-OFC) cortices triggered by personalised obsession-inducing images. These metabolic responses in anterior cingulate and left but not right orbitofrontal clusters diminished as patients improved and continued to decrease after stabilisation of clinical symptoms at the outset of therapy. Preliminary analyses of fCMRI using the three fMRI clusters as regions of interest indicate that while ACC/L-OFC connectivity decreased through therapy, R-OFC connectivity with either region did not change. These results, if confirmed by further analyses, confirm the ACC and OFC as key regions in OCD pathophysiology, which may provide by themselves and through their functional interactions promising biological markers of response to treatment in OCD.

P1.120

Inter-areal causal interactions in the Gamma and Beta frequency bands reflect the anatomical hierarchy of primate visual system

Vezoli, J. (Frankfurt am Main)[1], Bastos, A. (Frankfurt am Main)[1,2,3], Bosman, C. (Nijmegen)[2], Schoffelen, J.-M. (Nijmegen)[2], Oostenveld, R. (Nijmegen)[2], Kennedy, H. (Bron)[4,5], Fries, P. (Frankfurt am Main)[1]
[1]Ernst Strüngmann Institute (ESI) for Neuroscience in Cooperation with Max Planck Society, Frankfurt am Main, Germany, [2]Donders Institute for Brain, Cognition and Behaviour, Radboud University Nijmegen, Nijmegen, Netherlands, [3]Center for Neuroscience and Center for Mind and Brain, University of California- Davis, Davis, United States, [4]Stem Cell and Brain Research Institute, InsermU846, Bron, France, [5]Université de Lyon, Lyon1, Lyon, France

Cortico-cortical connectivity has been shown to be hierarchically organized such that bottom-up and top-down information are conveyed through the well-defined feedforward and feedback counter-streams, respectively. The anatomical patterns of these two main pathways in terms of source and termination layers allow dissociation of top-down and bottom-up pathways. However, it remains unclear which neurophysiological markers characterize feedforward and feedback connections. Further, strong interlaminar connectivity means that these signals could quickly become locally intermixed. The cortex might thus retain additional mechanisms to functionally segregate these different paths of information flow. Recent electrophysiological studies suggest that Gamma rhythms might function as a forward information process because they are predominantly found in the supragranular layers whereas Beta rhythms might function as a backward information process because they are strongest in the deep layers (Buffalo et al., 2011). We thus tested this hypothesis by analyzing causal interactions in the Gamma and Beta frequency bands between seven visual areas of

macaque monkeys performing a visuospatial attention task. LFP signals were recorded through electrocorticography (ECoG) and analyzed through spectrally resolved Granger causality. We found that Gamma-band influences were predominant in the bottom-up direction, whereas Beta-band influences were predominant in the top-down direction. This functional asymmetry was significantly correlated with quantitative anatomical data. We exploited these results to build a cortical hierarchy based on functional data alone. The resulting model was highly similar to the anatomical hierarchical model thus pointing to the strong link between structure and function in the cortex. These results open the possibility for the *in vivo* investigation of functional hierarchies in the healthy and pathologic human brain.
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J.V, A.M.B and C.B contributed equally to this work.

P1.121
Adaptation of motor preparation to lapses in attention: a transcranial magnetic stimulation study

Derosière, G. (Montpellier)[1,2], Billot, M. (Montpellier)[1], Ward, T.E. (Maynooth)[2], Perrey, S. (Montpellier)[1]
[1]*Movement to Health Laboratory (M2H), EuroMov, UFR STAPS, Montpellier, France,* [2]*National University of Ireland Maynooth (NUIM), Biomedical Engineering Research Group (BERG), Maynooth, Ireland*

Sustained attention tasks are characterized by a gradual decrease in attentional resources across time usually investigated through an increase in reaction time (RT). Weissman et al. (2006) depicted by means of functional magnetic resonance imaging (fMRI) that neural adaptations to lapses in attention involve an increase in the activity of the fronto-parietal network. Specifically, the primary motor cortex (M1) exhibits an increase in stimulus-related fMRI response once attentional lapses occur (Weissman et al., 2006; Yarkoni et al., 2009). In the present study, we investigated whether pre-stimulus neural activity increases when attentional lapses occur. We hypothesized that the increased stimulus-related fMRI response previously found once attentional lapses occur was related to an increase in motor preparation occurring during inter-stimuli intervals. To examine this, the time course of corticospinal excitability was investigated by means of single-pulse transcranial magnetic stimulation (TMS) throughout a sustained attention RT task of 32 min duration. Single-pulse TMS was applied at 5 min intervals, while subjects were preparing for the upcoming stimulus. Attentional lapses were observed as revealed by a significant increase in RT ($p < .001$) after 9 min of sustained attention. Friedman ANOVA showed that motor-evoked potential amplitude, reflecting the level of corticospinal excitability, (i) significantly increased between a period 15 min into the task compared to pre-task resting state values ($p < .01$) and (ii) returned to the pre-task resting state values once the task finished. In accordance with the interpretation given by Weissman et al. (2006), we propose that the motor structures become increasingly active once lapses in attention occurs to cope with the increasing task demand over time. This over-engagement is not reflected by an increase in stimulus-related neural response of the M1 area only but also involves an increase of the corticospinal excitability during the motor preparation phase.

P1.122
Spinal delta opioid receptors regulate both heat and mechanical noxious stimuli in the rat

Luccarini, P. (Clermont-Ferrand)[1], Normandin, A. (Sherbrooke)[2], Molat, J.L. (Clermont-Ferrand)[1], Gendron, L. (Sherbrooke)[2], Dallel, R. (Clermont-Ferrand)[1]
[1]*Clermont Université, INSERM U1107 'Neuro-Dol', Équipe Douleur Trigéminale et Migraine, Clermont-Ferrand, France,* [2]*Université de Sherbrooke, Laboratoire de Physiologie et Biophysique, Sherbrooke, Canada*

Expression of delta opioid receptors (DOPR) in polymodal nociceptors has been recently challenged. Indeed, DOPR was shown to be almost exclusively expressed by small non-peptidergic, IB4-positive primary afferents. Moreover, spinally-delivered DOPR agonist has been shown to attenuate mechanical- but not heat-induced nociception. In the present study, we investigated the role of DOPR

in heat and mechanical pain regulation by using electrophysiological and immunohistochemical approaches in rats. In electrophysiological studies, we assessed the effect of intrathecal deltorphine II, a DOPR-selective agonist, on heat- and mechanical-induced diffuse noxious inhibitory controls (DNIC). We recorded from wide-dynamic range (WDR) neurons in the spinal trigeminal nucleus of halothane-anesthetized rats. Trains of 105 electrical shocks (0.66 Hz) were delivered to the excitatory cutaneous receptive field at 3 times the threshold for C-fibers activation. To trigger DNIC (that induced inhibition of the C-fibers-evoked responses of WDR neurons), either heat (immersion in a 49°C waterbath) or mechanical (pressure 250 g) noxious stimulation was applied to the inflamed (complete Freund adjuvant) hindpaw between the 36^{th} and 60^{th} stimuli. To determine whether peptidergic primary afferents are involved in DNIC triggered by noxious heat and mechanical stimulation, measurement of substance P release in the spinal cord was performed by visualizing NK_1 receptors internalization. We found that deltorphine II strongly attenuates DNIC triggered either by heat or mechanical noxious stimulation of inflamed hindpaw. Such reductions were reversed by systemic administration of the selective DOPR antagonist, naltrindole (4 mg/kg). In superficial laminae of the spinal cord, deltorphine II significantly reduced NK_1 receptor internalization induced by noxious heat and mechanical stimuli. Altogether, our results reveal that activation of spinal DOPR efficiently relieves both heat- and mechanical-induced pain and therefore further suggest that this receptor is expressed on polymodal, substance P-expressing neurons.

P1.123

Long-term olfactory memory in depression and Alzheimer disease

Naudin, M. (Tours)[1], Mondon, K. (Tours)[1,2], El-Hage, W. (Tours)[1,3], Desmidt, T. (Tours)[2], Lorette, A. (Tours)[2], Belzung, C. (Tours)[1], Gaillard, P. (Tours)[1,3], Hommet, C. (Tours)[1,2], Atanasova, B. (Tours)[1]
[1]INSERM U930 ERL, Université François Rabelais, Tours, France, [2]ALS Center, CHRU Bretonneau, Department of Neurology, Tours, France, [3]Clinique Psychiatrique Universitaire, CHRU de Tours, Tours, France

Many studies have revealed olfactory deficits in psychiatric and neurodegenerative disorders. The detection of specific olfactory markers in these diseases could improve the screening and the therapeutic care of the patients.
The aim of the present study was to investigate the long term olfactory memory in depressed and Alzheimer's disease patients in order to find specific olfactory markers. We included 20 depressed patients, 20 patients with Alzheimer's disease and 24 healthy subjects matched in age, gender and smoking status. We investigated neuropsychological profile (psychometrics scales) and olfactory memory ability for familiar and unfamiliar odors.
On the one hand, our results demonstrated a decrease of olfactory memory performance both for familiar and unfamiliar odors in depressed patients compared to controls. On the other hand, Alzheimer patients failed only for unfamiliar odors compared to controls. We found no significant difference between depressed and Alzheimer patients.
For the first time, our study investigated long term olfactory memory using familiar and unfamiliar odors in Depressed and Alzheimer's disease patients. The confirmation and validation of a preserved olfactory memory for familiar stimuli in a large population of Alzheimer patients could allow clinicians to use this olfactory memory test as a complementary tool to clarify the diagnosis.

P1.124

Multiple reference frames in saccadic adaptation

Lévy-Bencheton, D. (Bron)[1], Pélisson, D. (Bron)[1], Panouillères, M. (Bron)[1], Urquizar, C. (Bron)[1], Tilikete, C. (Bron)[1,2], Pisella, L. (Bron)[1]
[1]INSERM U1028; CNRS UMR5292; Université Lyon 1, Lyon Neuroscience Research Center, Impact Team, Bron, France, [2]Hôpital Neurologique Pierre Wertheimer, Neuro-Ophthalmology Department, Bron, France

Saccadic adaptation allows changes of saccade over time after repeated post-saccadic error (McLaughlin, 1967). Adaptation is known to occur in oculocentric coordinates and to be unidirectional: adaptation of rightward saccades does not transfer to leftward saccades. However, Zimmermann et al. (2011) proposed that adaptation can unfold in spatiotopic coordinates: outward adaptation of rightward scanning saccades transferred to all *memory-guided double-saccades* (including those performed in the non-adapted direction) such that their endpoint shifted rightward (as if the visual target was re-localized). But the authors questioned whether their subjects could have used the visible frame of the computer screen as a reference, thus allowing adaptation in allocentric -rather than in spatiotopic- coordinates. In the present study, we re-assessed this question by using targets LEDs presented in complete darkness to avoid any visual reference. Nine subjects performed the paradigm of Zimmermann et al. (2011) in this *no-frame condition* and in a *frame condition*, in two separate sessions. Repeated measures ANOVA (factors: frame/no-frame and pre/post) performed on the amplitude of leftward memory-guided double-saccades revealed a significant decrease after adaptation only when a frame was provided (interaction $p< 0.01$), in agreement with the allocentric hypothesis of adaptation. Note however that the remapping processes which encode the parameters of the second saccade in this double-saccade task may have favored allocentric adaptation. Indeed, by testing *memory-guided single-saccades* after adaptation of rightward scanning saccades, we found a main effect of phase ($p=0.026$) but no interaction with the *frame* versus *no frame* conditions. This indicated that leftward memory-guided single-saccades significantly decreased after adaptation, with or without the frame, in agreement with spatiotopic adaptation. A last experiment testing the effect of the same adaptation on *immediate single-saccades* revealed oculocentric adaptation. Taken together, these three experiments indicate that the same saccadic adaptation can involve allocentric, spatiotopic and oculocentric coordinates.

P1.125

The dynamics of the ongoing spontaneous neuronal activity in zebrafish larvae

Romano, S.A. (Paris)[1], Pietri, T. (Paris)[1], Sumbre, G. (Paris)[1]
[1]*Ecole Normale Supérieure, IBENS, Section de neurosciences, Paris, France*

In a state of sensory deprivation, sensory brain areas remain active. Once interpreted as irrelevant random noise, these activities have been found to exhibit coarse spatiotemporal structures,however, the biological significance of this ongoing neuronal activity still remains elusive.
To address this issue, we are currently monitoring brain activities of head-restrained zebrafish larvae, using two-photon imaging of genetically encoded calcium indicators (GCaMP3). This method allows recording the dynamics of large neuronal circuits, with single-cell resolution, from several brain areas of an intact behaving vertebrate. We are applying dimensionality reduction techniques and clustering analysis to describe circuit dynamics.
Our results demonstrate that the optic tectum, a brain region homolog to the mammalian superior colliculus and the largest visual center of the zebrafish, presents a precise spatiotemporal structure in its ongoing population activity and that this process seems to be driven by local recurrent circuitry. Moreover, these patterns resemble, at the single-cell level, functional visual maps, arguing for their biological significance.

P1.126

Neuropathic and cancer pains depend upon D-serine co-activation of spinal NMDA receptors

Amadio, A. (Bordeaux)[1], Lefèvre, Y. (Bordeaux)[1], Vincent, P. (Bordeaux)[1], Devoize, L. (Clermont Ferrand)[2], Dallel, R. (Clermont Ferrand)[2], Oliet, S.H.R. (Bordeaux)[1], Nagy, F. (Bordeaux)[1], Voisin, D.L. (Bordeaux)[1]
[1]*Université de Bordeaux, Neurocentre Magendie INSERM U862, Bordeaux, France,* [2]*Université d'Auvergne, INSERM U1107, Clermont Ferrand, France*

Chronic neuropathic pain relies upon spinal NMDA receptor activation and first results suggest this might be the case for cancer pain too. It has been shown that astroglia can regulate NMDA receptor activation by releasing the NMDA receptor co-agonist D-serine. The present study has investigated the role of NMDA receptor and D-serine in models of cancer and neuropathic pain.
The experiments followed the ethical guidelines of the International Association for the Study of Pain and the European Community Council directive (2010/63/EU). The neuropathic pain model was obtained by right L5-L6 spinal nerve ligation in male Wistar rats (sham rats: same procedure but with no ligation). Bone cancer pain was induced by injecting MRMT-1 rat mammary gland carcinoma cells

into the right tibia of Sprague-Dawley female rats (sham animals: injection with vehicle only). Mechanical allodynia and hyperalgesia were quantified using von Frey hairs. Drugs were administered either acutely or chronically using osmotic pumps, through intrathecal catheters chronically implanted in animals.
We found that:
1) intrathecal administration of a single dose (25 nmoles in 5 µL vehicle) of the NMDA receptor antagonist AP-5 significantly reduced mechanical allodynia and hyperalgesia at day 14 in neuropathic rats (n=10) and at day 21 in cancer rats (n=8);
2) chronic administration of fluoroacetate (5 nmoles/h for 14 days) partially reduced mechanical allodynia at day 14 in neuropathic rats (n=10) but had no effect in cancer rats at day 21 (n=6);
3) the effect of chronic fluoroacetate in neuropathic rats was partially reversed by acutely administered intrathecal D-serine (100 µg in 5 µL vehicle, n=9); intrathecal D-serine had no effect in cancer rats (n=5);
4) Intrathecal administration of a single dose of D-aminoacid oxidase, which degrades D-serine, reduced mechanical allodynia and hyperalgesia in neuropathic rats (n=8) and in cancer rats (n=10).
These results show that neuropathic pain and cancer pains depend upon D-serine co-activation of spinal NMDA receptors but only neuropathic pain requires functional spinal cord glia.

P1.127

Brain prediction of auditory emphasis by facial expressions during audiovisual continuous speech

Barone, P. (Toulouse)[1], Strelnikov, K. (Toulouse)[1], Foxton, J. (Toulouse)[1], Marx, M. (Toulouse)[2]
[1]CNRS CERCO UMR 5549, Toulouse, France, [2]Service d'Oto-Rhino-Laryngologie, Hopital Purpan, Toulouse, France

Background: Previous studies have shown that visual features related to word emphasis in a phrase can facilitate the discrimination of its auditory features. However, as visual stimuli are very close in time to the auditory information in the non-ecological studies of syllables and short words, the brain reaction to them usually overlaps with the auditory stimulation; the neural dynamics underlying visual facilitation in ecological speech remains unclear. The purpose of our study was to describe the temporo-spatial neural dynamics underlying this visual facilitation in continuous speech processing. Our hypothesis was that the visual cues would mimic an increase in amplitude/pitch changes that will be detected as such by the auditory system and expressed by a brain response similar to the auditory mismatch negativity response. We presented video clips with even continuous speech as frequent stimuli (standard) and with one word visually emphasized by the speaker as non-frequent stimuli (deviant).
Results: We performed a contrast analysis by comparing the EEG signal of the standard and deviant audiovisual conditions. Negativity for the deviant stimulation was detected in this contrast after the start of the emphasizing articulatory movements but before the auditory stimulus; no such negativity was present in the control visual-only condition. This negativity occurred about 200 ms before the beginning of the auditory deviance and is probably related to the prosodic visual gestures. The frame by frame analysis revealed that the negativity occurs immediately after the initiation of a large eyebrows lift in the emphasized video clip. The spatial dynamics was characterized by the posterior-anterior-posterior propagation of this negativity, which formed a loop between the visual and fronto-temporal regions.
Conclusions: The MMN-like activity that reaches the auditory regions suggests that the prosodic facial movements of the head and eyebrows act as a predictive representation of the forthcoming auditory inputs. Thus, in continuous ecological speech, the visual modality evokes predictive coding for the auditory speech, which is analysed by the brain in the context of the phrase even before the arrival of the corresponding auditory signal.

P1.128

Views to a place: posterior parietal and hippocampal activity in the rhesus monkey during a virtual navigation task

Wirth, S. (Bron-Lyon)[1], Planté, A. (Bron-Lyon)[1], Duhamel, J.-R. (Bron-Lyon)[1]
[1]*Centre de Neuroscience Cognitive, CNRS, Bron-Lyon, France*

Many goal-directed behaviours of everyday life require individuals to find their way towards specific locations. Spatial orientation and navigation require an internal model whereby important environmental landmarks are encoded relative to the subject or to each other. The contribution of the hippocampus (HPC) and the posterior parietal cortex (PPC) to the representation of space is well documented via imaging and lesion studies in human. Yet, neuronal activity underlying spatial representation has been so far only largely studied in the rodent. Here, we aimed at understanding how neurones in the PPC and the HPC support navigation in the closest experimental model to human, the macaque monkey. To this end, we trained two rhesus macaques to manoeuvre in a virtual reality environment with a joystick. Animals moved on the paths of a radial star maze, learning by trial and error, the location of a reward in one of the arms. Only distal landmarks outside the paths allowed locating the goal. We recorded 184 cells in the HPC and 130 cells in the PPC in two monkeys. More than half of the cells showed activity modulated by the task. These activities were more frequent at the starting and decisional points. In the HPC, only less than 10% of the cells exhibited pure encoding of the animal's position inside the virtual maze. These rare cells resembled typical place cells in rodents. The majority of cells showed activity in multiple positions that was explained by brief visual saccades to a meaningful landmark regardless of the position of the animal. The relevance of the landmark to navigation appeared more important then its physical properties. Indeed, when the goal position was changed after learning, cells exhibited new selectivities related to the newly learned goal position. In the PPC, cells coded egocentric positions relative to specific landmarks. As in the HPC, the meaning of landmarks appeared more important than their visual appearance. In summary, cells in the HPCs carried allocentric represention by signalling external landmarks irrespective of self-positions, and the PPC, cells carried a mixture of egocentric and allocentric representations by signalling self-position but relative to the landmark. Both activities would appear critical to navigation.

P1.129

Odor-evoked LFP oscillations in the piriform cortex: a code for odorant molecular features?

Litaudon, P. (Lyon)[1], Garcia, S. (Lyon)[1], Buonviso, N. (Lyon)[1]
[1]*Lyon Neuroscience Research Center, CNRS UMR5292, INSERM U1028, UCB Lyon 1, Team: Olfaction: from Coding to Memory, Lyon, France*

A central question for chemical senses is to understand the way odorant molecules are represented in the brain. It has been shown that the olfactory system recognizes some particular structural features of the molecules which are represented through a spatio-temporal pattern of activity in the olfactory bulb so that pairs of odorants with the smallest difference in molecular features evoked the most similar patterns. Several studies also demonstrated that odor stimuli could be represented through temporal features of brain activity such as oscillation of the local field potential (LFP) or spikes synchronization. Thus, we have shown that, at the olfactory bulb level, the code for odorant quality and intensity may be partially contained in the oscillations generated by the network. We wanted to test if such a code is retained in the piriform cortex (PC) which is the main area of projection of bulbar output cells.
We recorded both LFPs and spiking activity in the piriform cortex of urethane anesthetized rats in response to series of aliphatic odorants varying in their molecular features (carbon chain length and functional group). Recording have been performed both in anterior and posterior part of the PC in order to take into account the functional heterogeneity of this brain structure. PC activity was analyzed in term of oscillation frequency (beta, gamma), spike discharge, and temporal relationship with breathing. We also analyzed phase coupling between spikes and network oscillations.

First, our results showed that the probability of occurrence of beta and gamma oscillations differed between anterior and posterior part of the PC confirming that odor processing differed between the two PC subunits. Oscillatory activity in the anterior PC closely resembles OB oscillatory activity confirming the functional link between these two structures. Second, as previously demonstrated in the OB, the probability of occurrence of beta and gamma oscillations seemed to vary according to the functional group and the carbon chain length of the odorants. Analyses of PC cell activity are still in progress. Preliminary results showed that some odor coding mechanisms observed in the OB are partly retained in the anterior PC.

P1.130

Representation of egomotion in non-human primate

Cottereau, B.R. (Toulouse)[1], Fize, D. (Toulouse)[1], Trotter, Y. (Toulouse)[1], Smith, A.T. (London)[2], Durand, J.-B. (Toulouse)[1]
[1]CerCo UMR 5549, Toulouse, France, [2]Royal Holloway University, London, United Kingdom

Forward motion through the environment generates an expanding pattern of optic flow on the retina whose monitoring by the central nervous system is essential during navigation. Electrophysiological recordings have led to the general belief that in primate, visual area MST has a central role in this process. Recent imaging studies in human have however challenged this view by demonstrating that two other areas, in the parietal and cingulate cortex, are also strongly responsive to egomotion-compatible optic flow (Wall and Smith, 2008). The aim of this study is to determine the cortical networks that process egomotion-compatible optic flow in non-human primate. fMRI recordings were performed at 3T in one awake behaving macaque using surface coils positioned above the animal's head; MION contrast agent was used to enhance the recorded signal (Vanduffel et al, 2001). The experimental protocol was similar to the one used in a previous human study (Wall and Smith, 2008): we used a block design paradigm where the stimuli alternated between a baseline (i.e. fixation point only), a single patch of egomotion-compatible optic flow and an array of nine similar optic flow patches that was inconsistent with egomotion. Eye position was monitored while the monkey performed a fixation task and only the runs where the percentage of correct fixation was above 80% were processed. Data were analyzed using SPM8 and the cortical areas responding specifically to egomotion-compatible optical flow were obtained from the general linear model. Consistent with human data, significant activations ($p < 0.05$, corrected) were found in the cortical network known to be involved in complex motion processing, particularly in dorsal MST and in the parietal cortex. We also found activation in the posterior cingulate cortex in an area that might be a macaque homologue of the cingulate sulcus visual area (CSv). Altogether, our results suggest that human and monkey may share similar cortical networks to guide their navigation.

Bibliography:

- Wall MB, Smith AT (2008) The representation of egomotion in the human brain. Curr Biol 18: 191-194.
- Vanduffel W, Fize D et al (2001) Visual motion processing investigated using contrast agent-enhanced fMRI in awake behaving monkeys. Neuron 32(4): 565-77

P1.131

The endocannabinoid system controls food intake *via* olfactory processes

SoriaGomez, E. (Bordeaux)[1], Bellocchio, L. (Bordeaux)[2], Bendahmane, M. (Paris)[3], Reguero, L. (Leioa)[4], Ruehle, S. (Mainz)[5], Remmers, F. (Mainz)[5], Desprez, T. (Bordeaux)[2], Matias, I. (Bordeaux)[2], Wiesner, T. (Bordeaux)[2], Cannich, A. (Bordeaux)[1], Verrier, D. (Bordeaux)[2], Vincent, P. (Bordeaux)[2],

Massa, F. (Bordeaux)[1], Ferreira, G. (Bordeaux)[6], Lutz, B. (Mainz)[5], Guzman, M. (Madrid)[7], Gurden, H. (Paris)[3], Grandes, P. (Leioa)[4], Marsicano, G. (Bordeaux)[1]
[1]Université Bordeaux Segalen, Neurocentre Magendie - INSERM U.862, Endocannabinoïdes et Neuroadaptation, Bordeaux, France, [2]INSERM, Bordeaux, France, [3]CNRS UMR8165, Paris, France, [4]Faculty of Medicine and Dentistry, University of the Basque Country UPV/EHU, Department of Neurosciences, Leioa, Spain, [5]Institute of Physiological Chemistry, University Medical Center of the Johannes Gutenberg University, Mainz, Germany, [6]CNRS, Bordeaux, France, [7]Dept. of Biochemistry and Molecular Biology I, Sch. of Biology, Complutense Univ., Madrid, Spain

One of the clearest examples of how internal states regulate perception and behaviour is the property of hunger to link olfaction and food intake. However, the neuronal mechanisms governing this link remain poorly understood. Cortical feedback projections decrease neuronal activity in the olfactory bulb and regulate olfactory processing, but the physiological impact of these circuits are largely unknown. Here we show that cannabinoid type-1 (CB_1) receptors regulate food intake of fasted mice by controlling olfactory processing *via* inhibition of glutamatergic centrifugal feedback inputs onto granule cells of the main olfactory bulb (MOB). Selective genetic deletion and rescue of CB_1 receptors in glutamatergic neurons of the anterior olfactory cortical nucleus (AON) revealed that CB_1 receptor-dependent decrease of centrifugal glutamatergic transmission in the olfactory bulb is both necessary and sufficient to promote fasting-induced food intake. Specific pharmacogenetic stimulation of glutamatergic transmission from the AON to the MOB decreased food intake and reversed the hyperphagic effects of CB_1 receptor agonists. CB_1 receptor-dependent control of glutamatergic transmission in the main olfactory bulb determined the threshold of olfactory detection and olfactory habituation, eventually leading to increased food intake. *In vivo* electrophysiological recordings in the MOB revealed that activation of CB_1 receptors inhibits habituation of beta-type membrane potential oscillations evoked by repeated odour presentations. Altogether, these results show that cortical centrifugal feedback projections to the MOB are crucial in the regulation of food intake *via* CB_1 receptor signalling, which links the feeling of hunger to stronger and more persistent odour processing. Thus, CB_1 receptor-dependent control of cortical feedback projections in olfactory circuits couples internal states to perception and behaviour.

P1.132

Breaking the boundaries of somatosensory plasticity: improved touch at the fingers transfers to the lips

Muret, D.-A. (Bron)[1], Dinse, H.R. (Bochum)[2], Urquizar, C. (Bron)[1], Farnè, A. (Bron)[1], Reilly, K.T. (Bron)[1]
[1]Lyon Neuroscience Research Center, INSERM U1028, CNRS UMR5292, ImpAct Team, Bron, France, [2]Institute for Neuroinformatics, Neural Plasticity Laboratory, Ruhr-University, Bochum, Germany

As cortical plasticity is now well known to occur in the adult brain and to be involved in many brain functions, understanding the rules governing this phenomenon and its functional consequences is extremely important. A cutaneous coactivation (CA) protocol relying on the Hebbian postulate has been successfully used to study somatosensory plasticity in humans (see Godde et al, 2003). The aim of this study was to evaluate whether pure somatosensory plasticity, experimentally induced at the right index fingertip can cross somatotopically defined boundaries and functionally affect a physically distant but cortically adjacent body-part, the perioral lip region. A two-point discrimination (2PD) task was used to determine the spatial discrimination thresholds at the right and left index fingers (RD2 & LD2) and at the right and left sides of the upper lip area. This test was performed during 4 sessions on 2 consecutive days, with the CA applied for three hours at the RD2 fingertip between the third and fourth sessions. Thirty participants were randomly assigned to either a test group (n=15, mean age = 20.53 ± 2.26 years) in which subjects received a true CA, or a control group (n=15, mean age = 23.5 ± 3.25 years) in which the stimulator was fixed at the fingertip but turned off. In the test group three hours of CA led to a significant decrease of the 2PD threshold at RD2 fingertip of approximately 15%, thus replicating previous results. Critically, a significant 2PD threshold decrease was also found at

both sides of the upper lip area (on average 10% decrease). No effects were found at either the LD2 fingertip in the test group or at any of the four regions tested in the control group. These results demonstrate that experimentally-induced plasticity following RD2 coactivation improves tactile discrimination performance not only at the coactivated body site but also at a cortically adjacent body site. CA-induced somatosensory plasticity appears to cross somatotopically defined boundaries between body-part representations, spreading its functional consequences to cortically adjacent, but peripherally unstimulated body parts.
Godde, B. et al. (2003). Behavioral significance of input-dependent plasticity of human somatosensory cortex. *Neuroreport*, *14*(4), 543-6.

P1.133

Retention of oculomotor changes after adaptive lengthening of voluntary saccades

Habchi, O. (Lyon)[1], Salemme, R. (Lyon)[1], Urquizar, C. (Lyon)[1], Pelisson, D. (Lyon)[1]
[1]*Centre de recherches en neurosciences de Lyon-Univ Lyon 1, INSERM U1028 - CNRS UMR 5292, Impact, Lyon, France*

Adaptation of saccadic eye movements is one of the most studied model of sensori-motor adaptation. Several studies have investigated saccadic adaptation over the short-term, but the retention of adaptive oculomotor changes over days or weeks remains debated.
Here, we tested whether the amplitude lengthening adaptation of voluntary saccades (VS) can be retained over several days. We induced adaptation in seven human subjects, using a modified version (Panouillères et al. JEMR. 2012) of the target double-step paradigm (Mc Laughlin. Percept & Psychophys. 1967). Adaptation was induced progressively by displacing the target during horizontal VS and in the same direction (forward step of 30% in the first three blocks of trials and of 45% in the last three blocks); both leftward and rightward saccades were simultaneously adapted. Saccadic gain changes relative to the pre-adaptation baseline were calculated immediately after and at five different time points after adaptation: 5 min (day 0) and 1, 5, 11, and 19 days.
Results revealed significant increases of saccadic gain relative to baseline, reaching 15% and 16% immediately after adaptation of leftward and rightward VS, respectively. Further, the gain of leftward VS remained significantly elevated on days 0, 1, 5 and 11 (average saccadic gain change: 13%, 6.8%, 3.3% and 3.7%, respectively). In contrast, for rightward VS, the average change of gain remained significant on day 0 only (13 %) and decreased to a non significant 1.3% value on day 1.
These data reveal that adaptation after-effects persisted up to 11 days for leftward VS but had already disappeared after 1 day for rightward VS.
In agreement with a similar study of amplitude shortening adaptation of reactive saccades (RS) (Alahyane et al. Learn & Mem. 2005), these results suggest that saccade direction (leftward or rightward) interacts with the long term retention of adaptive oculomotor changes.

P1.134

Identification of the visuo-tactile convergence network: a functional Magnetic Resonance Imaging (fMRI) study in awake monkeys

Guipponi, O. (Bron)[1], Wardak, C. (Bron)[1], Pinède, S. (Bron)[1], Ben Hamed, S. (Bron)[1]
[1]*Centre de Neurosciences Cognitives, UMR 5229, Bron, France*

Multisensory integration is the process by which sensory information coming from different sensory channels is combined at the level of the single neuron. It is thought to improve the detection, localization, and discrimination of external stimuli and to produce faster reaction times. This process requires the convergence of inputs from different sensory modalities on the same neurons. In the non-

human primate, multisensory convergence has mostly been described from single cell recording or connectivity studies. Here, we use functional magnetic resonance imaging (fMRI) in behaving monkeys to provide a whole brain view of visuo-tactile (VT) convergence.
Visual and tactile modalities are tested in blocks in two monkeys while they maintain active fixation. Visual stimulations consist in 14° static pictures (see Guipponi et al., 2013). Tactile stimulations are directed to the center of the face thanks to airpuffs. Amongst the cortical areas activated both by visual and tactile stimulations, we identify parietal areas (PIP, VIP, 7op), prefrontal areas (46p, 6Vam, 8as, PMZ), lateral area Pi, cingulate area 24d and orbitofrontal areas (11 and 13). Interestingly, we also identify VT convergence in low-level visual areas (V1, V2d, V3, V3A), as well as in temporal area MST, consistent with what has been described in the human brain.
Two different visual and tactile response profiles are observed. The response of some areas (mainly the occipital and parietal areas) is significantly higher for the visual modality than for the tactile modality. The response of the remaining areas respond equally well to either modalities. A functional connectivity analyses allows us to propose a model of functional hierarchy within the identified cortical visuo-tactile convergence network.
A major prediction of this work is that VT integration will be observed in all of the identified cortical sites of VT convergence. This is addressed in a twin poster (Clery, Guipponi et al., *Société française des neurosciences*, Lyon, 2013).
Guipponi*, O., Wardak*, C., Ibarrola, D., Comte, J-C., Sappey-Marinier, D., Pinède, S., Ben Hamed, S. Multimodal convergence within the intraparietal sulcus of the macaque monkey. *J. Neurosci.* (in press).

P1.135

Categorization between rising vs. falling sweeps affects the functional properties and temporal precision of auditory cortex neurons after aversive and appetitive tasks

Gaucher, Q. (Orsay)[1], Huetz, C. (Orsay)[1], Tith, C. (Orsay)[1], Adenis, V. (Orsay)[1], Gourevitch, B. (Orsay)[1], Edeline, J.-M. (Orsay)[1]
[1]*Centre de Neuroscience Paris Sud, CNRS UMR 8195, Université Paris Sud, Orsay, France*

Recently, it has been claimed that the aversive or appetitive nature of the reward influences the type of changes that are detected in auditory cortex (ACx) during a learning task (David et al., PNAS, 2011). These data were obtained in behaving, animals during tasks where the animals' attention and strategies could be different in the aversive and appetitive conditions. To clarify this problem, we analyzed here the consequences of such training in anesthetized conditions when no differences in terms of attention and strategies are involved.
Rats were trained in a shuttle box to discriminate between rising (CS+ predicting a footshock) and falling FM sweeps (CS-) in an aversive task. After reaching 90% of correct responses, they were trained to generalize to 3 different sets of rising and falling sweeps. Performance decreased when a new pair of CS+/CS- was introduced but returned to 90% of correct responses for each pair. Water-deprived guinea pigs were trained to lick a spout, then to discriminate between a CS+ (ascending or descending sweep) and a CS- (ascending or descending sweep). Water was available during the CS+, not during the CS-. Once the animals have learned the initial discrimination, 3 other sets of rising and falling sweeps were introduced (same as for the rats). Both in rat and in guinea pig, spectro-temporal receptive fields (STRFs) of ACx neurons were tested under urethane anesthesia 48h after completion of training.
In rats, the STRFs of the trained animals were larger both in terms of bandwidth (2.8 vs. 1.9 octaves) and of duration (20 vs.11 ms) than in control animals. The response strength was also higher (51 vs. 29 spikes/sec). In guinea pigs, the STRFs of the trained animals were larger in terms of response duration (34 vs.29ms) but not in terms of bandwidth (2 vs. 2 octaves) than in control animals. The response strength was also higher (43 vs. 36 spikes/sec) in trained vs. untrained animals. These results suggest that when tested under anesthesia, the consequences of a categorization task are quite similar in aversive and appetitive conditions. Thus, the differences previously reported could be the consequences of different strategies or different attention load.

P1.136

Role of astrocytes connexin in the regulation of sleep oscillatory pattern

Lacroix, M. (Paris)[1], Roux, L. (New York)[2], Giaume, C. (Paris)[3], Benchenane, K. (Paris)[1]
[1]ESPCI - Paris Tech, Laboratoire de Neurobiologie CNRS UMR7637, Paris, France, [2]New York University, Medical School, Neuroscience, New York, United States, [3]College de France, Institute of Biology, Paris, France

Coordination across brain structures is thought to be crucial for the appropriate consolidation of memory trace. Cortical slow oscillations modulate the timing of hippocampal sharp wave-ripples (SPW-Rs) occurrence which support neuronal reactivations. Hippocampal SPWR-s are then followed sequentially by cortical reactivation, cortical delta waves and finally cortical spindles.
Our project aims at challenging the classic view of the “neurocentric” concept in brain rhythms regulation during sleep. Indeed, recent evidences showed that astrocytes can regulate cortical slow oscillations during sleep and are involved in brain processes related to memory deficits induced by sleep deprivation, both mechanisms being mediated by the activation of A1 adenosine receptors.
One of the key feature of astrocytes is their expression of connexins that form either hemichannels (leading to the release of neuroactive molecules) or gap junction (making astrocytes organized into networks of communicating cells).
We therefore investigated the role of connexins in the fine regulation and coordination between hippocampal, cortical and thalamic oscillations during natural sleep, by multi-site recordings in wild-type mice or transgenic mice double knock-out for astrocytic connexins Cx43 and Cx30 (dKO).
Our results shows that there is a massive decrease of slow oscillations during sleep in dKO mice in olfactory bulb, confirming the results obtained in vitro by Lisa Roux.
Our results support the involvment of astrocytes in regulation of neuronal network functioning and brain oscillatory activity.

P1.137

Investigation of sleep and dreaming using intracranial EEG

Moreno, Y. (Lyon)[1], Vidal, J.R. (Lyon)[1], Perrone, M. (Lyon)[1,2], Kahane, P. (Grenoble)[3], Ruby, P. (Lyon)[1], Lachaux, J.-P. (Lyon)[1]
[1]Lyon Neuroscience Research Center, CNRS UMR5292, INSERM U1028, UCB Lyon 1, DYCOG Team, Lyon, France, [2]Université Claude Bernard Lyon 1, Lyon, France, [3]CHU Grenoble and Department of Neurology, INSERM U704, F-38043, Grenoble, France

While great improvements have been made in the description of the default mode network (DMN) activity during wakefulness, very little is known about the DMN activity during sleep and especially during REM sleep. Recent studies, in StereoElectroEncephalography (SEEG) showed that the typical deactivation of the DMN during cognitive tasks usually described with fMRI could be also identified using broadband gamma power. SEEG technique thus appears as an alternative to fMRI for DMN investigation through the different sleep stages. Unlike the technique generally used, SEEG provides both high spatial and temporal resolution. This study aims at investigating the DMN activity during the different sleep stages as compared to executive brain regions (i.e. lateral prefrontal cortex). In addition, as some brain regions of the DMN are involved in dreaming (temporo-parietal junction, TPJ and medial prefrontal cortex, MPFC) we will also investigate the dream related activity in these regions. Rapid eye movement (REM) sleep and non REM (NREM) sleep are associated with quantitatively and qualitatively different dream reports. We will thus contrast the broadband gamma power in TPJ and MPFC between REM and NREM sleep. These data could participate to a better understanding of the dream report differences between NREM and REM sleep.

P1.138

Cochlear implantation in macaque monkey: surgical anatomy and radiological data with cone beam CT scan

Girard, P. (Toulouse)[1,2], Marx, M. (Toulouse)[1,3], Escudé, B. (Toulouse)[4], Barone, P. (Toulouse)[1], Fraysse, B. (Toulouse)[3], Deguine, O. (Toulouse)[1,3]
[1]CNRS CERCO UMR 5549, Toulouse, France, [2]INSERM, Toulouse, France, [3]CHU Purpan, Otology & Neurology Department, Toulouse, France, [4]Clinique Pasteur, Toulouse, France

Background: Large animal models of implantable hearing devices are needed to assess innovative technologies before using them in humans. The rhesus macaque monkey is the most widespread primate model for both electrophysiological and behavioral investigations because of its complex cognitive abilities. It has been used in the past but with non commercial implants or no detailed radiological descriptions of the surgical procedures.
Methods: We present detailed radiological data (CT scan and Cone beam computed tomography) from 7 heads of adult male rhesus macaque monkeys obtained from autopsy materials. Several comparative measurements were performed with 5 human temporal bones to emphasize similarities and differences between the macaque and the human inner ear. The radiological analyses helped planning the surgical approach for cochlear implant insertion in the macaque.
Results: We managed to perform one full (720°) and three partial insertions (190° to 330°) of cochlear implants (MedEl® Medium electrode array) in four rhesus macaque cochleae from different animals. All implantations were documented by cone beam computed tomography reconstructions. Radiological data confirmed that cochlear dimensions in the macaque are close to that observed in humans, hence modern cochlear implants could be used in the rhesus. However, we observed marked differences in the orientation of the external auditory canal and the basal turn that must be taken into account in the surgical approach. We suggest that the removal of the inferior wall of tympanal bone provides the optimal axis for electrode array insertion.
Conclusion: The rhesus macaque monkey is a valid and close-to-human animal model for unmodified cochlear implants insertion. Since this species is widely used in both behavioral and physiological studies, we expect that functional implants can be coupled with electrophysiological recordings to study the mechanisms of auditory compensation.

P1.139

How short can a vowel be and still be recognized by auditory cortex (ACx) neurons?

Occelli, F. (Orsay)[1], Suied, C. (Paris)[2], Pressnitzer, D. (Paris)[2], Edeline, J.-M. (Orsay)[1], Gourévitch, B. (Orsay)[1]
[1]Centre de Neurosciences Paris-Sud, CNRS UMR 8195, Orsay, France, [2]Ecole Normale Supérieure (ENS), Equipe audition, DEC, Paris, France

Vowels have been extensively used to investigate the neural coding of timbre, from the auditory nerve up to ACx (Bizley et al 2009). Here we recorded ACx neurons in anaesthetized guinea-pigs to a set of vowels of different durations to investigate whether the coding abilities of ACx neurons would be robust even in situations where potentially less spikes are emitted as well as fuzzier stimulus spectrum are available. Short segments of 5 vowels were selected with durations of 2, 4, 8, 16, 32, 64 or 128ms. These stimuli were used for a psychophysical experiment with human listeners. At 128 ms, human listeners performed the task almost perfectly. Performance decreased with duration but stayed above chance down to 4-ms duration. The same 5 vowels were presented (at 73dB SPL) while recording (with a 16-electrodes array) ACx neurons in urethane anesthetized guinea pigs. The tuning of each site was determined before presenting the vowels. Confusion matrices were computed for each

cortical site as well as for populations of neural responses. Mutual information was applied to confusion matrices to quantify the ability of neurons or populations to discriminate the vowels. In general, the discrimination ability of each cortical site was poor, lower than 2 vowels, even at 128ms. Actually, it was possible to approach performance from psychoacoustics based on the activity of populations of neural responses but never from that of a single cortical site. High frequency neurons were better than low frequency ones at discriminating vowels. This could result from the difference in sound energy between vowels, which was larger for higher harmonics than lower ones. The highest harmonics (~5-10 kHz) have a very short time period and therefore have a narrow spectrum even at a 2ms duration. Thus, high frequency neurons were able to identify some vowels ("E") even at 4ms. This result suggests that high frequencies neurons do not strongly influence perception when the task is to discriminate very short broadband sounds. Those trends were also broadly consistent with a simple distance measure between the spectrograms of vowels. This suggests that, down to very short duration, both cortical neurons and listeners still have access to a reasonable representation of the stimulus acoustics.

P1.140

Morphological development and electrophysiological properties of PKC gamma interneurons within the medullary dorsal horn

Antri, M. (Clermont Ferrand)[1], Chatila, N. (Clermont Ferrand)[1], Peirs, C. (Clermont Ferrand)[1], Artola, A. (Clermont Ferrand)[1], Monconduit, L. (Clermont Ferrand)[1], Dallel, R. (Clermont Ferrand)[1]
[1]*Inserm/UdA, U1107, Neuro-Dol, Trigeminal Pain and Migraine, Clermont Ferrand, France*

Protein kinase C gamma (PKCγ), which is concentrated in interneurons of the inner part of lamina II and lamina III of the medullary dorsal horn (MDH), plays a key role in pain and particularly allodynia by converting touch into pain. Nothing is known about the postnatal development of these interneurons and their electrophysiological characteristics in the MDH. The aim of this study was 1) to determine the spatial and temporal distribution of PKCγ immunoreactive neurons in the MDH of rat from postnatal day 3 (P3) to postnatal day 21 (P21) and 2) to investigate basic electrophysiological properties (resting membrane potential, input resistance, rheobase, and discharge patterns) at the mature stage. The recorded neurons were injected with biocytin for histochemical processing and morphological analysis. The results show that PKCγ interneurons appear at P6 in laminae II and III of the MDH. Their number increases very significantly during the following days with the appearance of the neuropil from P8/P9 in lamina II. In lamina III, their number decreases after P9. The distribution of the PKCγ interneurons was very similar for different rostro-caudal level (0,-1200, -2400 from the obex) and somatotopic regions of the MDH (ophtalmic division: V1, maxillary division: V2 and mandibular division: V3). The soma morphology and dendritic fields of PKCγ interneurons both increase in complexity with postnatal age and appear to be mature at about P21.
Whole-cell patch-clamp recordings from PKCγ interneurons were then performed in MDH slices of mature rat aged P28-P35. Half of the interneurons fired regularly, while the others adapted at all depolarization levels and fired with only 1-5 spikes at the beginning of the pulse. The majority of PKCγ interneurons had a hyperpolarization-induced inward rectification, whereas rebound excitation at the offset of hyperpolarization was seen in only few interneurons. All of the recorded PKCγ interneurons have cells bodies located in lamina IIi and dendrites arbors extending outside lamina IIi.

P1.141

Auditory efferent feedback, speech perception in continuous and fluctuating noise maskers and hemispheric specialization

Moulin, A. (Lyon)[1], Garcia, S. (Lyon)[1], Andeol, G. (Paris)[2]

[1]*Centre de Recherches en Neurosciences de Lyon, Inserm U1028 - CNRS UMR5292, Lyon, France,*
[2]*IRBA Institut de Recherche Biomédiale des Armées, Paris, France*

The mammalian auditory system contains descending neural pathways, some of which project onto the cochlea amplifier via the medial olivocochlear system (MOCS). By dampening cochlear amplification, the MOCS enhances signal to noise ratios. Hence, the strength of the MOCS reflex, as measured, in humans, by contralateral acoustic suppression of outer hair cell functioning, (i.e. suppression of otoacoustic emissions amplitude), has been related to abilities in speech perception and sound localization in noise. This study sought to investigate if the “listening in the dips” abilities, i.e. ipsilateral unmasking by modulated maskers, were related to MOCS functioning in humans. In twenty-five normally-hearing right-handed young subjects, both MOCS efficiency and speech recognition scores (SRC) in fluctuating and continuous ipsilateral maskers were investigated (study approved by the local ethics committee, RCB 2011-A01233-38).
Subjects performed a 4 alternative forced choice task consisting in recognizing one word amongst 4 alternatives differing by one phoneme, in 3 different types of continuous background noises, all equal in rms amplitude and spectra: a flat noise (FN), a 32 Hz modulated noise (Mod. N) and a cocktail party noise (CPN), at 4 different signal-to-noise (SNR) ratios.
No significant differences were obtained between the ears concerning speech perception in the 3 different types of noises. No significant correlation were obtained between left ear (LE) and right ear (RE) speech perception performance (measured as the SNR giving a score of 50 rau) in the FN, however ears performances were significantly linked for the Mod. N and CPN ($r=0.55$, $p< 0.005$).
MOCS reflex strength increased significantly with speech perception abilities in mod. N mostly for the LE ($r=0.48$, $p< 0.02$) and in CPN only for the RE ($r=0.41$, $p< 0.05$), with a significant difference with the LE (Z score=-1.91, $p=0.05$). Moreover, MOCS strength increased significantly as masking release increased in the LE only ($r=0.67$, $p< 0.001$).
Relationship between speech in noise perception and efferent reflex strength depends both on the ear considered (left/right) and the level of processing, being significantly stronger for tasks involving higher processing.

P1.142

The role of the subthalamic nucleus in selecting actions that produce the most desirable outcome

Apicella, P. (Marseille)[1], Espinosa-Parrilla, J.-F. (Marseille)[1], Baunez, C. (Marseille)[1]
[1]*Institut de Neurosciences de la Timone, Marseille, France*

The subthalamic nucleus (STN) is a key basal ganglia structure involved in motor control and motivation. There is evidence supporting the idea that this nucleus may be well positioned to mediate action selection. However, the properties of STN neurons with respect to selection among competing motor alternatives remain to be clarified. To address this issue, we recorded single STN neurons in two monkeys while they performed a free choice task in which they chose between two actions based on reward preference. In each trial, two colored visual stimuli predicting juice or water rewards were presented simultaneously on the left and right sides of a panel, spatial locations and reward types alternating randomly from trial-to-trial. After a fixed delay, two target stimuli appeared on the same two locations and monkeys were required to select one of the two possible target responses. The animal´s choices in favor of juice provided an operational measure of the value that the monkey assigned to the reward available at each target. We identified 59 neurons that significantly changed their spiking activity at the moment of action, as the monkey chose the target associated with the most desirable outcome (juice). Most of these neurons also showed changes in their firing rate during a postreward period. Using additional control procedures in order to better differentiate changes in neuronal activity along visuomotor and cognitive domains, we found that neuronal activity changes were not markedly dependent on whether the preferred reward was offered on the left or on the right. Also, the perimovement changes in STN activity were not influenced by choice accuracy. On the other hand, in sessions in which monkeys were wrong with the choice of selecting the preferred outcome (less than 70% of juice trials per block), neuronal activity during the postreward period was greater in water trials

than in juice trials. These results suggest that the STN activity is influenced by both the need to choose between actions that led to a preferred reward and the consequences of the chosen actions. The observed changes in STN activity could provide signals useful for selecting actions that are guided by expected reward outcomes.

P1.143

How does learning modulate the sensorimotor control of the sniff?

Lefevre, L. (Lyon)[1], Courtiol, E. (Lyon)[1], Buonviso, N. (Lyon)[1]
[1]*INSERM U1028 - CNRS UMR5292 - CRNL, Centre de Recherche en Neuroscience de Lyon, Equipe Codage et Mémoire Olfactive, Lyon, France*

Sensory function depends on a combination of feedforward flow of information from sensory organs to the brain and the ensuing readjustment of sense organs by feedback from the brain. As eye movements actively sample a visual scene to allow acute visual perception, the active sampling process in olfaction is sniffing. Sniffing is highly dynamic (Youngentob et al., 1987): it is rapidly modulated in frequency and flow rate. While these modulations strongly impact neural activity (Courtiol et al., 2011a, b), their functional significance is still not well understood. As first suggested by Schoenfeld and Cleland (2005), sniffing variations would improve olfactory capabilities by optimizing the deposition of odor molecules through the olfactory epithelium. Our aim here is to test this hypothesis and gain an insight into the contribution of sniffing variations in olfactory perception.
In a first series of experiments, rats were trained to discriminate odors in a double choice discrimination task while their sniffing activity was accurately recorded using a whole-body plethysmograph. We showed that sniffing parameters were adjusted 1) quickly and 2) relative to odorant chemical properties, revealing a true olfactomotor control of the sniff. The next question is to know if those adjustments are just perceptual effects or if they could be related to learning. In this latter case, sniffing variations would be correlated to a discrimination improvement. A new series of experiments are in progress to answer this question.
Courtiol et al. (2011a) PLoS One, 6(1):e16445.
Courtiol et al. (2011b) J Neurophysiol. 106:2813-24.
Schoenfeld & Cleland (2005) TINS, 28:620-7.
Youngentob et al., (1987) Physiol Behav. 41:59-69.

P1.144

Inactivation of B3 serotonergic neurons by RNA interference: effects on the descending control of nociception in the rat

Gautier, A. (Paris)[1], Salam, S. (Paris)[2], Kayser, V. (Paris)[1], Hamon, M. (Paris)[1], Vodjdani, G. (Paris)[2], Bernard, J.-F. (Paris)[1]
[1]*INSERM U894 CPN, UPMC, Paris, France,* [2]*CRICM, UPMC/CNRS UMR7225/INSERM UMRS975, Paris, France*

Pain is a multifactorial phenomenon involving not only numerous ascending pathways but also descending pathways, which modulate the transmission of nociceptive messages at the level of the spinal cord. Although it is generally accepted that bulbar serotonergic neurons of the B3 group play a critical role in the descending control of nociception, so far the tools used were not specific enough to assess unambiguously the role of serotonin in these controls. The aim of this study was to reappraise this question using RNA interference (RNAi) to selectivity inhibit serotonin synthesis in B3 serotonergic neurons in the rat.

Adult male Sprague-Dawley rats were stereotaxically injected into the B3 group, with a recombinant lentiviral vector, which produces siRNA interfering with the expression of Tryptophan Hydroxylase 2 (TPH2). Control animals were injected with a paired lentiviral vector inactive on TPH2. Nociceptive responses to mechanical and thermal noxious stimuli were assessed using validated behavioral tests and spinal c-Fos immunolabeling was quantified in the spinal dorsal horn in both groups of rats. The localization of injected virus and the TPH2 inhibition were immunohistochemically controlled in every animal.
RNAi - induced blockade of TPH2 expression and the resulting serotonin depletion were almost complete (80 - 90% range) in the whole rostro-caudal extent of B3 group. However no significant changes in mechanical and thermal nociceptive thresholds were noted in serotonin depleted rats compared to controls injected with the inactive vector. In contrast, c-Fos immunolabeling revealed that high temperature (50°C) nociceptive stimulation activated a larger number of neurons ($p < 0.01$) in the laminae I and II of the spinal dorsal horn in rats whose B3 group had been depleted in serotonin (59.9 ± 2.5, n = 12) compared with control rats (48.7 ± 2.0, n = 7). This difference was not observed at 46°C nociceptive stimulation.
These results suggest that serotonergic neurons of the B3 group do not exert any modulatory influence upon spinal nociceptive neurons under moderate noxious conditions but contribute to a descending inhibitory control when the latter neurons are activated by a strong nociceptive stimulation (50°C).

P1.145

Chronic cervical high injury and delayed respiratory diaphragm rehabilitation using post-traumatic nerve bridging in the rat. A potential clinical application for ventilator dependant spinal cord injured patients

Gauthier, P. (Marseille)[1], Matarazzo, V. (Marseille)[2], Tadié, M. (Paris)[3], Felix, M.-S. (Marseille)[4]
[1]Université Aix-Marseille, CNRS UMR 7260 Laboratoire LNIA, Pôle 3C, Marseille, France, [2]Université Aix-Marseille, INMED INSERM UMR901, Marseille, France, [3]Hopital de Bicêtre, Service de Neurochirurgie, Paris, France, [4]Université Aix-Marseille, PSNM, Marseille, France

Cervical spinal cord injury still has a devastating impact on the respiratory system, leading to acute and chronic respiratory insufficiency which mainly results from diaphragm paralysis due to interruption of the descending respiratory pathways commanding the phrenic motoneurons or to direct injury of those motoneurons whose axons constitute the phrenic nerve (PN) commanding the diaphragm. Respiratory complications are frequent after cervical SCI and contribute significantly to associated morbidity, mortality and economic burden.
Although electrical stimulation of the phrenic nerve or the diaphragm remains the current treatment for ventilator dependent patients, this method is still associated with side effects and high costs and does not allow optimum physiological control of respiration. In this context, repair strategies that may result in respiratory functional recovery or improvement after high spinal cord injury are therefore still required.
A potential tissular therapy for reinnervating the diaphragm consists in nerve bridging between laryngeal recurrent nerve (LRN) and phrenic nerve. The LRN expresses a spontaneous respiratory activity in phase with the phrenic nerve but doesn't emerge from the spinal cord, thus making it safe after spinal cord injury. The rationale is thus to reinnervate one hemidiaphragm by laryngeal respiratory fibers.
Recurrent-phrenic nerve anastomosis has already been proven to work in healthy animals or after acute SCI (Gauthier et al., 2006) but was never tested in a post-traumatic circumstance. Hence, the original feature of this report concerns the efficiency of the strategy after chronic high spinal cord injury in the rat.
We show that the nerve bridging strategy can induce a dramatic diaphram rehabilitation in the rat, even when the strategy is applied as long as one month after a chronic unilateral C2 spinal cord injury. In non treated animals, the injury induces a long-term persistent ipsilateral hemi-diaphragm paralysis whereas in transplanted animals, an ipsilateral diaphragm recovery starts at 2 months to be achieved at 5 months post-bridging. The present study confims that diaphragm rehabilitation originates from laryngeal re-innervation and illustrates the time course and amount of diaphragm recovery.

P1.146

Human Acid-Sensing Ion Channel 3 dynamically adapts its activity to sense the extracellular pH in both acidic and alkaline directions

Lingueglia, E. (Valbonne)[1,2], Delaunay, A. (Valbonne)[1,2], Gasull, X. (Barcelona)[3], Salinas, M. (Valbonne)[1,2], Noël, J. (Valbonne)[1,2], Friend, V. (Valbonne)[1,2], Deval, E. (Valbonne)[1,2]
[1]Institut de Pharmacologie Moléculaire et Cellulaire, CNRS/Université de Nice-Sophia Antipolis UMR 7275, Valbonne, France, [2]LabEx Ion Channel Science and Therapeutics, Valbonne, France, [3]Facultat de Medicina, Universitat de Barcelona, Neurophysiology Laboratory, Barcelona, Spain

Aim of investigation: Acid-Sensing Ion Channels (ASICs) are depolarizing cation channels gated by extracellular acidosis. These channels are expressed in both the central and peripheral nervous systems, including nociceptors. Different ASIC subunits have been identified in rodents (ASIC1a, ASIC1b, ASIC2a, ASIC2b, ASIC3 and ASIC4), and recent studies have demonstrated the important role of ASIC3 in inflammatory and post-operative pain in rat. Comparatively, little is known about the human ASIC3 channel, which comprises three splice variants (hASIC3a, hASIC3b and hASIC3c) differing in their C-terminal domain.
Methods: We have combined molecular biology approaches (quantitative PCR and site-directed mutagenesis) and electrophysiology approaches (patch-clamp recordings in cell lines and primary-cultured neurons, two-electrode voltage-clamp recordings in *Xenopus* oocytes) to assess the detailed tissue distribution and biophysical properties of the human ASIC3 channels.
Results: The hASIC3a mRNA is the most abundant ASIC3 transcript in both human peripheral and central nervous systems (sensory neurons, dorsal spinal cord and brain). Interestingly, we show that human ASIC3 channels are not only opened by acidification (up to pH 5.0), but also by alkalization (up to pH 8.0), a property never described for ASICs. The channel displays two functioning modes: (i) a transient mode in response to extracellular acidification, and (ii) a sustained mode in response to extracellular alkalization. Both modes lead to membrane depolarization that might increase neuronal excitability. The alkaline sensitivity requires two arginine residues localized in the extracellular loop of human, but not rodent, ASIC3. Interestingly, the alkaline sensitivity is not always present and can be induced and/or potentiated by arachidonic acid.
Conclusions: Human ASIC3 channels are thus able to sense the extracellular pH in both directions and therefore to dynamically adapt their activity between pH 5.0 and pH 8.0. This property likely participates to the fine tuning of neuronal membrane potential and to neuron sensitization in various pH environments and under different physiological and pathophysiological conditions including pain.
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P1.147

Olfactory bulb and glucose sensing

Julliard, A.K. (Lyon)[1], Al Koborssy, D. (Lyon)[1,2], Salem, R. (Lyon)[1], Romestaing, C. (Villeurbanne)[3], Paulignan-Palouzier, B. (Lyon)[1]
[1]Centre de Recherche en Neurosciences de Lyon (CRNL). Inserm U1028-CNRS 5292- UCBL1, Team-Olfaction: From Coding to Memory, Lyon, France, [2]The Florida State University, Department of Biological Science, Program in Neuroscience and Molecular Biophysics, Thallahassee, United States, [3]Laboratoire d'Ecologie des Hydrosystèmes Naturels et Anthropisés (LEHNA) CNRS 5023 - UCBL1, Villeurbanne, France

Brain derives its energy source almost exclusively from glucose which extra cellular fluid (ECF) levels vary within brain regions according to the neural activity. In the brain, glucose homeostasis is crucial to the point that systems specifically dedicated to glucose-sensing are found in areas involved in energy regulation and feeding behavior. Olfaction is one of the major sensory modalities that regulate food consumption and conversely, the nutritional status modulates olfactory detection. An increasing body

of evidence indicates that this interplay between olfaction and food intake is mediated by a direct action of endocrine and metabolic molecules on the olfactory system specifically on the olfactory bulb (OB) which is a target for hormones related to metabolism and food-intake regulation. In the OB, neuronal glucose consumption is high in order to supply the high energetic demands and what could be considered as a critical signal to integrate metabolic cues. Sodium-coupled glucose transporters (SGLTs) and glucose transporters (GLUTs) are proposed to be markers of neuronal glucose-sensitivity. Since GLTU3 and GLUT4 have been described in the OB this strongly suggests that some OB neurons could be glucose-sensitive in the same way as hypothalamic neurons. In order to test this hypothesis, we first measured ECF glucose concentration ($[Gluc]_{ECF}$) in anesthetized rats, in the OB and the cortex (Ctx) simultaneously using glucosensors. We showed that $[Gluc]_{ECF}$ in the OB is higher than $[Gluc]_{ECF}$ in the Ctx and that it follows glycemia. Using immunostainning, we demonstrated that GLUT4 is strongly expressed in some mitral cells (MCs) and glomeruli (GI). The SGLT1 is also present in some GI and specifically in the inner part of the EPL. We quantified GLUT4 and showed a preferential distribution in the rostral and caudal parts of the OB. We showed that the feeding state induces changes in the mapping of the GI GLUT4 staining and in OB GLUT4 expression (evaluated by Western blot). Finally, in vitro electrophysiological recordings showed that MCs firing changed according to the ECF glucose content.
In conclusion, these data emphasize the importance of glucose for the OB network and provide the first arguments towards establishing the OB as a glucose sensing organ.

P1.148

Hippocampal temporal coding during backward movement

Cei, A. (Paris)[1], Girardeau, G. (Paris)[2], El Kanbi, K. (Paris)[2], Wiener, S. (Paris)[2], Dupras, Y. (Paris)[2], Rateau, S. (Paris)[2], Zugaro, M. (Paris)[2]
[1]Collège de France, Umr 7152 Physiologie de la Perception et Action, Paris, France, [2]Umr 7152 Physiologie de la Perception et Action, Paris, France

When a rat explores and environnement, hippocampal pyramidal cells called place cells, fire specifically in one or few places (the cell´s place fields). In parallel, the local field potential oscillates at theta frequencies (between 8-12 Hz). Interestingly, place cell fires ealier and earlier compare to theta while the rat advances in the place field. This phenomenon called phase precession could constitute a temporal code of the spatial information. In spite, when we consider few place fields that are successively crosses by the rat, phase precession allows the corresponding place cells to fire sequentially in the same order than their place fields within each theta cycle, ie in a time window compatible with plasticity. Phase precession could so plays an instrumental role in the memorization of spatio-temporal sequences.
To explain the neurophysiological mecanisms of phase precession different computationnal models have been proposed : dual oscillators models, somato-dendritic interference models or models based on the network properties. One way to discriminate between these different models is to perturbate the para-hippocampal system and to compare the data with models´ predictions.
We recorded hippocampal neurons when animals were moved backward. In this context, the relationship between place and theta phase was reversed compared to forward displacement. However the theta cycle firing phase remains the same in time. These results are not compatible with all models of phase precession and will give new future orientation for exploring their neurophysiological mecanims. Besides if the firing kept precessing relative to time regardless of the direction of movement, that means that phase precession is an endogenous mecanism more than a phenomenon controled by sensory inputs. This supports the view that phase precession is crucial in the formation and memorization of spatio-temporal sequences. These sequences could correspond in humans to episodic memory.

P1.149

Anticipation during auditory sequence processing: a human electrophysiology experiment on the role of temporal structure

Azzi, J. (Lyon)[1,2], Varnet, L. (Lyon)[2], Schwartze, M. (Leipzig)[3], Wilson, C. (Lyon)[1], Dominey, P.F. (Lyon)[1], Kotz, S.A. (Leipzig)[3], Procyk, E. (Lyon)[1], Hoen, M. (Lyon)[2]
[1]Inserm U846, Stem Cell and Brain Research Institute, Lyon, France, [2]Lyon Neuroscience Research Center, INSERM U1028 - CNRS UMR5292 - Université Claude Bernard Lyon, Lyon, France, [3]Max Planck Institute for Human Cognitive and Brain Sciences, Neuropsychology Department, Leipzig, Germany

A still unresolved issue in the fields of cognitive neuroscience of language is how the unique ability to process syntactic structural rules emerged in humans. According to the Functional Equivalence Hypothesis (FEH) (e.g. Dominey, Inui & Hoen, 2009), grammatical processing evolved from rule-based anticipatory mechanisms originally underpinning non-linguistic sequence processing. The FEH also states that the neural substrate of these structural rule-governed mechanisms that emerged from sensory, sensory-motor to linguistic sequence processing are embodied in fronto-striatal loops. Furthermore, this brain system in its capacity to tag temporal attributes (e.g. Kotz et al., 2009) may be responsible for the synchronization of predictive temporal and sequential cues. In 2012, we started a collaborative project involving French and German research teams (ORIGRAM, ANR/DFG funded). The project aims at comparing electrophysiological correlates of anticipatory mechanisms involved in auditory sequence processing in both human and non-human primates. In the current communication, we will present data from a first series of experiments in which we adapted a mismatch negativity (MMN) paradigm developed by Schwartze et al. (2011), to study the sensitivity of human participants to rhythmic regularities in auditory sequences. In the original paper the authors used Isochronous (predictable rhythmic structures) and random (non-predictable) auditory sequences and showed that in an attentive session, the P3b component, following the MMN, was smaller for deviant tones embedded in irregular temporal structure compared to regular temporal structures. In the current version of the paradigm, we added one new type of deviant in temporally predictable sequences: an omission (no tone) and we ran time-frequency analyses in order to identify the oscillatory correlates of the missing tone that was predicted from the temporal structure. Preliminary data from this study suggest that brain entrainment to temporal regularities is reflected in modulations of power in the alpha frequency band. These data will be discussed in the context of recent observations suggesting a role for alpha oscillations as pulsatile inhibition of ongoing cortical activity and in the general framework of the FEH.

P1.150

Changes in spinal inhibitory networks induced by furosemide in humans

Klomjai, W. (Paris)[1,2], Lackmy-Vallée, A. (Paris)[1], Roche, N. (Garches)[3,4], Bensmail, D. (Garches)[4], Katz, R. (Paris)[1,5]
[1]ER6 - UPMC,Service de Médecine Physique et Réadaptation, Groupe Hospitalier Pitié Salpétrière, Paris, France, [2]Faculty of Physical Therapy, Mahidol University, Bangkok, Thailand, [3]Univ. Versailles-Saint-Quentin, EA 4497, Garches, France, [4]APHP Service de Médecine Physique et Réadaptation, Hôpital Raymond Poincaré, Garches, France, [5]APHP Service de Médicine Physique et Réadaptation, Hôpital Pitié-Salpêtrière, Paris, France

It has been shown in animals that during neural development, GABAergic and glycinergic neurons are first excitatory, and then become inhibitory during maturation. This developmental transition is generated by the differential expression of cation-chloride transporters. The chloride inward co-transporter (NKCC1) is highly expressed in immature neurons, while the chloride outward co-transporter (KCC2) is dominant in mature neurons. This results in lower intracellular chloride, inducing neuronal inhibition response to GABA and Glycine. Spinal cord transection in animals showed a down-regulation of co-transporter KCC2, which reversed the inhibitory pattern of GABAergic and glycinergic neurons back towards their immature and excitatory state. This may contribute to hyper-excitability of

spinal reflexes and reduction of synaptic inhibition. The reversal of mature inhibitory synapses may contribute to the reflex hyper-excitability after SCI in humans. In this study, we demonstrated the effects of furosemide, a cation-chloride co-transporter antagonist on spinal networks excitability in healthy subjects. Electrophysiological recordings of the Hoffman reflex were used to assess pre- and post-synaptic inhibitions of the spinal cord. Subjects were given 40 mg furosemide by oral administration during recording. Our findings showed that furosemide, at doses commonly used in clinical medicine, significantly reduced inhibitory synapse excitability. This reduction was also dose-dependent. A control study of inhibitory synaptic excitability without drug administration was also performed, allowing exclusion that this reduction could be due to spontaneous variations during the recording time (70 minutes). The results with furosemide provide indirect evidence that the cation-chloride transport system may play a role in modulating of inhibitory synapse excitability in humans. The purpose of this study was to explore the use of furosemide administration as a first step towards future studies in paraplegic patients exploring the hypothesis that SCI in humans reverses the inhibitory synapses back towards the immature state.

P1.151

Anodal transcranial direct current stimulation of the motor cortex induces opposite modulations of reciprocal inhibition in wrist extensor and flexor muscles

Lackmy-Vallée, A. (Paris)[1], Klomjai, W. (Paris)[1], Bussel, B. (Garches)[2], Katz, R. (Paris)[1], Roche, N. (Garches)[3]
[1]Univ. Pierre et Marie Curie Paris 6, Er6-UPMC, Hôpital de la Pitié Salpétrière, service de Medecine Physique et Réadaptation, Paris, France, [2]Univ. Versailles-Saint-Quentin, Hôpital Raymond-Poincaré, service de Médecine Physique et Réadaptation, Garches, France, [3]Univ. Versailles-Saint-Quentin, EA 4497, Hôpital Raymond-Poincaré, Service de Médecine Physique et Réadaptation, Garches, France

Development of non-invasive brain stimulation techniques in humans allowed to change cortex excitability and thus to study physiological and physio-pathological mechanisms of cortical plasticity. Recently, it was shown that transcranial direct current stimulation (tDCS) applied over the motor cortex can also affect neural networks in the spinal cord. This opens the way to use tDCS to explore cortical descending control onto spinal neural networks. In the present study, tDCS (in anodal polarity) was applied over the hand motor area so as to explore effects of increased motor cortex excitability onto reciprocal inhibition pathways innervating wrist flexors and wrist extensors. For the first time, tDCS effects were tested in parallel on reciprocal inhibition directed from wrist flexors to wrist extensors and in reverse situation i.e reciprocal inhibition directed from wrist extensors to wrist flexors. We showed that modulations of reciprocal inhibition between flexors and extensors during anodal tDCS application are opposite: tDCS depresses reciprocal inhibition directed from flexors to extensors but increases reciprocal inhibition from extensors to flexors. This asymmetry but to a lesser extent was also observed when ipsilateral motor cortex was stimulated by tDCS. Our results suggest that reciprocal inhibition between flexors and extensors at wrist level is not symmetrical. The functional significance is that reciprocal inhibition is arranged to favor the extensor contraction which is required in a large sample of wrist and finger movements in humans. We will study the effect of tDCS in patients with central nervous system lesion to explore if anodal tDCS applied over the hand motor cortex area may be used as a tool to favor extensor contraction.

P1.152

Differential processing of sensory input by neighbouring layer 2 pyramidal neurons in whisker barrel cortex revealed by immediate-early-gene expression

Jouhanneau, J.-S. (Berlin-Buch)[1,2], Ferrarese, L. (Berlin-Buch)[1,2], Brecht, M. (Berlin)[3], Barth, A.L. (Pittsburgh)[4], Poulet, J.F.A. (Berlin-Buch)[1,2]
[1]Department of Neuroscience, Max-Delbrück Center for Molecular Medicine (MDC), Berlin-Buch, Germany, [2]NeuroCure, Cluster of Excellence, Neuroscience Research Center, Charité-Universitätsmedizin Berlin, Berlin, Germany, [3]Bernstein Center for Computational Neuroscience, Humboldt University of Berlin, Berlin, Germany, [4]Department of Biological Sciences and Center for the Neural Basis of Cognition Carnegie Mellon University, Pittsburgh, United States

Neocortical pyramidal neurons display considerable heterogeneity in spontaneous and sensory evoked firing rates. The underlying circuit and synaptic mechanisms that leads to sensory response diversity have not been determined, in part because of the lack of molecular markers for pyramidal neuron subpopulations. Recently, we used expression of the activity-dependent immediate-early gene c-fos to identify a more active subnetwork of layer 2 pyramidal neurons in whisker primary somatosensory cortex using a fosGFP transgenic mouse. Here we investigate the spontaneous activity and sensory response properties of fosGFP+ve and fosGFP-ve layer 2 pyramidal neurons using in vivo dual two-photon targeted whole-cell recordings in the urethane anaesthetized mouse. We show that fosGFP+ve neurons receive larger amplitude depolarising synaptic input as compared to neighboring (<100µm) fosGFP-ve neurons both during spontaneous upstates and airpuff triggered sensory responses. Surprisingly, analysis of the subthreshold sensory response shows that fosGFP+ve neurons also respond with a significantly shorter latency. Further investigation has demonstrated that fosGFP+ve neurons have a larger receptive field integrating sensory input from wider range of input within the barrel filed. The postero-medial nucleus of the thalamus (POm) is known to be activated by multiwhisker stimulation and his projection target the entire barrel cortex field. We used optogenetic stimulation to activate specifically POm and found that fosGFP+ve neurons receive a stronger and shorter latency input. Thus, a subpopulation of more active layer 2 pyramidal neurons expresses the immediate early gene c-fos and integrate sensory input from a wider receptive field.

P1.153

Localization of glycinergic neurons selectively activated during paradoxical sleep in rats: potential role in the control of muscle atonia

Valencia Garcia, S. (Lyon)[1], Clément, O. (Lyon)[1], Libourel, P.A. (Lyon)[1], Arthaud, S. (Lyon)[1], Charrie, P. (Lyon)[1], Luppi, P.-H. (Lyon)[1], Fort, P. (Lyon)[1]
[1]Centre de Recherche en Neurosciences de Lyon INSERM U1028 - CNRS U5292 - Université Claude Bernard, Lyon, France

During paradoxical sleep (PS), somatic motoneurons are inhibited causing a tone loss in the whole skeletal musculature, a motor event characteristic of this sleep state. As revealed by intracellular recordings of masseter and lumbar motoneurons in cats, glycine is the main inhibitory neurotransmitter involved in their PS-specific hyperpolarization. In our current functional model, glutamatergic neurons of the pontine sublaterodorsal nucleus (SLD) are responsible for the genesis of PS atonia by means of their excitatory projections to glycinergic premotoneurons. However, the exact location within the brainstem and/or spinal cord of the glycinergic premotoneurons selectively activated during PS remains a matter of debate. To bridge this gap, we used 4 groups of rats:
1) control (PSC);
2) PS deprived during 72h (PSD, n=4);
3) allowed to recover for 150 minutes of such deprivation during which they experienced 40% of PS (PSR, n=4); and
4) forced to walk for 2h (STEP, n=4).
PSR rats were previously injected with retrograde tracer Fluorogold (FG) into the ventral T13-L1 spinal cord. Brainstem and spinal cord sections were then processed for double labeling combining Fos immunohistochemistry (a marker of neuronal activity) with non-radioactive *in situ* hybridization of GlyT2 mRNA (the neuronal glycine reuptake transporter). Sections from PSR rats were also submitted to Fos/FG double immunostaining.

Within the brainstem, Fos+/GlyT2+ neurons were quite exclusively observed in PSR rats, with the highest number of double-labeled cells detected within the ventral gigantocellular (GiV) and lateral paragigantocellular (LPGi) medullary nuclei. In both areas, more than 80% of Fos+ neurons also expressed GlyT2. Within the lumbar cord, occasional Fos+/GlyT2+ neurons were seen in PSC, PSD and PSR rats while they were numerous in STEP rats with a sustained locomotor activity prior to sacrifice. Finally, Fos/FG double immunostaining in PSR rats unravel that 45% of Fos+ neurons in GiV and 5% in SLD project to the ventral spinal cord.
Our functional data clearly indicate that the glycinergic neurons of GiV, selectively activated during PS and projecting to the spinal cord, are responsible for the PS-specific hyperpolarization of spinal somatic motoneurons, causing muscle atonia.

P1.154

Selective anticipation and processing of visual cues in motor cortex

Kilavik, B.E. (Marseille)[1], Confais, J. (Marseille)[1], Riehle, A. (Marseille)[1]
[1]*CNRS, Institut de Neurosciences de la Timone (INT), Marseille, France*

Motor cortex holds an established role in movement preparation and execution. But, it also exhibits short-latency (~100ms) phasic responses to cues, particularly evident in delay tasks that separate in time the presentation of sensory information from movement execution. Moreover, pre-cue anticipatory modulations occur when the cue timing is predictable. We here studied in more detail the motor cortical activity associated with visual cue anticipation and processing, furthermore linking it to the behavioral performance and the activity during movement execution. We recorded neuronal spiking activity and local field potentials (LFPs) in two macaques performing a visuomotor task containing both a pre-cue and a preparatory (pre-GO) delay, which durations (both short or long) was indicated at each trial-start.
Almost half of the neurons showed either increasing or decreasing pre-cue anticipatory activity. Many of those that increased their pre-cue firing rate had a short-latency phasic response to the cue, while those that decreased their pre-cue firing rate were more active during movement execution. However, both subsets had similar proportions of directionally selective neurons, throughout the preparatory delay, suggesting concurrent involvement in movement preparation. Furthermore, pairs of neurons with the same pre-cue anticipatory activity pattern had a positive trial-by-trial pre-cue firing rate correlation, while pairs with opposite pre-cue anticipatory activity patterns had a negative trial-by-trial pre-cue firing rate correlation.
The LFPs contained directionally selective visual evoked potentials (VEPs) to the cue that were larger in short-delay trials, while movement related potentials (MRPs) around movement onset were larger in long-delay trials. The same effects of delay duration were also observed in the spiking response to the cue and around movement onset, albeit weaker. Moreover, VEPs were negatively and MRPs positively correlated with behavioral reaction times.
We propose that the motor cortical activity during cue anticipation and processing is embedded in a timing network. It may reflect a presetting mechanism that complements the subsequent processing during movement execution, while prohibiting a premature response.

P1.155

The ADHD-susceptibility gene *Iphn3.1* modulates dopaminergic neuron formation and locomotor activity during zebrafish development

Lange, M. (Gif sur Yvette)[1], Norton, W. (Leicester)[2], Chaminade, M. (Gif sur Yvette)[1], Vernier, P. (Gif sur Yvette)[3], Lesch, K.-P. (Würzburg)[4], Bally-Cuif, L. (Gif sur Yvette)[1]

[1]*Institut de neurobiologie Alfred Fessard, CNRS UPR 3294-Neurobiologie & Développement Equipe ZEN, Gif sur Yvette, France,* [2]*Department of Biology, University of Leicester, Leicester, United Kingdom,* [3]*Institut de neurobiologie Alfred Fessard, CNRS UPR 3294-Neurobiologie & Développement Equipe "Development and Evolution of Neurotransmission", Gif sur Yvette, France,* [4]*Department of Psychology, Biological Psychology, Clinical Psychology, and Psychotherapy, University of Würzburg, Würzburg, Germany*

Attention-deficit/hyperactivity disorder (ADHD) is a developmental disease characterized by hyperactivity, impulsivity and inattention. However, although there is a significant genetic component to ADHD, relatively few risk genes have been identified and characterized. Furthermore the etiology of ADHD is poorly understood and there are few animal models for the disease. Recently, linkage analysis followed by fine-mapping identified *LPHN3* as a candidate gene for ADHD. *LPHN3* encodes an orphan G protein-coupled receptor whose function is poorly understood.
In recent years, the zebrafish (*Danio rerio*) has been established as a model to study the genetics and developmental bases of behavior. To validate *LPHN3* as a potential ADHD risk factor, we analyzed the function of *lphn3.1* in zebrafish larvae.
We used morpholino injection to achieve a transient reduction of *lphn3.1* activity, and characterized the larval behavioral phenotype of *lphn3.1* morphants. We found that at six days of development, *lphn3.1* morphant larvae display two major ADHD-like behavioral endophenotypes: hyperactivity, and motor impulsivity. This hyperactivity can be rescued by application of the pharmacological treatments for ADHD methylphenidate and atomoxetine, and correlates with altered development of dopaminergic (DA) neurons. We are now in the process of using drugs that target the DA pathway in order to understand the the dysfunction in signal transduction that occurs in morphants. We are applying agonists and antagonists of DA receptors and then examining their effects on locomotion. Promising results provide evidence of an impairment in synaptic DA signaling.
This work provides evidence of Lphn3's role in the control of locomotor activity and DA development. *lphn3.1* morphants have the potential to be developed into a new animal model for ADHD locomotor phenotypes and will help us to expand our knowledge of Lphn3 function.

P1.156

Understanding fronto-polar function: an intracranial EEG study of flexibility in human decision-making

Domenech, P. (Paris)[1], Wyart, V. (Paris)[1], Isnard, J. (Lyon)[2], Koechlin, E. (Paris)[1]
[1]*Ecole Normale Supérieure, Laboratoire de Neurosciences Cognitives, Paris, France,* [2]*Hospices Civils de Lyon, Service de Neurologie fonctionnelle et d'épileptologie, Lyon, France*

Behavioral flexibility in the face of uncertain and changing environments is a hallmark of Human cognition. In this context, decision-making involves exploring possible courses of action and learning about their consequences, to establish -and then exploit- mappings associating stimuli and actions to outcomes. Because natural environments are ever changing, uncertain and open-ended, behavioral fitness further requires being able to drop the mapping in use whenever it no longer predicts action outcomes reliably, and to trigger new exploration phases.
Human fMRI studies suggest that the fronto-polar cortex (FPC) is pivotal in learning new mappings (or *task sets*, TS), departing from them and managing them. FPC expanded dramatically during Human evolution. To date, only one study investigated monkey FPC neural activity and showed it monitored action outcomes. However, the significance of this result for Human FPC function is unclear, as inter-species homologies are not well established yet.
To characterize Human FPC function, we recorded local field potentials in the FPC and the orbito-frontal cortex (OFC) from 6 epileptic patients performing a variant of the Wisconsin Card Sorting Test. Patients learned mappings between 3 stimuli and 4 buttons from noisy feedbacks (90% reliability). Correct mappings were changed pseudo-randomly every 33 to 48 trials. Each patient performed 1950 trials (48 TS) over 2 sessions on successive days. Patients were able to learn correct mappings and swiftly enter new exploration phases after TS changes. Preliminary analyses in a patient showed distinct ERPs to correct and incorrect feedbacks in lateral OFC and FPC. OFC response preceded

that of FPC by 200ms. Unlike previous results in monkeys' FPC, there was no evidence of feedback anticipation. Finally, ERP time-locked at stimulus onset showed differential activity between exploratory and exploitatory trials. Again, OFC activity preceded that of FPC: in OFC, response was phasic and peaked around 400ms, whereas FPC exploration specific response started around 400ms and was sustained for 800ms.
These data are the first direct experimental evidence of TS management in Human FPC. Our findings highlight the inter-species differences in this Phylogenetically recent part of the Human brain.

P1.157

Plasticity in the main olfactory bulb of leptin-deficient ob/ob obese mice

Chelminski, Y. (Orsay)[1], Scotto-Lomassese, S. (Paris)[2], Sezille, C. (Orsay)[1], Martin, C. (Orsay)[1], Genoux, A. (Orsay)[1], Sébrié, C. (Orsay)[3], Gurden, H. (Orsay)[1]
[1]Laboratoire Interdisciplinaire d'Imagerie et de Modélisation en Neurobiologie et Cancérologie, CNRS UMR8165, Université Paris Diderot et Paris-sud, Orsay, France, [2]Institut du Fer à Moulin, INSERM UMR-S 839, Paris, France, [3]Imagerie par Résonance Magnétique Médicale et MultiModalité, UMR8081 CNRS, CNRS UMR8081, Université Paris-sud, Orsay, France

Olfactory cues are crucial for feeding behavior. Olfaction makes possible both the detection and the processing of odor related to food location and palatability. Interestingly, some receptors to anorexigen and orexigen hormones and neuropeptides found in the hypothalamus are also detected in the main olfactory bulb (MOB), suggesting that feeding state has an impact on odor representation. Moreover, our previous work showed that fasting reduces the threshold of odor-evoked spatiotemporal activity in the MOB.
What about MOB plasticity in obesity? Leptin, a peptidergic hormone produced by adipocytes, is a major regulator of the metabolic activity and inhibits food intake. Ob/ob mice are deficient in leptin from birth and are used as a murine model of obesity since they are hyperphagic and are rapidly obese. We used two functional neuroimaging techniques (Manganese Enhanced MRI and intrinsic optical signals imaging) to monitor food odor-evoked spatial maps, and found that these maps cover a larger space in the MOB of ob/ob mice than in control animals. IP injection of leptin carried on obese animals seems to reduce the area of the odor-evoked maps detected by MEMRI in obese mice.
To pinpoint what cellular/molecular mechanisms can be responsible for these changes in the olfactory maps, we first quantified mRNA expression of neuronal (OMP), astrocytic (GFAP, S100B), and microglial (IBA1, ED1) molecular targets by RT-PCR, but did not find any significant changes between ob/ob and control mice. We also quantified bulbar adult neurogenesis and found that 21 days after BrdU injections, a cell birth marker, ob/ob mice showed an increased number of both new periglomerular (28%) and granular (14%) cells as compared to control, suggesting that leptin regulates new neuron elimination. However, it remains to determine whether this increased number of immature cells contributes to the modified olfactory maps in ob/ob mice.
Since obese mice display abnormal odor-evoked maps associated with increased adult neurogenesis in the MOB, we are currently testing their motivational and behavioral phenotype in a Go/No-Go odor discrimination task. In addition, we will probe local network activity in the deep layers of the MOB by recording local field potentials during behavior.

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Mamba toxins reveal new roles for Acid-Sensing Ion channels in pain

Baron, A. (Valbonne)[1], Diochot, S. (Valbonne)[1], Salinas, M. (Valbonne)[1], Douguet, D. (Valbonne)[1], Scarzello, S. (Valbonne)[1], Dabert-Gay, A.-S. (Valbonne)[1], Debayle, D. (Valbonne)[1], Friend, V. (Valbonne)[1], Alloui, A. (Clermont Ferrand)[2], Lazdunski, M. (Valbonne)[1], Lingueglia, E. (Valbonne)[1]
[1]Institut de Pharmacologie Moléculaire et Cellulaire, CNRS /Université de Nice-Sophia Antipolis UMR 7275, LabEx Ion Channel Science and Therapeutics, Valbonne, France, [2]Neuro-Dol, Inserm / Université d'Auvergne UMR 1107, Clermont Ferrand, France

Peptide toxins isolated from animal venoms are invaluable tools to understand physiological and physiopathological functions of ion channels. We have identified from the deadly venom of the African snake black mamba two 57-aa isopeptides, mambalgin-1 and mambalgin-2, which define a new class of three-finger peptides with the property to specifically inhibit Acid-Sensing Ion Channels (ASICs). ASICs are excitatory Na^+ channels largely expressed in the peripheral and the central nervous systems and involved in pain. Mambalgins inhibit all the ASIC channel subtypes expressed in central neurons, *i.e.* homomeric ASIC1a and heteromeric ASIC1a + ASIC2a and ASIC1a + ASIC2b channels, as well as ASIC1b-containing channels that are specific of sensory neurons.
Mambalgins show potent analgesic effects in mice on acute pain and inflammatory hyperalgesia upon central and peripheral injections, without any apparent toxicity. In the central nervous system, the analgesic effects can be as potent as morphine but are resistant to naloxone and do not involve opioid receptors. Mambalgins also seem to produce fewer side effects than morphine. The central analgesic effects are absent in ASIC1a-knockout mice, demonstrating the specificity of the effects and the essential implication of ASIC1a-containing channels. They are also reduced by *in vivo* silencing of the ASIC2a subunit, which supports the involvement of heteromeric ASIC1a+ASIC2a channels in the central analgesic effects. Subcutaneous peripheral injection of mambalgins in the mice hindpaw also induces analgesic effects that are unchanged in ASIC1a-knockout mice but reduced after *in vivo* silencing of the ASIC1b subunit in nociceptors, supporting the involvement of ASIC1b-containing channels in these effects.
These findings identify a potent role in the central and peripheral pain pathways for different ASIC channel subtypes never associated with pain before (*i.e.*, central heteromeric ASIC1a+ASIC2a channels and peripheral ASIC1b-containing channels), and introduce at the same time new natural peptides that block them and could be of potential therapeutic value.
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P1.159

PIXSIC - characterization of a β+ intracerebral wireless probe for PET measurements coupled with behavioral studies in freely moving rats

Balasse, L. (Orsay)[1,2], Maerk, J. (Marseille)[3], Benoit, D. (Orsay)[1], Benoit, M. (Geneva)[4], Clémens, J.-C. (Marseille)[3], Fieux, S. (Lyon)[2], Fourgeron, D. (Marseille)[3], Janvier, B. (Orsay)[1], Jevaud, M. (Marseille)[3], Genoux, A. (Orsay)[1], Gisquet-Verrier, P. (Orsay)[5], Lefebvre, F. (Orsay)[1], Menouni, M. (Marseille)[3], Pain, F. (Orsay)[1], Tourvieille, C. (Lyon)[6], Morel, C. (Marseille)[3], Zimmer, L. (Lyon)[2,7], Lanièce, P. (Orsay)[1]
[1]CNRS, Université Paris sud, UMR8165 IMNC, Orsay, France, [2]CRNL, CNRS UMR5292, INSERM U1028, BioRan, Lyon, France, [3]CNRS, CPPM, Centre de Physique des particules de Marseille, Marseille, France, [4]CERN, PH-LCD, Geneva, Switzerland, [5]CNPS, UMR 8195, CNRS, Orsay, France, [6]CERMEP - Centre d'imagerie du Vivant, Lyon, France, [7]CERMEP - Centre d'imagerie du Vivant, Lyon, France

Understanding of neurophysiological mechanisms to decode the functional specificity of brain regions based on small animal in vivo studies requires the development of original and well-adjusted methods and instruments. In this field, an exciting challenge remains in the combination of brain imaging techniques and behavioral studies, which helps to associate molecular processes of neuronal communications to the related actions they initiate. However, recent approaches in this context, such as RATCAP and β-Microprobe, are still affected by important constraints considering the investigation of awake and freely moving animals. PIXSIC presents a novel strategy using a submillimetric telemetric pixellated probe for β+ radiotracer detection based on a reverse biased, high-resistivity silicon diode. The detector permits local time-activity measurements with high sensitivity and additional imaging features.
The sensor is 200 µm thick, 690 µm wide and 17 mm long and comprises 10 pixels, with dimensions of 200 µm x 500 µm. The probe is wire-bonded to a head socket fixed on the skull that supports the specific micro-ship (ASIC) for the parallel signal processing of each pixel. This head socket is stereotaxically implanted in the region of interest in rodent brain and is connected to a back-board

worn by the animal in a backpack that supports the components required to drive the ASIC and to deliver a RF telemetric signal to the acquisition board on a PC. The RF module renders the setup fully autonomous and therefore limits stress induced in the animal during acquisition and enables behavioral studies.
The first biological validation was performed on anaesthetized rats implanted in region of interest (hippocampus or striatum) with PET radiotracers injection ([^{11}C]-Raclopride for D_2 receptors or [^{18}F]-MPPF for $5HT_{1A}$ receptors). We demonstrated the feasibility of using PIXSIC to measure radioactive concentrations in specific regions of the rat brain. The specific binding curves obtained in vivo were reproducible and showed a clear correlation between the level of radioactivity measured by each pixel of PIXSIC and their location in the brain (cortex/striatum or cortex/hippocampus). In addition, PIXSIC allowed us to make the first kinetics on awake and freely moving rats.

P1.160

Slow and fast adaptive processes in tactile perceptual decision making: behavioral and MEG evidence

Sanchez, G. (Lyon)[1,2], Partouche, J. (Lyon)[1,2], Maby, E. (Lyon)[1,2], Bertrand, O. (Lyon)[1,2], Mattout, J. (Lyon)[1,2]
[1]INSERM U1028 - CNRS UMR5292, Lyon Neuroscience Research Center - DYCOG Team, Lyon, France, [2]University Lyon 1, Lyon, France

Perceptual decision making and its neuronal correlates are often studied by minimizing the influence of adaptive processes induced by practice or contextual sets. Such processes, which may have profound impacts on performance at different time scales, are largely unknown although they reflect central mechanisms of perception and decision making.
In two separate studies, using a typical comparison task of two consecutive tactile stimulations, we investigated the effects and interplay of a slow adaptive process and the fast implicit learning of a contextual rule.
1st study (effect of practice): 20 subjects performed 6 similar sessions, where the first stimulation remained constant (F1=30Hz), while the second ranged between 22 and 38Hz (F2).
2nd study (interaction between practice and implicit context learning): Three groups of subjects performed 6 sessions of the same task, except for sessions 4 and 5, where F2 was always 2Hz higher or lower than F1. During those two sessions, in group 1 (G1), F1 varied and ranged between 25 and 35Hz (mean=30Hz). In group 2 (G2), F1 ranged between 21 and 31Hz (mean=26Hz). In group 3 (G3), F1 ranged between 29 and 39Hz (mean=34Hz).
1st study: the behavioral data emphasize a learning phase during which performance improves, followed by a plateau. Evoked responses to F1 also reflect these two phases by suggesting that the precise encoding of F1 in each trial is essential in the first phase only. In that phase, we show the crucial role of inter-connections between the inferior frontal gyrus and the somatosensory cortices.
2nd study: the learning of a contextual rule in sessions 4 and 5 was revealed by a contraction bias. Interestingly, this effect was further biased, although temporarily, towards the previously acquired reference at 30Hz, in groups G2 and G3. In contrast, performances in session 6 were not degraded at all compared to the ones in session 3, whatever the direction of the induced contraction bias.
These results shed light on the interplay of two mechanisms that bias perceptual decision making: the relatively slow implicit learning of a perceptual reference and the faster up-dating of a contextual prior based on the recent history of past stimulations.

P1.161

Decrease of cerebral mast cell deganulation after systemic administration of lipopolysaccharide

Dubayle, D. (Paris)[1], Héron, A. (Paris)[2]
[1]*Université Paris Descartes - UFR Biomédicale, CNRS UMR 8119, Paris, France,* [2]*Université Paris Descartes - Faculté de Pharmacie, Département de Biologie Expérimentale, Paris, France*

Mast cells (MC) are the only immunocytes known to infiltrate the brain in physiological conditions, and are especially numerous in the thalamus. Their exact cerebral function remains to be elucidated. The aim of the present study was to investigate change of the thalamic MC population in a weak model of inflammation. The number of MC and the degree of degranulation were quantified and analyzed with acidified toluidine blue histochemistry, in two groups of five adult male Wistar rats, four hours after saline (control) or systemic injection of gram-negative bacterial lipopolysaccharide (LPS, 100 micrograms/kg, i.p). Results showed that MC had always a preferential perivascular localization. LPS treatment significantly decreased the degranulated MC number by 59%, (p=0.01) in the lateral thalamus nuclei with no significant effect on the total MC number. We hypothesize that, a weak inflammation induced by LPS could thus prevent the opening of blood brain barrier and cerebral sepsis.

P1.162
Evidence for a neuroendocrine pulse generator from organotypic hypothalamic cultures

Israel, J.-M. (Bordeaux)[1], Garenne, A. (Bordeaux)[2], Le Masson, G. (Bordeaux)[3], Oliet, S.H.R. (Bordeaux)[1], Ciofi, P. (Bordeaux)[1]
[1]*Université Bordeaux Segalen, Neurocentre Magendie - Inserm U.862, Interactions Glie-Neurones, Bordeaux, France,* [2]*Université Bordeaux Segalen, IMN, CNRS UMR 5293, Mnemosyne, Bordeaux, France,* [3]*Université Bordeaux Segalen, Neurocentre Magendie, INSERM U862, Pathologies du Système Moteur, Bordeaux, France*

Pituitary hormones are released in pulses mirroring bursting electrophysiological activity of neuroendocrine cells. Whether bursting is intrinsic to neuroendocrine cells or driven by afferents is unclear. We tested this in supraoptic (SON) oxytocin (OT) neurons that typically burst in response to pup suckling during lactation. In anterohypothalamic organotypic cultures of neonatal rats, intracellular electrophysiological pair recording of identified OT neurons revealed their synchronized bursting activity identical to that seen in suckled dams in vivo, suggesting a drive by intrahypothalamic networks present at birth (JN 18:6641; EJN 17:2619). If a specific network is involved, this must fit with canonical mechanisms of sexual differentiation irreversibly sculpting neural networks neonatally.
OT neurons displayed synchronized bursts in most (>90%) cultures harvested from female neonates at postnatal day 5 (PN5), PN7, PN9 or PN12, as well as in cultures from males at PN5 (>90%) and PN7 (50%) but not older, suggesting defeminization by circulating androgens. In line, bursting behavior in cultures harvested at PN9 was, rescued in males by orchidectomy at PN5, and abolished in females by testosterone given from PN5 to PN7. Testosterone also abolished bursting behavior *in vitro* when applied for 72h in tissues harvested at PN5, irrespective of sex. This *in vitro* effect of testosterone was, blocked by the aromatase inhibitor letrozole or the pan-caspase inhibitor zVADfmk, and mimicked by the estrogen receptor (ER) alpha selective agonist PPT. This suggests defeminization of OT cells bursting behavior by locally produced estrogen from aromatized testosterone acting via ERalpha to induce apoptosis of a determinant cell population. Histologically *in vivo*, Hoechst labeling revealed a very low number of apoptotic-like cell nuclei in the SON with no sex-difference, and a rather exclusive expression of ERalpha in the periventricular area (PeA). Accordingly, removal of the PeA from tissue slices harvested at PN5 abolished bursting activity irrespective of sex, and apoptotic-like cell nuclei were in both sexes equally abundant in the PeA with a decline after PN7.
Seemingly, a periventricular network generates bursts in OT cells and is specifically pruned postnatally in males.

P1.163

The endothelial-cell-derived factor Semaphorin 3A controls GnRH axon plasticity in the adult brain

Giacobini, P. (Lille)[1], Campagne, C. (Lille)[1], Parkash, J. (Lille)[1], Langlet, F. (Lille)[1], Casoni, F. (Lille)[1], Cagnoni, G. (Candiolo)[2], Hanchate, N.K. (Lille)[1], Mazur, D. (Lille)[1], Ciofi, P. (Bordeaux)[3], Bouret, S. (Lille)[1,4], Tamagnone, L. (Candiolo)[2], Prevot, V. (Lille)[1]
[1]Inserm U837, Universite de Lille 2, JPARC, Equipe 2, Lille, France, [2]Institute for Cancer Research and Treatment (IRCC), University of Torino Medical School, Candiolo, Italy, [3]Inserm, Neurocentre Magendie, U862, Université de Bordeaux, Bordeaux, France, [4]The Saban Research Institute, Childrens Hospital Los Angeles, Univ. Southern California, Los Angeles, United States

Semaphorin 3A (Sema3A) / Neuropilin-1 (Nrp1) signaling guides the development of the nervous and vascular systems during embryogenesis, but its role in the adult brain is unknown. Here, we report that Sema3A, the ligand of Nrp1, is selectively expressed in the capillary zone within the median eminence of the hypothalamus, the projection site of neurons secreting gonadotropin releasing hormone (GnRH), the neuropeptide controlling reproduction. We show that endothelial cells of the median eminence secrete this class 3 secreted semaphorin during a discrete window of the ovarian cycle and that Sema3A/Nrp1 signaling is required for the advancement of GnRH nerve terminals towards the vascular wall on the day of the preovulatory surge. Loss of function of Nrp1 in GnRH neurons also abrogates Sema3A-mediated structural remodeling. Finally, temporary and local *in vivo* infusion of antibodies that neutralizes Nrp1 or Sema3A function disrupts the ovarian cycle, which requires a pulsatile, coordinated delivery of GnRH into the hypothalamo-hypophyseal portal system. These data thus suggest that endothelium-to-neuron communication processes involving Sema3A/Nrp1 signaling are functionally and physiologically relevant to the adult reproductive brain. Altogether our results also raise the intriguing possibility that endothelial cells may actively participate in synaptic plasticity in specific functional domains of the adult brain.

P1.164

Gap junction signaling disclosed as a stress-regulated component of adrenal neuroendocrine stimulus-secretion coupling *in vivo*

Guérineau, N.C. (Angers)[1], Jourdan, C. (Montpellier)[2], Toutain, B. (Angers)[1], Hormuzdi, S.G. (Dundee)[3], Desarménien, M.G. (Montpellier)[2]
[1]Lab. de Biologie Neurovasculaire Intégrée, CNRS UMR6214-INSERM U1083-Univ. Angers-UFR Sciences Médicales, Angers, France, [2]Institut de Génomique Fonctionnelle - CNRS UMR5203-INSERM U661-Univ. Montpellier, Montpellier, France, [3]Division of Neuroscience, Univ. Dundee, Dundee, United Kingdom

Catecholamine release from the adrenal medulla is vital to cope with stress but excessive plasma concentration may be deleterious, thus secretion needs tight regulation. Catecholamine release is chiefly triggered by synaptic transmission from the splanchnic nerve but recent *ex vivo* studies state that stimulus-secretion coupling can be modulated by gap junction-mediated communication between chromaffin cells. Indeed, gap junctions ensure propagation of electrical and ensuing Ca^{2+} signals and increase catecholamine release in response to stimulation of a single cell. Whereas converging arguments support that gap junctional coupling contributes to catecholamine release in acute slice preparation, its contribution to excitation-secretion coupling *in vivo* is still unknown. Therefore, we developed a microsurgical approach allowing combined monitoring of catecholamine concentration from adrenal vein blood and splanchnic nerve stimulation in anesthetized mice. Gap junction contribution was assessed pharmacologically by *i.p.* injection of the uncoupling agent carbenoxolone (CBX) or its inactive analog glycyrrhizic acid. In control mice, CBX reduced by 58% norepinephrine release evoked by high (4 Hz), but not low (0.1 Hz), frequency stimulation. Epinephrine release was not affected. Immunohistofluorescence and qRT-PCR demonstrated that Cx36 is the major connexin expressed in the mouse adrenal medulla. In stressed mice (5-day cold exposure), expression of adrenomedullary Cx36 was up-regulated. Under these conditions, secretion of both norepinephrine

and epinephrine evoked by high frequency stimulation was drastically inhibited (by 76%) by CBX. Interestingly, in Cx36$^{-/-}$ mice, a severe impairment of splanchnic nerve stimulation-evoked catecholamine secretion was observed. The failure likely occurred presynaptically as evidenced by a substantial reduction in both neurofilament and vesicular acetylcholine transporter staining. Our data demonstrate a positive contribution of gap junctions to catecholamine secretion *in vivo* and argue for a novel developmental role for gap junctional coupling in the rodent adrenal medulla.

P1.165

Role of the chemokine network in T-cell brain infiltration and neurodegeneration in a mouse model of Parkinson´s disease

Hunot, S. (Paris)[1], Bekaert, A. (Paris)[1], Brochard, V. (Paris)[1], Dalard, C. (Paris)[1], Monnet, Y. (Paris)[1], Lobsiger, C. (Paris)[1], Hirsch, E.C. (Paris)[1], Combadière, C. (Paris)[2]
[1]Hôpital de la Pitié-Salpétrière, INSERM UMRS 975-CNRS UMR 7225- Institut du cerveau et de la moelle épinière, Paris, France, [2]INSERM U543 - Hôpital de la Salpêtrière, Cellular and Tissue Immunology, Paris, France

Mounting evidence suggests that non-cell autonomous mechanisms play a role in the demise of dopaminergic neurons (DN) in Parkinson´s disease (PD). In line with this, we have recently reported that peripheral CD4+ T cells not only invade the brain in PD patients and MPTP-treated mice (an experimental model of PD) but also actively participate to DN death suggesting that therapeutic strategies aimed at preventing lymphocyte extravasation may be of interest. To unravel the mechanisms underlying such T cell infiltration we here focused on the role of the chemokine network. We first investigated the chemokine signature in the ventral mesencephalon of MPTP-treated mice using TaqMan® Low Density Arrays. We found a strong upregulated expression of the chemokine ligands Cxcl10, Ccl2,3,4 and 5. Similar up-regulation of CCL3, CCL4 and CXCL10 was evidenced by ELISA assay. Furthermore, double immunofluorescent staining on mesencephalic tissue sections demonstrated that CCL3 and CCL4 were exclusively expressed by Iba1-positive microglial cells whereas CXCL10 was expressed by GFAP-postive astrocytes. Interestingly, expression analysis of the most promising candidates by qPCR on human post-mortem substantia nigra tissues revealed an increased expression of Ccl3 and Ccl4 in PD patients whereas Ccl5 and Cxcl10 were expressed at similar levels as in age-matched control individuals. To get functional mechanistic insights into the role of the chemokine network in T cell-associated neuronal cell death, we then assessed the impact of chemokine receptor deficiency on disease outcome after MPTP challenge in mice. Our results indicate that deletion of either CCR5 or CXCR3 in mice does not modify the overall toxicity of MPTP. To test whether compensatory and/or redundant mechanisms may underline this null effect we next attempted to block CXCR3 and CCR5/CCR1 simultaneously using a combinatory genetic and pharmacologic approach. This was achieved by delivering Met-RANTES, a CCR5/CCR1 antagonist, to CXCR3-deficient mice. In this condition, a significant neuroprotection (50%) was observed. Overall, our data indicate that modulation of the chemokine network associated with glial cell activation may participate to the recruitment of pathogenic T cells during nigrostriatal pathway injury.

P1.166

Collapsin Response Mediated Protein -2 (CRMP2), a possible peripheral biomarker of neuroinflammation

Giraudon, P. (Lyon)[1], Nicolle, A. (Lyon)[1], Varrin-Doyer, M. (Lyon)[1], Bertrand, C. (Lyon)[1], Cavagna, S. (Lyon)[1], Benetollo, C. (Lyon)[2], Marignier, R. (Lyon)[1]
[1]Centre de Recherche en Neuroscience de Lyon INSERM 1028 CNRS UMR5292 Univ Lyon 1, Neurooncology Neuroinflammation Team, Lyon, France, [2]Centre de Recherche en Neuroscience de

Lyon INSERM 1028 CNRS UMR5292 Univ Lyon 1, Plateforme Genetique Fonctionnelle et Optogénétique, Lyon, France

Recruitment of T lymphocytes in the CNS is a crucial phase in neuroinflammatory disease such as Multiple sclerosis (MS). However, little is known on the migratory mechanism of immune cells. Our studies have revealed an unexpected role for the phosphoprotein CRMP2, a cytoskeleton organizer first described in neural cells. CRMP2 is involved in T-cell polarization and migration (Vincent et al., 2005). In neural cells, CRMP2 transduces semaphorin signal; in immune cells, CRMP2 function is regulated by chemokines and the CXCL12/CRMP2/PI3K signalling pathway. Chemokine-treated T-cells induced differential phosphorylation of CRMP2, modulating its contribution to the cytoskeleton reorganization. The functional importance of the CRMP2-p479 form in T-cell migration was demonstrated (Varrin-Doyer et al., 2009). In model of virus-induced neuroinflammation, high CRMP2 expression in activated T-cell correlated the brain infiltration and clinical scores (Vuaillat et al., 2008). This pointed out the potential use of CRMP2 as a peripheral indicator of neuroinflammation. We investigated CRMP2 expression in MS patients *versus* Healthy Donors. Cytometry analysis of peripheral blood mononuclear cells (PBMC) showed the differential expression of CRMP2 in various cell subsets (T-cells; T-regulators, B-cells; NK cells; M1/M2 macrophages). In MS patients, CRMP2 was specifically elevated in activated (HLA-DR+) T-cells . Selection of these cells from HD and MS patients and gene analysis showed an association between CRMP2 level expression, enhanced migratory rate and activation of molecular pathways of cell motility. CRMP2-p479 level, quantified in total PBMC (WBlot), was specifically and differentially enhanced in RR-MS and PR-MS. Additional study showed a link between CRMP2-p479 level and chemokine signal in human immune cells *ex vivo*. Thus, the elevated CRMP2-p479 expression seen in MS patients presumably reflects an efficient contact of blood immune cells with chemokine(s), hence a stimulus to migrate and invade neural tissues. Similar data were observed on T-cells from mouse lymph nodes in experimental autoimmune encephalitis model EAE. All these data confirm our interest for CRMP2, notably CRMP2-p479 as a potential peripheral biomarker of neuroinflammation useful at least in MS.

P1.167

Hypothyroidism induces Alzheimer's disease-related pathological hallmarks and associated memory impairments

Chaalal, A. (Orsay)[1], Poirier, R. (Orsay)[1], Blum, D. (Lille)[2], Gillet, B. (Orsay)[3], Le Blanc, P. (Orsay)[1], Basquin, M. (Lille)[2], Buée, L. (Lille)[2], Laroche, S. (Orsay)[1], Enderlin, V. (Orsay)[1]
[1]Center of Neuroscience Paris-Sud, CNRS-UMR8195, Orsay, France, [2]Centre de recherche Jean Pierre Aubert, INSERM U837, Lille, France, [3]Imagerie par Résonance Magnétique Médicale et MultiModalité, UMR8081 CNRS, Orsay, France

Alzheimer's disease (AD) is a multifactorial disease and to date no single cause of the sporadic forms, which accounts for over 99% of the cases, has been established. Although transgenic mice models contributed significantly to our understanding of the molecular pathogenesis of AD, an alternative and potentially fruitful approach is to develop models of dysfunctional mechanisms associated with risk factors for AD. Converging evidence suggests a possible link between AD and thyroid dysfunctions. In this context, using a hypothyroid rat model induced by propylthiouracil (PTU) treatment, we have recently reported that hypothyroidism increases the vulnerability to the formation of amyloid deposits in the hippocampus, a brain structure affected in early stages of AD and which plays a crucial role in memory processes. The aim of the present study was to explore the possibility that adult hypothyroidism represents an etiopathogenic mechanism of AD. We investigated the consequences of PTU treatment on Tau phosphorylation, a pathological hallmark of AD, and on alterations of markers of inflammation and of molecules involved in synaptic plasticity and cognitive functions. *In vivo* MRI revealed a progressive decrease in cerebral volume of PTU-treated rats compared to controls. In the hippocampus, hyperphosphorylation of Tau was observed in hypothyroid rats, together with an increase in neuroinflammatory cytokines (IL1β, TNFα, Il6, CRP) and a reduced expression of signalling molecules important for learning and memory, including neurogranin (RC3), calmodulin kinase II (CaMKII), cAMP response element-binding protein (CREB) and early growth response protein 1 (EGR1). These alterations were associated with impaired spatial learning and memory.

Together, these data support the idea that disruption of thyroid signalling could participate in the aetiology of AD and present a physiopathological model pertinent for the future exploration of preventive approaches against AD.

P1.168

Periplaques form extensive lesions in multiple sclerosis spinal cords

Lhuillier, A. (Lyon)[1], Chanal, M. (Lyon)[1], Androdias, G. (Lyon)[1], Reynolds, R. (London)[2], Confavreux, C. (Lyon)[1], Nataf, S. (Lyon)[1]
[1]Centre de Recherche en Neurosciences de Lyon (CRNL). Inserm U1028-CNRS 5292- UCBL1, Équipe neuro-inflammation et neuro-oncologie, Lyon, France, [2]Center for Neuroscience, Imperial College, London, United Kingdom

Our knowledge of multiple sclerosis (MS) neuropathology has benefited from a number of studies that precisely depicted different types of plaques in white or grey matter areas. Also, both inflammation and diffuse axonal loss in the normal appearing white matter (NAWM) were extensively described. However, little attention was given to the so-called periplaque, which is usually defined as a partially demyelinated ribbon of tissue surrounding the plaque. Whether periplaques correspond to expanding lesions, sites of ongoing remyelination or areas of tract degeneration remains uncertain. In this context, our study aimed to bring quantitative insights to the neuropathology of MS periplaques in the spinal cord of primary or secondary progressive MS patients. A neuropathological quantitative assessment of inflammation, axonal loss and myelin loss was performed concurrently in the periplaques, plaques and normal-appearing white matter (NAWM) of cervical spinal cord samples from 16 patients with progressive MS. Periplaques formed large areas of incomplete myelin loss that extended distant away from the border of plaques. Axonal loss was quantitatively similar in plaques and periplaques but signs of axonal dystrophy were predominantly observed in plaques. Surprisingly, axons that remained myelinated in periplaques presented a thicker myelin sheath than myelinated axons of the NAWM. Inflammation in the periplaque was mainly characterized by an accumulation of macrophages/microglia that were closely apposed to myelin sheaths but exerted poor phagocytic activity. Finally, we found that neuropathological features of periplaques were overall disconnected from that of plaques with regard to size, shape, inflammation and axonal integrity. Our work indicates that in MS spinal cords, periplaques correspond to demyelinating rather than remyelinating lesions. It further suggests that in progressive forms of MS, periplaques are likely to impact significantly on neurological disability. Finally, we propose that tract degeneration might not be the only cause of periplaque extension and that slowly-expanding demyelination, temporally remote from plaque formation, might occur in periplaques. Molecular results will be presented that support these hypotheses.

P1.169

Morphological changes associated to microglial activation after spinal cord injury: from amiboid phenotype to multinucleated cell formation

Thomaty, S. (Marseille)[1], Xerri, C. (Marseille)[1], Brezun, J.-M. (Marseille)[1]
[1]Aix Marseille Université, Laboratoire de Neurosciences Intégratives et Adaptatives CNRS / UMR 7260, Marseille, France

The neuroinflammatory processes after injury are characterized by a cascade of both cellular and molecular events. Interactions between glial, vascular and resident microglial cells lead to major morphological changes. Activated microglia classically exhibits an involution of its processes and becomes amiboid in shape. Other changes have been reported in specific models such as persistent microbial infection or post-injury chronic inflammation. Indeed, in these circumstances, some microglial cells and/or macrophages can trigger cell-cell fusion leading to multinucleated giant cell formation. Little is known about the mechanisms associated to these events, however it has been proposed that they are promoted when phagocytosis is outstripped.
In this study, we investigated the fusion of microglial cells after spinal cord injury (SCI). Our model consists in a C4-C5 medio-lateral spinal hemisection performed in 3-month-old Wistar male rats. Histological investigations were processed over the first month after SCI using Iba-1 immunoreactivity (Iba-1-Ir) as a microglial marker and Hoechst labelling for nuclei detection. Phagocytosis was evaluated with ED1-Ir labelling of the lysosomal compartment. The cell fusion was determined using confocal microscopy analysis. Fusioned cells were defined by the lack of Iba-1-Ir between two nuclei.

Our results point out the presence of fusioned cells in the perilesion area until the 1 month postlesion period examined. However, the number of nuclei per cell is smaller and these fusioned cells are systematically located in Iba-1-Ir aggregates compared with previous descriptions. Most of the Iba-1-Ir clusters are constituted by non-fusioned cells. For both the aggregated and fusioned Iba-1-Ir cells the intra-cytoplasmic ED1-Ir is mainly located in the injured white matter tracts, while weakly detected in the grey matter. Furthermore, the Ed1-Ir exhibit a strong variability among clusters.
These results strongly suggest that two different mechanisms, i.e. aggregation and moderate fusion, are involved in the atypical microglial activation reported herein. The function of multinucleated giant cells is still unclear but one could hypothesize that these cells contribute to adapt the inflammatory response to lesion severity.

P1.170

The location of feedback-related activity in the midcingulate cortex is predicted by local morphology

Amiez, C. (Bron)[1,2], Neveu, R. (Bron)[3], Warrot, D. (Bron)[1,2], Petrides, M. (Montreal)[4], Knoblauch, K. (Bron)[1,2], Procyk, E. (Bron)[1,2]
[1]INSERM U846, Bron, France, [2]Université de Lyon, Lyon 1, Unité mixte de recherche S-846, Lyon, France, [3]Neuroscience Research Center, Bron, France, [4]McGill University, Montreal Neurological Institute, Montreal, Canada

Information processing in the medial frontal cortex is often said to be modulated in pathological conditions or by individual traits. This has been observed in neuroimaging and event-related potential studies centered in particular on midcingulate cortex (MCC) functions. This region of the brain is characterized by considerable intersubject morphological variability. Whereas in a subset of hemispheres only a single cingulate sulcus (cgs) is present, a majority of hemispheres exhibit an additional sulcus referred to as the paracingulate sulcus (pcgs). The present functional magnetic resonance imaging study defined the relationship between the local morphology of the cingulate/paracingulate sulcal complex and feedback-related activity. Human subjects performed a trial-and-error learning task in which they had to discover which one of a set of abstract stimuli was the best option. Feedback was provided by means of fruit juice, as in studies with monkeys. A subject-by-subject analysis revealed that the feedback-related activity during exploration was systematically located in the cgs when no pcgs was observed, but in the pcgs when the latter sulcus was present. The activations had the same functional signature when located in either the cgs or in the pcgs, confirming that both regions were homologues. Together, the results show that the location of feedback-related MCC activity can be predicted from morphological features of the cingulate/paracingulate complex.

P1.171

Phasic dopamine release and choice behaviour in a changing environment

Baudonnat, M. (Oxford)[1], Lin, E. (Cardiff)[2], Huber, A. (Oxford)[1], Walton, M.E. (Oxford)[1]
[1]Oxford University, Experimental Psychology, Oxford, United Kingdom, [2]University of Cardiff, School of Psychology, Cardiff, United Kingdom

We live in a changeable environment and, to make advantageous choices, it is vital to be able to adjust behaviour appropriately when the world changes. One system that is likely important to this process is the mesolimbic dopamine (DA) projection to the nucleus accumbens (NAc), which is involved in aspects of reinforcement learning, motivation and decision making. However, little is known about what precise role DA transmission on a trial-by-trial basis plays in tracking parameters such as value, uncertainty and behavioural preference, particularly in situations when the value of an option can change dynamically during the course of learning. To investigate this issue, we employed fast-scan cyclic voltammetry (FCV) during an operant decision making task in which rats chose between two levers associated with a certain amount of work (cost) to gain a certain quantity of reward (benefit). While the cost/benefit associations of one option remained fixed throughout testing, we selectively manipulated either the cost or the benefit associated with the other option in order to influence which lever they would prefer. Using this manipulation, we could therefore compare dopamine transmission in the NAc core region to a change in the absolute value of an option with changes in the relative value of an option caused by a change in reward size / work requirement of the alternative option. While both manipulations of work and reward caused comparable changes in

behaviour, dopamine at the time of reward delivery only scaled with unexpected changes in reward magnitude. While DA at the initial cue was not always a good predictor of behavioural preference, there were marked boosts of DA at when the levers were presented, which was particularly strong when updating preference towards the changing option. These data suggest that phasic DA may provide a permissive signal to explore the environment to gain reward, particularly at times of uncertainty.

P1.172

Early-life experience differentially affects cynomolgus monkey depressive-like behaviours in farming conditions: the cumulative effect of adversity?

Camus, S. (Bordeaux)[1], Rochais, C. (Paimpont)[2], Blois-Heulin, C. (Paimpont)[2], Li, Q. (Manchester)[3], Hausberger, M. (Rennes)[4], Bezard, E. (Bordeaux)[5]
[1]Université Bordeaux Segalen, IMN CNRS UMR 5293, Bordeaux, France, [2]UMR 6552 CNRS Ethologie animale et humaine, Université de Rennes 1, Paimpont, France, [3]Motac Neuroscience Ltd, Manchester, United Kingdom, [4]UMR 6552 CNRS Ethologie animale et humaine, Université de Rennes 1, Rennes, France, [5]CNRS, IMN UMR 5293, Bordeaux, France

Adverse early-life experience might lead to the expression of abnormal behaviours in animals and the predisposition to psychiatric disorder in Humans. Common breeding processes employ weaning and housing conditions different from what happens in the wild. Following the identification of spontaneous atypical/abnormal behavioural- and their associated physiological-profiles among 40 single-housed cynomolgus macaques, we investigated whether early-life experience impacts the possible existence of these spontaneous atypical behaviours in 40 captive-born and 40 wild-born socially-housed counterparts. Data were collected in farming conditions using an unbiased ethological scan-sampling method followed by multifactorial correspondence and hierarchical clustering analyses. In each housing or origin conditions, we identified several distinct profiles that significantly differed on many behaviours, body postures, body orientations, gaze directions, distances between individuals and locations in the cage. Data suggest that 4 single-housed, 4 captive-born and 1 wild-born animals present depressive-like symptoms, unnatural early life events thereby increasing the risk of developing pathological symptoms. General differences were also highlighted between the captive- and wild-born populations, implying the expression of differential coping mechanisms in response to the same captive environment.
Unsurprisingly single-housing promotes the development of atypical ethologically-defined behavioural profiles, reminiscent of certain depressive-like symptoms. But birth origin also impacts the expression of such profiles and should be considered when setting up a preclinical research protocol. The use of unbiased behavioural observations might allow the identification of animal models of human mental/behavioural disorders and their most appropriate control groups.

P1.173

Brain regions involved in forgetting during spatial working memory

Youssef, M. (Lyon)[1], Fraize, N. (Lyon)[1], Arthaud, S. (Lyon)[1], Libourel, P.A. (Lyon)[1], Parmentier, R. (Lyon)[1], Salin, P.A. (Lyon)[1], Malleret, G. (Lyon)[1]
[1]CNRS UMR 5292, Lyon, France

A distinction is made between Working Memory (WM) and Reference Memory (RM). WM is a specific form of short-term memory that refers to the ability to retain information within a single trial. This information can then be stored into reference memory. RM refers to the long-term information storage that remains constant over time and that is gradually acquired over many training sessions. However,

not everything should be memorized and not all information that comes from WM must be transferred into RM. Insignificant data is better to be erased in order not to overload the brain with irrelevant things. It has been demonstrated that WM is very sensitive to proactive interference which is a phenomenon whereby information learned in the past interferes with the learning or memory of more recently presented material. Consequently, forgetting this interfering information would be necessary to perform everyday tasks requiring WM abilities. To study the mechanism of how these interferences could be forgotten during WM, we designed different spatial tasks in an 8-arm radial maze requiring the use of spatial orientation and memory. Then, we carried out an immunohistochemical study in rats in order to identify brain regions involved in processing RM and WM with and without interference across two stages of learning. The immediate early gene *zif268* was used as an indirect marker of activity and plasticity and its expression was examined following four and ten days of training. Our results suggest a significant increase in Zif268 expression in the hippocampus, Entorhinal cortex and Prefrontal cortex in the three tasks on day ten compared to controls and to day four. However, the Dentate Gyrus displayed the most unique pattern of activity, with expression of Zif268 remained constant and not significantly different from controls across the two days. This pattern was specifically observed in WM task with high level of interference. This suggests that the non-activation of the Dentate Gyrus is necessary to accomplish the task and overcome interference. Our work sheds light not only on the question of how the Dentate Gyrus responds to interferences, but also on the mechanisms of "forgetting" what should be forgotten.

P1.174

High-frequency stimulation of the hippocampus blocks both fear learning sensitization and return of extinguished fear

Deschaux, O. (Marseille)[1], Koumar, O.-C. (Nice)[2], Spennato, G. (Nice)[2], Moreau, J.-L. (Basel)[3], Garcia, R. (Marseille)[1]
[1]Institut de Neurosciences de la Timone, Equipe SteVe, Marseille, France, [2]Université de Nice Sophia-Antipolis (UNS), Sciences de la Vie, Nice, France, [3]Pharma Division, Hoffmann-La Roche, Basel, Switzerland

Patients with post-traumatic stress disorder (PTSD) are known to present both fear learning sensitization and fear return following extinction, both phenomena being associated with hippocampal deactivation. Rats submitted to a strong fear conditioning can also display sensitization of a subsequent weak conditioning. Following fear extinction, exposure of rats to a weak fear conditioning can also provoke return of extinguished fear, which can be prevented by hippocampal high-frequency stimulation (HFS). Our goal was to examine whether such a treatment also interfere with fear learning sensitization in rats. With field potential recording, we found long-term potentiation in the PFC following hippocampal HFS. Our behavioral results showed that hippocampal HFS, which was applied before the weak conditioning, was able to block both fear learning sensitization and fear return. This finding indicates a pivotal role of the hippocampus in preventing both proactive and retroactive effects of successive conditionings. These data also support the idea according to which hippocampal deactivation in PTSD patients may be involved in fear learning sensitization and fear return seen in these patients.

P1.175

Regulation of the HPA axis and response to antidepressant: evidence from an animal model of depression

Khemissi, W. (Tours)[1], Farooq, R.K. (Tours)[2], le Guisquet, A.M. (Tours)[1], Sakly, M. (Bizerte)[3], Belzung, C. (Tours)[1]
[1]UFR Sciences et Techniques, Tours, France, [2]UFR Sciences et Techniques, Département Neurosciences, Tours, France, [3]Faculté des sciences de Bizerte université de carthage, Bizerte, Tunisia

Antidepressant therapy does achieve remission only in a subpopulation of depressed patients. Interestingly, altered regulation of the HPA axis is a predictor of treatment response in humans but the mechanisms involved in this are poorly understood. As further progress involves animal models, it becomes necessary to mimic this resistance and the contribution of the HPA axis regulation in animals. The Unpredictable Chronic Mild Stress (UCMS) in mice is an animal model of depression which has the advantage of reproducing the role of socio-environmental stress in the onset of a depressive episode and to induce a set of alterations suggestive of symptoms of depression and to

predict effectiveness of the response to antidepressants. As a first step, we subjected BALB/C mice to UCMS in order to induce depressive-like alterations. After two weeks of stress exposure we measured the regulation of the HPA axis using the dexamethasone (0.1mg/kg, ip) suppression test. According to the percentage of corticosterone suppression after dexamethasone injection, we divided the mice into two groups: Strong responders (FR) and weak responders (NR). From the fifth week onwards, we administered fluoxetine (an antidepressant from SSRI group) at 15mg/kg (i.p.) daily. At the end of 7th week, we applied a battery of behavioral tests assessing the emotional, cognitive, and motor aspects of UCMS induced depressive-like behavior. Our results show that fluoxetine-induced antidepressant effects were observed with higher amplitude in FR when compared to NR in various behavioral aspects like coat state, novelty suppression of feeding, splash test and Nest test. The same profile was found concerning the immunohistochimical analysis of ki-67 positive cells in the dentate gyrus of the hippocampus, which is a marker of neuronal proliferation. This suggests that the failure of fluoxetine to induce antidepressants effects was associated to poor ability of the compounds to stimulate cell proliferation in the hippocampus.

P1.176

At what age are rat pups able to encode time? An investigation using odor fear conditioning

Boulanger Bertolus, J. (Lyon)[1], Ahers, J. (New York)[2], Londen, E. (New York)[2], Hegoburu, C. (Lyon)[1], Rousselot, J. (Lyon)[1], Doyère, V. (Orsay)[3], Sullivan, R.M. (New York)[2], Mouly, A.-M. (Lyon)[1]
[1]Université Lyon 1, CRNL - UMR 5292 - INSERM U1028, Lyon, France, [2]Emotional Brain Institute, Nathan Kline Institute, Child & Adolescent Psychiatry, New York University School of Medicine, New York, United States, [3]Center of Neuroscience Paris-Sud, CNRS-UMR8195, Neurobiology of Executive Function, Orsay, France

In fear conditioning, an initially neutral stimulus predicts an aversive stimulus at a fixed time interval. Evidence indicates that in associative learning, temporal relations between events are encoded from the early stages of the learning. Moreover, an increasing set of evidence suggests an involvement of the striatum and the prefrontal cortex in this timing memory. However the ontogenesis of this time encoding is poorly understood. We addressed this question using odor fear conditioning in rats. In this task, an odor is presented to the animal and after a fixed interval (30 seconds) a mild footshock is applied. This paradigm can be applied in very young animals as it does not require complex movements and olfaction is fully functional since birth. We first investigated the ontogenesis of interval timing at the behavioral level. For this we used an experimental setup developed and validated in the lab allowing the simultaneous recording of respiration, ultrasonic vocalizations (USVs) and freezing in rats. These parameters greatly improve the sensitivity of fear behavior assessment, thus increasing the probability of detecting transient anticipatory fear responses. Previous results in adults have shown that an anticipatory response develops after a few odor-shock pairings, characterized by a decrease in respiratory rhythm and an increase in USVs emission and freezing a few seconds before shock delivery. We investigated from what age this anticipatory response occurs in pups. Three ages were considered: 12-days-old (infants), 22-days-old (juveniles) and 80-days-old (adults). In parallel, 2-Deoxyglucose metabolic mapping was used to investigate learning-induced brain activation at these three ages mainly focusing on structures known to be involved in timing in adults: striatum and prefrontal cortex. Our results suggest that CS timing may emerge at weaning and is coincident with the functional maturation of prefrontal and striatal structures, although future studies are required to assess causation.

P1.177

A novel approach for the investigation of olfactory episodic memory in humans

Saive, A.-L. (Lyon)[1], Ravel, N. (Lyon)[1], Thévenet, M. (Lyon)[1], Garcia, S. (Lyon)[1], Royet, J.-P. (Lyon)[1], Plailly, J. (Lyon)[1]
[1]Lyon Neuroscience Research Center, Olfaction: from Coding to Memory, Lyon, France

Episodic memory is characterized as the vivid and conscious recollection of a personal event (What) in its spatial (Where) and contextual (Which context) environment. In humans, two historical methods are used to study episodic memory: the ecological method, which investigates autobiographical memory, and the laboratory-based method, which investigates recognition memory. Recently, McDermott et al. have underlined the interest to propose a new method to the study of human episodic memory, avoiding existing approaches' drawbacks and being both highly controlled and as ecological as

possible. Toward this end, we developed a novel behavioral design to investigate episodic memories triggered by odors.
In our approach, the three to-be remembered episodes were rich and close to real-life situations. They were made-up of three unfamiliar and unnamable odors (*What*), positioned at specific locations (*Where*) in a visual context, the picture of a landscape (*Which context*), and were freely encoded, one episode per day. After consolidation, the evaluation of the memory content accuracy was performed using recognition and retrieval tasks. We used odors as cues because they are known to be especially evocative reminders and difficult to identify, which limit the use of verbal processes.
The results demonstrated that the participants were highly competent at recognizing unfamiliar target odors, demonstrating the validity of using odors as recall cues. Moreover, the participants were able to retrieve the spatio-contextual environment of an episode cued by an odor.
To conclude, our protocol investigated the memory of rich olfactory episodes composed of unnamable odors located spatially within a visual context, allowing the controlled study of the encoding, the retention delay and the retrieval of complex episodes, as close as possible to real-life situations. This approach will also allow us to investigate the requirement of conscious recollection during the retrieval and therefore the episodic nature of these memories, and to explore the as-yet-unexplored neural bases of odor episodic memory.

P1.178

Role of the dorsal premotor cortex (PMd) and the lateral prefrontal cortex (LPFC) of macaques in action selection based on self-determined abstract behavioral goals

Hoshi, E. (Tokyo)[1], Hashimoto, M. (Tokyo)[1], Saga, Y. (Tokyo)[1,2], Tremblay, L. (Bron)[2], Tanji, J. (Sendai)[3]
[1]Tokyo Metropolitan Institute of Medical Science, Tokyo, Japan, [2]Centre de Neuroscience Cognitive, UMR5229 CNRS, Bron, France, [3]Tohoku University, Brain Science Center, Sendai, Japan

Accumulating evidence indicates that the frontal association cortex plays a crucial role in the volitional control of behavior. In the present study, we sought to reveal area-specific involvement of the LPFC and PMd in self-determined behavior. We particularly focused on the two major aspects of behavioral representations: the abstract behavioral goal and the action (Nakayama et al. 2008, Yamagata et al. 2009). We trained two monkeys to determine an abstract behavioral goal in a voluntary fashion based on a reward delivered after each action; the monkey were required to determine whether to reach toward the right or left target despite the physical absence. After a choice cue (with two potential targets) was presented at various locations on the screen, the monkeys specified a forthcoming movement (action) based on a behavioral goal associated with a reward, and they then reached toward a target. Through analysis of cellular activity recorded from the LPFC and the PMd, we obtained three major findings. 1) The abstract behavioral goal was more strongly represented in the LPFC than in the PMd during the period preceding the choice cue presentation. 2) After the appearance of the choice cue, neurons in the LPFC and PMd began to reflect the action (i.e., the actual direction of a reaching movement). 3) Toward the execution of reaching movements, the LPFC was characterized by activity reflecting the behavioral goal, whereas the PMd was by the activity reflecting the action. These results suggest that the LPFC plays a crucial role in determining the abstract behavioral goal based on the outcome, in specifying the action based on the goal, and in guiding the reaching movement based on the maintained representation of the goal, whereas the involvement of PMd in these processes is limited to retrieving a specified action and then preparing for and executing the reaching movement.

P1.179

Emotional resonance could drive empathic social decision making in Non-Human Primates

Ballesta, S. (Bron)[1], Pozzobon, M. (Bron)[1], Duhamel, J.-R. (Bron)[1]
[1]Centre de Neurosciences Cognitives, UMR 5229, Bron, France

Considering that Non-Human Primates (NHP) live in social groups which contribute to their individual survival, actively supporting social cohesion would seem to be an evolutionary relevant behavior. To do so, animals should be able to perceive others' emotional arousal in order to adopt appropriate social behaviors. Hence, we ask the following questions: do NHP take into account the wellbeing of other group members when making social decision? If yes how? We developed a social decision paradigm designed to study the cognitive encoding of the perception of others' emotional states. The monkeys sat face to face and made choices by manually touching one of two visual targets that were

virtually projected on a transparent touch-sensitive panel interposed between the two animals. Each target was associated with unique set of outcomes for the active monkey, the recipient monkey or nobody (a plastic container). Outcomes were defined as positive, i.e. a drop of apple juice, or negative, i.e. an air puff delivered close to the eyes. Because the use of animal models precludes direct assessment of personal perceptions, to infer the emotional state of the monkeys during social decisions and experience of their consequences, we recorded the gaze, pupil size, eye blink rate, oro-facial movement and skin temperature of both animals. The results show that, depending on the identity of their partner, monkeys can be prone or averse to procure rewards. They can also be more or less averse to inflict air puffs to his partners. Interestingly, day to day variations in choice behavior show that the more the active monkey chose to give apple juice, the more he avoided delivering air puffs to the recipient monkey. In other words, prosocial behavior is consistent across outcome valence, arguing for « supramodal » social motivation. Complementary results show that a set of emotional proxies displayed similar variations when the active monkey experienced an outcome and when he observed the recipient monkey experiencing the same outcome. In conclusion, we confirmed that monkeys take into account the wellbeing of others while making social decision and we argue for specialized neural mechanisms mediating emotional resonance and empathy-driven behavior.

P1.180

Modular structures of functional networks in true and false odor recognition memory

Meunier, D. (Lyon)[1], Fonlupt, P. (Bron)[2], Saive, A.-L. (Lyon)[1], Plailly, J. (Lyon)[1], Royet, J.-P. (Lyon)[1]
[1]*Centre de Recherche en Neurosciences de Lyon, Codage et Mémoire Olfactive, Lyon, France,*
[2]*Centre de Recherche en Neurosciences de Lyon, Dynamique Cérébrale et Cognition, Bron, France*

Complex network tools allow to study systems composed of nodes and edges. They have been widely applied to analysis of data coming from social, technology and biological sciences but also to neuroscience data, such as anatomical and resting-state functional networks (Bullmore and Sporns, 2009). Modular structure decomposition allows to study the existence of sub-networks whose components gathered in a same module are working tightly together, and weakly with components located in other modules. This processing is of interest for studying memory, a cognitive process known to be widely distributed. We propose a new method to study modular structure of task-related functional MRI networks. The modular structure is obtained directly from the correlation coefficients, retaining information about both signs and weights. This method allows to avoid both issues arising when applying an arbitrary threshold to obtain binary graphs from correlation matrices, and considering absolute values, if we consider anti-correlated signals correspond to systems that work in opposition to each other's.
The method is applied to data acquired during a yes-no odor recognition memory task performed by 16 young and 22 elderly adults (Royet et al., 2011). Four response types were previously explored: correct (Hit) and incorrect false alarms (FA) recognition, and correct (CR) and incorrect (Miss) rejection. We showed that 44 brain areas were differentially activated as a function of these response types and age groups. We here extract times series for 36 areas among them and calculate condition-based weighted correlation matrices (Dodel et al., 2005).
Using partition similarity-based statistics and posteriori statistical analyses, we show that
1) Several areas, including the hippocampus, and parahippocampal and middle temporal gyri, were more connected between them for Hit responses than for other response types in young adults;
2) Condition-based modular partitions were more homogeneous in young than elderly subjects. We also show that modularity was negatively correlated to memory scores and positively correlated to bias scores.
Bullmore, Sporns (2009) Nat Rev Neurosci 10:186-198.
Dodel et al. (2005) Philos Trans R Soc Lond Biol Sci, 360:921-935.
Royet et al. (2011) Front Hum Neurosci, 5:65.

P1.181
Top-down and bottom-up competition in auditory attention in human

Bidet-Caulet, A. (Bron)[1,2], Bottemanne, L. (Bron)[3], Bertrand, O. (Bron)[1,2]
[1]Centre de Recherches en Neurosciences de Lyon, Inserm U1028-CNRS UMR 5292, Bron, France, [2]Université Lyon 1, Lyon, France, [3]Institut des Sciences Cognitives, CNRS UMR 5229, Bron, France

Attention is the brain function by which we, voluntarily or not, improve the processing of specific information in our environment. The entry of information to the limited-capacity system is controlled by top-down (TD) and bottom-up (BU) processes. On one hand, TD attention enables the good performance of an on-going task by selecting relevant information. On the other hand, one's attention can be involuntarily captured by an unexpected salient stimulus and thus diverted from the previously on-going task. This BU form of attention is necessary to be aware of important events, though irrelevant to the on-going task (e.g. fire alarm). A good balance between BU and TD mechanisms is thus crucial to be task-efficient while being aware of our surrounding environment. BU and TD mechanisms of attention have been mostly explored in separated trials or experiments; really few studies have explored how these mechanisms dynamically interact.
We set up a new paradigm to assess both BU (attentional capture) and TD (anticipation) attention mechanisms, at the same time. We adapted a Posner paradigm using central visual cues and monaural auditory targets. To assess TD anticipation, visual cues could be either informative or uninformative. BU attentional capture was triggered by a binaural distracting sound (phone ring, doorbell…) played during the cue-target period, in 25% of the trials. Behaviorally, in the absence of distracting sound, informative cues compared to uninformative ones shortened RT. The presence of distracting sounds also shortened RT, suggesting an increase in arousal after distracting sound presentation. Scalp EEG was recorded from 18 young adults performing this detection task. In the absence of distracting sound, an enhanced early-CNV to informative cue was followed by a reduced P300 to target. N100 and early-P3a to distracting sounds were found to be reduced when the preceding cue was informative, suggesting that BU attentional capture becomes less effective with enhanced TD engagement. Finally, the impact of BU attentional capture by distracting sounds on target processing was revealed as a delayed latency of the N100 response to target sounds. These results provide crucial information on how BU and TD mechanisms interact and compete in the human brain.

P1.182
Tone detection in the ´continuous´ and ´rhythmic´ modes of sensory processing

Zoefel, B. (Magdeburg)[1], Heil, P. (Magdeburg)[1,2]
[1]Leibniz-Institute for Neurobiology, Auditory Learning & Speech, Magdeburg, Germany, [2]Center for Behavioral Brain Sciences, Magdeburg, Germany

Neuronal excitability fluctuates continuously, resulting in alternating brain states of high and low responsiveness to external stimulation. The instantaneous state of excitability is assumed to be reflected in EEG (electroencephalogram) phase. In line with this notion, the probability of detecting near-threshold stimuli in the visual and somatosensory systems has been shown to depend on the phase of spontaneous oscillations recorded in the EEG. In audition, recent studies reported a dependence of stimulus detection on phase, but only when neural oscillations are driven by background sounds far above threshold or by transcranial direct current stimulation. Also, phase entrainment to the rhythmic presentation of sounds has been associated with attentional selection. Here, we examined in humans whether detection of near-threshold sounds in quiet depends on EEG phase, presenting stimuli at regular and irregular intervals. When presented at irregular intervals, detection probability was independent of phase, consistent with the idea of a ´continuous mode´ of sensory processing (Schroeder and Lakatos, 2009). When presented at regular intervals, we observed phase entrainment, revealing an adjustment of the system to the regular stimulation, and consistent with a ´rhythmic mode´ of stimulus processing. Phase entrainment went in line with slightly, but

significantly, decreased reaction times. However, and in contrast to expectations in the literature, detection remained independent of phase at frequencies >= 1 Hz. These results indicate that an operation in the ´rhythmic mode´ does not necessarily go in line with a modulation of stimulus detection, but might rather reflect stimulus anticipation or motor preparation.
Finally, we show that phase extraction by common acausal algorithms results in an apparent phase entrainment and in a dependence of detection probability on δ-phase, similar to findings in the literature. We show that these effects are artifacts from ERP ´smearing´, resulting in phase distortion at stimulus onset. The study was supported by the Deutsche Forschungsgemeinschaft (SFB-TRR 31 A6).

P1.183

Maintaining vs enhancing: respective roles of striatum and hippocampus in sleep-related motor sequence memory consolidation

Albouy, G. (Montreal)[1], Fogel, S. (Montreal)[1], Laventure, S. (Montreal)[1], Patenaude-Veilleux, L. (Montreal)[1], Benali, H. (Paris)[2], Karni, A. (Haifa)[3], Carrier, J. (Montreal)[1], Robertson, E. (Boston)[4], Doyon, J. (Montreal)[1]
[1]Universite de Montreal, Montreal, Canada, [2]U.M.R.-S 678, I.N.S.E.R.M., U.P.M.C., Paris, France, [3]Laboratory for Functional Brain Imaging and Learning Research, Haifa, Israel, [4]Harvard Medical School and Beth Israel Deaconess Medical Center, Boston, United States

Activity in both striatum and hippocampus has been shown to play a crucial role in sleep-related motor sequence memory consolidation. The aim of this study was to specify their respective roles in consolidation processes using a spatial/motor manipulation of a motor sequence learning task and fMRI in young healthy individuals.
Using fMRI, cerebral activity of 55 young healthy volunteers was recorded during training on an explicit finger tapping sequence task. After training, subjects were tested either on a spatial (allocentric [ALLO]) or motor (egocentric [EGO]) representation of the sequence. Subjects were again divided in two groups according to whether they were allowed to take a 90-minute nap, or stayed in quiet wakefulness during this period. Subjects of the 4 groups were then retested on the representation they were tested on. Performance speed was measured using block duration.
A sleep-dependent enhancement in performance was observed for the allocentric representation whereas consolidation of the egocentric representation of the sequence did not depend on sleep. Brain imaging data showed that the allocentric representation was supported by activity in hippocampo-cortical networks whereas the egocentric representation recruited striato-motor circuits. Regression analyses suggest that activity in the striatum repressed the emergence of offline gains in performance in the ALLO, hippocampal-dependent, representation. In contrast, activity in the hippocampus promoted improvement in performance in the EGO representation, but only when sleep was allowed after training. Finally, while sleep-dependent enhancement was supported by increased connectivity between hippocampo-frontal areas in the ALLO representation, maintenance in performance in the EGO representation was supported by reinforced connectivity within striatal networks.
Our results suggest that activity in the hippocampus is specifically related to sleep-dependent offline mechanisms promoting performance enhancement, whereas activity in the striatum rather supports maintenance of the memory trace, a process that would be facilitated by sleep. Our study provides the first evidence of distinct roles for both the striatum and hippocampus during motor sequence memory consolidation.

P1.184

The anti-nociceptive and orexigenic activities of the aqueous extract of Pistacia lentiscus leaves

Mohamed Yeslem, O.M. (Marrakesh)[1], Mountassir, M. (Marrakesh)[2], Khalki, H. (Marrakesh)[2], Ferehane, H. (Marrakesh)[2], Benharref, A. (Marrakesh)[2], Chait, A. (Marrakesh)[2]
[1]Faculté des Sciences Semlalia, Université Cadi Ayyad, Lab. pharmacologie, neurobiologie et comportement, Marrakesh, Morocco, [2]Faculté des science Semlalia, Marrakesh, Morocco

Pistacia lentiscus is an evergreen shrub of the family Anacardiaceae, can reach 3 m in height, grows wild in arid areas and is characteristic of Mediterranean countries. The aerial part hastraditionally been used as a stimulant, for its diuretic properties, and to treat hypertension, coughs, sore throats, eczema, stomach aches, kidney stones and jaundice.The aim of this work, is to study the anti-

nociceptive and orexigenic activities of the leavesof Moroccan P. lentiscus. Male Swiss mouse, weight between 24g and 30 g, and male Sprague-dawley rats weighting between 180-230g are used in this study. All tested animalshave receiving the aqueous extract of Pistacia lentiscus leaves. The plant was carried from Rfala, locality of AZILAL Moroccan town. Results have shown that no toxicity was observed at the used doses. No antalgic effect wasobtained in the hot plat test. In the writhing test, the extract of this plant has shown asignificant antalgic effect. In food intake test, the extract decreases quantity of consumedaliments in tested rats. The aqueous extract of P. Lentiscus, has demonstrated a significant antalgic and anorexigenicactivities in mouse and rat.

P1.185

Social hierarchies in the brain: learning the value of others through reinforcement

Ligneul, R. (Bron)[1], Girard, R. (Bron)[1], Scheiber, C. (Bron)[1], Dreher, J.-C. (Bron)[1]
[1]*ISC - Centre de Neuroscience Cognitive, Bron, France*

Introduction: Performance-based competition is a widespread phenomenon, which has a central role in the emergence and the maintenance of social hierarchies. Here, we hypothesize that motivation to compete against other individuals is encoded in the brain similarly to other primary motivations, recruiting the brain valuation system. Furthermore, we hypothesized that optimizing strategic interactions in such contexts requires adapting to the relative rank of our rivals; a process which may be implemented as a reinforcement learning problem.
Methods: To test the hypotheses mentioned above, we designed an optimized fMRI study in which each subject played a competitive perceptual task against three opponents; in an additional condition, subjects played a cooperative interaction task, matched for visual stimulation. Blood samples were also taken to assess the circulating levels of testosterone and cortisol.
Results: The results of our model-based analysis clearly demonstrated that the ventral striatum, the ventromedial prefrontal cortex and the anterior cingulate cortex were more activated by victories than by defeats. Moreover, while BOLD responses in the caudate nucleus increased with the rank of the opponent in case of victories -possibly reflecting a prediction error signal, the lateral orbitofrontal cortex seemed sensitive to the rank in case of defeats, reflecting the negative emotions elicited by losing against a low-ranked player.
Conclusion: Applying a computational model to our fMRI data, we showed that a brain network previously associated with valuation and reinforcement-learning in nonsocial contexts was also recruited to learn the value of others in a social competition task. Although this neurocomputational approach is not widely developed in social decision making, our findings demonstrate its efficiency to enlighten a fundamental mechanism underlying social valuation. Moreover, our study provides a promising starting point to investigate the well-known interactions between hormonal and behavioral responses to social competition.

P1.186

Effects of perinatal exposure to waterborne fluoxetine on memory processing in the cuttlefish Sepia officinalis

Boulouard, M. (Caen)[1], Bellanger, C. (Caen)[1], Darmaillacq, A.-S. (Caen)[1], Dickel, L. (Caen)[1], Di Poi, C. (Caen)[1]
[1]*Normandie Universités Université de Caen Basse-Normandie, GMPc EA 4259, Caen, France*

Recent ecotoxicological studies highlight the increasing presence of pharmaceuticals

discharged in the aquatic environment. Amongst them is the antidepressant fluoxetine (FLX), a selective serotonin reuptake inhibitor, primarily indicated for treatment of depression. The effect of chronic exposure to FLX was evaluated on memory processing in 1 month-cuttlefish Sepia officinalis. Three groups of new-borns were reared in different conditions: one control group (no FLX) and two groups exposed to environmental concentrations of FLX (1 and 100 ng/L) from 15 days pre-hatching to 1-month post-hatching. Acquisition and retention performances were assessed using the ´prawn-in the-tube´ procedure. Perinatal exposure to fluoxetine led to significant changes in memory processing of the animals. The lowest observed effect concentration of this antidepressant on learning and retention was 1 ng/L which is under the range of environmental contamination. Cuttlefish exposed at low FLX dose had impaired acquisition capabilities and animals exposed at high FLX concentration displayed a deficit of memory retention compared to the control group that had non impaired initial acquisition and retention performances. The results subsequently suggested that FLX-induced changes in cognitive capacities could potentially lead to inappropriate predatory behaviors in the natural environment. The study provides the basis for future studies on how pharmaceutical contaminants disrupt cognition in ecologically and economically relevant marine invertebrates.

P1.187
Prefrontal parvalbumin-expressing interneurons control fear expression

Courtin, J. (Bordeaux)[1], Chaudun, F. (Bordeaux)[1], Rozeske, R. (Bordeaux)[1], Gonzalez-Campo, C. (Bordeaux)[1], Bienvenu, T. (Bordeaux)[1], Wurtz, H. (Bordeaux)[1], Herry, C. (Bordeaux)[1]
[1]*Neurocentre Magendie, U862, Circuits neuronaux des apprentissages associatifs, Bordeaux, France*

Accumulating evidence indicate that the medial prefrontal cortex is necessary for conditioned fear expression. Indeed, whereas inactivation of the prelimbic area prevents fear expression, its electrical stimulation facilitates conditioned fear responses. It is likely that neuronal activity occurring during fear expression depends on specific interactions between prefrontal excitatory neurons and inhibitory interneurons. However the precise role played by prefrontal excitatory and inhibitory circuits in mediating fear expression is still largely unknown. To address this question, we used single unit and field potential recordings combined with optogenetics in behaving mice submitted to auditory fear conditioning. Our results indicate that tone-evoked inhibition in a subpopulation of prefrontal PV-expressing interneurons inversely correlates with fear behavior and coincides with an increase of neuronal excitability in prefrontal principal neurons. Furthermore, targeted optogenetic inhibition or excitation of prefrontal PV-expressing interneurons during conditioned tone presentations respectively enhanced or decreased fear expression. Interestingly, inhibition of PV-expressing interneurons during conditioned tone presentations or by optogenetic means was associated with theta phase resetting of prefrontal field potentials that synchronizes neuronal activity in prefrontal projection neurons during fear expression. Our results identify two distinct and coordinated neuronal mechanisms mediated by prefrontal fast spiking, PV-expressing interneurons, enhancing the excitability and neuronal synchronization of prefrontal principal neurons to gate fear expression.

P1.188
Functional connectivity of neural circuits involved in cocaine addiction

Simonnet, A. (Bordeaux)[1], Courtin, J. (Bordeaux)[1], Renault, P. (Bordeaux)[1], Fiancette, J.-F. (Bordeaux)[1], Herry, C. (Bordeaux)[1], Deroche, V. (Bordeaux)[1]
[1]*Université Bordeaux Segalen, Neurocentre Magendie, INSERM U.862, Bordeaux, France*

Cocaine addiction is a severe psychiatric condition defined as a compulsive drug taking despite severe negative consequences for the users. In our laboratory, we developed a rat model that closely mimics the behavior normally observed in a small proportion of human drug users. Accordingly, 15-20% of the rats displayed the behavioral hallmarks of addiction after a prolonged drug intake. This animal model provides excellent face and predictive validities and is a crucial prerequisite for studying pathophysiological mechanisms of addictive behavior. From an functional standpoint, a number of evidence suggest that distinct, but interconnected circuits, including the nucleus accumbens (NAc), the medial prefrontal cortex (mPFC) and basolateral amygdala (BLA) are at the core of pathological incentive processes and difficulties to control drug taking. However the functional interactions that occur between these neuronal structures during cocaine addiction are still poorly understood. To further address this question, we performed, simultaneous single unit and local field potential recordings in NAc, mPFC and BLA in rats behaviorally characterized as addict or non-addict animals after 3 months of cocaine self-administration. Our results indicate a reduction of the functional

connectivity between these neuronal structures in addict animals compared to non-addict animals as assessed using LFP coherence analysis and long-range neuronal correlations. These preliminary results support the notion that cocaine addiction is associated with specific functional alteration of dedicated neuronal circuits and could further provide, in part, an explanation to the individual differences in vulnerability to cocaine addiction.

P1.189

Zif268/egr1 gene controls the selection, maturation and functional integration of adult hippocampal newborn neurons by learning

Gros, A. (Orsay)[1,2], Bruel-Jungerman, E. (Orsay)[1,2,3], Rochefort, C. (Orsay)[1,2,4], Laroche, S. (Orsay)[1,2], Veyrac, A. (Orsay)[1,2]
[1]CNRS, Centre de Neurosciences Paris-Sud, UMR 8195, Orsay, France, [2]Université Paris-Sud, UMR 8195, Orsay, France, [3]INSERM UMR S975, CHU Pitié-Salpêtrière, UPMC, Paris, France, [4]CNRS UMR 7102, UPMC, Paris, France

New neurons are continuously added to the dentate gyrus (DG) of the adult mammalian brain. During a critical period of a few weeks after their birth while newborn neurons progressively mature, a restricted fraction is competitively selected to survive in an experience-dependant manner, a condition for their contribution to the storage of new memories. The mechanisms that control critical stages of experience-dependant functional incorporation of adult newborn neurons remain largely unknown. Here, we identify a novel transcriptional regulator of the functional integration of newborn neurons, the inducible immediate early gene zif268/egr1. We show that newborn neurons in zif268-knockout mice undergo accelerated death during the critical period of 2-3 weeks of their birth and exhibit deficient neurochemical and morphological maturation including reduced GluR1 expression, increased NKCC1/KCC2b chloride co-transporter ratio, altered dendritic development and marked spines growth defect. Investigating responsiveness of newborn neurons to activity-dependent expression of zif268 in learning, we demonstrate that in the absence of zif268 the occurrence of spatial learning during this critical period fails to recruit newborn neurons and to promote their survival, and leads to impaired long-term memory. This study reveals a previously unknown mechanism for the control of the selection, functional maturation and experience-dependent recruitment of DG newborn neurons that depends on the inducible immediate early gene zif268, processes that are critical for their contribution to hippocampal-dependent long-term memory. Supported by ANR-2010-BLAN-1413-01 to SL.

P1.190

Anxiety-like behavior induced by repetitive dural stimulations in the rat

Jacquot, F. (Clermont Ferrand)[1], Dallel, R. (Clermont Ferrand)[1], Monconduit, L. (Clermont Ferrand)[1], Artola, A. (Clermont Ferrand)[1], Luccarini, P. (Clermont Ferrand)[1]
[1]Faculté de chirurgie dentaire, INSERM U1107, Neurobiologie de la douleur trigéminale, Clermont Ferrand, France

Migraine is a neuro-vascular disorder with a major impact on productivity due to chronic pain but also from the co-morbid psychiatric disorders such as anxiety. In order to understanding the relationship ongoing between migraine and anxiety we use a behavioral and immunocytochemical approaches on a rat model of repetitive noxious meningeal stimulations. The rats received 1 to 4 injections (one per day spaced within 2 days) of inflammatory soup solution (IS) (10µL of Bradykinin 0.2mM; Histamin 2mM; Prostaglandin 2mM; Serotonin 2mM in HEPES ph 5,5) or of HEPES (vehicle). Each day, mechanical facial allodynia is verified with Von Frey filaments with *VOS* method. On days without injection, anxiety-like behavior is tested with elevated plus maze (EPM) or light/dark box (LDB) tests. Then, on days without injection, we used p-ERK as a neuronal activation marker to identify central structures involved in the behavioral anxiety-like responses observed.
In EPM test at the day after injection, for the rat received four injections of IS, the results show an increase of the time spent in closed arms ($n=12$; $p< 0.01$) and a decrease of the time spent in open arms ($n=12$; $p< 0.01$). In LDB at the day after injection, for the rat received four injections of IS, a decrease of first entries in dark side ($n=12$; $p< 0.001$) and time spent in light side ($n=12$; $p< 0.01$). These findings suggest that repeated stimulations of meninges are necessary to observe an anxiety-like behavior in rat.
Immunocytochemical study reveal, in cortical structures, that an increase of activity in insular cortex ($n=5$; $p< 0.01$) with the repetitive injection of IS whereas a depression of cingulate cortex ($n=5$; $p< 0.001$). In the same time, we observe in some amygdala nucleus an increase of their activity for the rat

received four injections, such as lateral nucleus (n=5; p< 0.05) and especially medial nucleus (n=5; p< 0.001) but not in central nucleus (n=5; ns).
These results suggest an influence of migraine crises on animals behavior and it is necessary to repeat the injections of IS to lead the appearance of anxiety-like behavior in the rat.

P1.191

RMS contrast normalization induces changes in the retinotopic processing of spatial frequency in scenes

Ramanoel, S. (Grenoble)[1,2], Kauffmann, L. (Grenoble)[1], Guyader, N. (Grenoble)[3], Chauvin, A. (Grenoble)[1], Pichat, C. (Grenoble)[1], Dojat, M. (La Tronche)[2], Peyrin, C. (Grenoble)[1]
[1]Laboratoire de Psychologie et Neurocognition, CNRS UMR5105, Grenoble, France, [2]Grenoble Institute of Neuroscience Inserm U 836, La Tronche, France, [3]Grenoble-Image-Parole-Signal-Automatique, CNRS UMR 5216, Grenoble, France

Since there is considerable evidence suggesting that visual perception is based on spatial frequency (SF) processing, a growing number of studies investigate the cerebral regions and mechanisms involved in the processing of low and high SF (LSF and HSF) information in complex visual stimuli (such as scenes). LSF and HSF stimuli are created using low- and high-pass filters that respectively attenuates signals with frequencies higher and lower than a cutoff frequency. The contrast (i.e. the difference in luminance values) is reduced in HSF relative to LSF images. Thus, recent fMRI studies normalized root mean square (RMS) contrast in image (the standard deviation of luminance values) in order to avoid that differential cortical activations in LSF and HSF processing might be due to contrast differences. In the present fMRI study, we investigated whether RMS contrast normalization induced change in retinotopic processing of SF during scene categorization. For this purpose, participants performed a categorization task using large black and white photographs of natural scenes (indoors vs. outdoors) filtered in LSF, HSF and non-filtered (NF) scenes, in eight block-designed functional scans. In four functional scans, both the mean luminance and the RMS contrast of LSF, HSF and NF scenes were equalized, while in the other four functional scans only the mean luminance was equalized. When RMS contrast was not normalized (mean luminance equalization only), results showed that LSF (relative to HSF) scenes elicited activation in the anterior half of the calcarine fissures linked to the peripheral visual field, whereas HSF (relative to LSF) scenes elicited activation in the posterior part of the occipital lobes, which are linked to the fovea, according to the retinotopic property of visual areas. However, RMS contrast normalization drastically increased activation for HSF scenes only, such as no significant activation was obtained for LSF scenes compared to HSF scenes. Our study suggests that RMS contrast normalization should be used with caution when investigating the neural basis of SF processing in retinotopic areas.

P1.192

Effect of action verbs on the performance of a complex motor movement

Rabahi, T. (Villeurbanne)[1], Fargier, P. (Villeurbanne)[2], Rifai Sarraj, A. (Beyrouth)[3], Clouzeau, C. (Limonest)[4], Massarelli, R. (Villeurbanne)[2]
[1]Univ. Claude Bernard Lyon I, Centre de recherche et d'innovation sur le sport, Bât. R. Dubois- EA 647, Villeurbanne, France, [2]Université Claude Bernard Lyon 1, Centre de Recherche et d'Innovation sur le Sport (CRIS, EA-647), Villeurbanne, France, [3]Université Libanaise, Département de Physiothérapie, Beyrouth, Lebanon, [4]Institut Supérieur d'Ostéopathie, Unité de Recherche, Limonest, France

The relation between language and motor action has been approached by studying the possible effect of action verbs upon the performance of a complex motor movement, the classical squat vertical jump (SVJ), in naive male subjects. The results showed a statistically significant improvement of the height of the jump after loudly or silently pronouncing, hearing or reading the verb *saute* (jump in French). Action verbs specific for other actions (e.g.: *pince* = pinch, *lèche* = lick) or non-specific (*bouge* = move) showed no or little effect. A meaningless verb for the French subjects (*tiáo* = jump in Chinese) showed no effect as did *rêve* (dream). The verb *gagne* (win) stimulated instead the SVJ height indicating the possible influence of emotional stimuli, as it might have been expected. Surprisingly the pronunciation of *perds* (lose) also improved significantly the height of the SVJs.
Finally, the improving effect of the specific action verb *saute* was similar to that obtained after kinaesthetic imagery of the SVJ and after mental subtraction of two digits numbers from three digits ones, possibly because of the intervention of language in calculus. As a possible conclusion, it appears that the effects of the specific action verb saute did not appear to be exclusive for the enhancement of the SVJs as other verbs unrelated to the action itself and even calculus might be effective in improving the height of the jump.

P1.193
Distinct pathway-specific prefrontal projection neurons control fear expression

Chaudun, F. (Bordeaux)[1], Courtin, J. (Bordeaux)[1], Herry, C. (Bordeaux)[1]
[1]*Université Segalen Bordeaux, Neurocentre Magendie - INSERM U.862, Circuits neuronaux des apprentissages associatifs, Bordeaux, France*

Numerous studies indicate that the dorsal medial prefrontal cortex (dmPFC) is involved in the expression of conditioned fear responses. In particular, it has been shown that inactivation of the prelimbic area decreases fear expression whereas its electrical activation facilitates conditioned fear responses. Furthermore, the activity of putative dmPFC pyramidal neurons (PN) is highly correlated with fear expression as measured with freezing responses. These data strongly suggest that changes in the activity of PN directly regulate the expression of conditioned fear responses. Interestingly, anatomical evidence indicate that dmPFC output neurons project to both the basolateral amygdala (BLA) and the ventral or dorsal periaqueductal gray (vlPAG/dlPAG) where they could potentially modulate conditioned fear responses. However, the specific contribution of these distinct neuronal pathways during fear expression is still largely unknown. To address this question, we used single unit and local field potential recordings combined with specific optogenetic manipulations of dmPFC-BLA or dmPFC-PAG neuronal circuits in behaving mice submitted to auditory fear conditioning. Our preliminary results indicate that dmPFC output PN can modulate freezing behavior through at least two distinct neuronal pathways. Specific optogenetics activation of dmPFC PN targeting the dlPAG decreases conditioned fear expression whereas optical activation of the dmPFC-vlPAG pathway produces the opposite effect. Furthermore, specific optical activation of BLA-projecting dmPFC PN increased conditioned fear expression. Our data indicate that fear behavior can be tightly regulated by at least two distinct dmPFC output pathways and further suggest that under some circumstances, dmPFC-PAG pathway can bypass the BLA to control fear responses.

P1.194
Metformin alters energy metabolism by reducing both food intake and energy expenditure

Rouquet, T. (Marseille)[1], Pierre, C. (Marseille)[2], Roux, J. (Marseille)[2], Troadec, J.-D. (Marseille)[1], Bariohay, B. (Marseille)[2], Lebrun, B. (Marseille)[1]

[1]Faculté Saint Jérôme - Univ. Paul Cézanne, PPSN, Marseille, France, [2]Biomeostasis, Marseille, France

Since the late 1950s, metformin, a biguanide derivate, is widely used to treat hyperglycemia in individuals with type 2 diabetes. The main effects of metformin are to reduce hepatic glucose production and to improve peripheral sensitivity to insulin. This drug has also been shown to enhance glucose uptake in the muscles, and to reduce plasma levels of triglycerides and non-esterified fatty acids. Recent evidences indicate that metformin is able to exert an anorectic effect on rodents in free-feeding condition, but the mechanisms of action remain unclear. To investigate mechanisms of action of metformin on food intake and energy metabolism, we performed acute treatment *(per os)* with metformin at a dose of 300mg/kg, prior to the dark phase, on C57Bl/6J adult mice housed in metabolic cages (Physiocages system). Their food intake, energy expenditure and spontaneous physical activity were recorded for the subsequent 48h. We showed that metformin induced a statistically significant decrease in cumulative food intake of C57Bl/6J mice. This reduction was significant from 3h to 12h post-treatment (50% and 30% respectively), when compared to control mice. Meal-pattern analysis revealed that the anorectic effect of metformin resulted from a significant decrease in meal size without affecting the meal frequency, suggesting that the anorectic effects of metformin does not result from a non specific adverse effect. Interestingly, our results also indicate, that in parallel to its anorectic effect, metformin induced a transient but significant decrease in energy expenditure during the 6h post-treatment of the dark phase, while the total spontaneous activity was not altered. We also showed that, compared to control animals, metformin-treated mice displayed a decrease in RQ (respiratory quotient). Moreover, we are currently investigating the impact of metformin on neuronal networks controlling food intake and body weight. To this goal, we are looking to the effect of peripheral and central metformin injections on neuronal activation using c-Fos protein as a marker. Taken together, our results support the view that in addition to its peripheral effects on glucose metabolism, metformin also interacts with the central control of food intake and energy expenditure.

P1.195

Action-related neuronal activity in the globus pallidus of macaques reflects multiple aspects of goal-directed behavior

Yosuke, S. (Bron)[1,2], Hashimoto, M. (Tokyo)[2], Tremblay, L. (Bron)[1], Tanji, J. (Sendai)[3], Hoshi, E. (Tokyo)[2]
[1]CNRS, UMR5229, Centre de Neuroscience Cognitive, Bron, France, [2]Tokyo Metropolitan Institute of Medical Science, Frontal Lobe Project, Tokyo, Japan, [3]Tohoku University, Brain Science Center, Sendai, Japan

Although the basal ganglia (BG) have been implicated in the generation of action, it is becoming evident that the BG are also involved in processing information relevant to aspects of cognitive behavior. The BG interconnect across synapses with the prefrontal cortex and higher-order motor areas in the frontal cortex. BG dysfunction is also known to cause cognitive function deficits as well as movement disorders. In the present study, we examined the role of the BG in cognitive control of self-determined behavior. We particularly focused on the two major aspects of behavioral representations: the abstract behavioral goal and the action (Nakayama et al. 2008, Yamagata et al. 2009). We trained two monkeys (*Macaca fuscata*) to determine an abstract behavioral goal without any instructions. The monkeys were required to select the goal based on a reward delivered after each action. After a choice cue (with two potential targets) was presented at various locations on the screen, the monkeys specified a forthcoming action based on a behavioral goal associated with a reward, and they then reached toward a target. We asked the monkeys to perform two types of tasks, each of which required them to achieve a behavioral goal belonging to either of two dimensions. In the spatial-goal task, the behavioral goal was to reach toward the right or left side of the two potential targets. In the object-goal task, the behavioral goal was to reach toward a circular or triangular target. We recorded neuronal activity in the external and internal segments of the globus pallidus (GP) while the monkeys performed the two behavioral tasks. By analyzing the activity of GP neurons during the execution of action, we found that GP neurons reflect the direction of movements as well as the goals of behavior. These

results suggest that the GP represents multiple levels of signals related to goal directed behavior during execution of action.

P1.196

Effect of the visualization of geometrical shapes on the performance of a complex motor act

Kabbara, M. (Villeurbanne)[1], Sarraj, A.R. (Beyrouth)[2], Rabahi, T. (Villeurbanne)[1], Fargier, P. (Villeurbanne)[1], Khalil, M. (Beyrouth)[2], Massarelli, R. (Villeurbanne)[1]
[1]Université Claude Bernard Lyon 1, Centre de Recherche et d'Innovation sur le Sport (CRIS, EA 647), Villeurbanne, France, [2]Université Libanaise, Département de Physiothérapie, Campus Rafic Hariri, Hadath Cedex, Beyrouth, Lebanon

Several studies have shown that the observation of a motor action activates cerebral cortical motor areas involved in the execution of the action itself and further sustained the suggestion that the visualisation of a motor act might improve the same movement in the observer. The hypothesis was advanced that the stimulus in the observation of a motor action might be the 'movement' rather than the human figure realising the action. The present experiments were devised to test whether simple geometrical shapes might have an influence on the performance of a complex motor act the *squat vertical jump* (SVJ). Two series of 2D objects ("*circle, square, cylinder*" and "*heart, ring, star*") were shown to 3 groups of naïve male (M) (n=15 for each group) and 3 groups of naïve females (F) subjects (same cohorts). Group 1 visualized immobile shapes while groups group 2 saw forms moving up-bottom and group 3 bottom-up. Groups M1, M2, F1, and F2 showed no significant improvement in the heights of jump. In group M3 the series *circle* improved significantly the height of the SVJ (+2.1 cm, P< 0.003), while the series *heart* showed no significance (+1.0 cm, p=0.3). The other groups (fixed or up-bottom) the height did not change (p=0.2-1.0). Group F3 significantly improved its SVJ performance after both series of images (series *circle*, +1.2 cm, p=0.006; series *heart* +1.1 cm, p=0.006). The results show that the performance of a complex motor action, the SVJ, might have been improved by the simple visualisation of 2D geometrical shapes, only if these were in congruent movement (bottom-up). Female subjects apparently performed better than the male counterparts as they improved similarly the height of the SVJ with both experimental series of shapes ("*circle, square, cylinder*" and "*heart, ring, star*"), again only when these were moving bottom-up.

P1.197

Characterization of aversive-related neuronal activity during the performance of imperative/choice task in non-human primate ventral striatum

Richard, A. (Bron)[1], Saga, Y. (Bron)[1,2], Hoshi, E. (Tokyo)[2], Sgambato-Faure, V. (Bron)[1], Tremblay, L. (Bron)[1]
[1]Institut des Sciences Cognitives, CNRS UMR 5229, Bron, France, [2]Tokyo Metropolitan Institute of Medical Science, Frontal Lobe Project, Tokyo, Japan

The ventral striatum (VS), which belongs to a limbic circuit, is implicated in reward processing. Recently, human imaging studies and animal pharmacological studies have suggested that the VS is also involved in aversive information processing. In the present study, we investigated how single VS neurons represent aversive information. We trained two monkeys to perform imperative and choice tasks that contain appetitive and aversive predictable cues and outcomes. In the imperative task, a conditioned stimulus (CS) was presented as a cue at the right or left side on the screen monitor for 1 sec. Each CS was associated with either appetitive outcome (juice) or aversive outcome (air puff). Following the CS presentation, delay period ensued for 1.5-2 sec. Subsequently, two green squares

that were potential targets appeared on the right and left sides of the screen. If monkeys reached with their hand a target located at the same side as the cue (approach), monkeys obtained an outcome associated with the CS after a second delay. If they touched a target located opposite to the CS (avoidance), they didn't receive any outcomes. The CSs predicting appetitive outcome were presented in 60% of trials, and in the remaining 40% of trials the CSs predicting aversive outcome were presented. In a great majority of trials, the monkeys approached the appetitive outcome and avoided the aversive one. In the choice task, the temporal sequence of task was identical to that of the imperative task, except that both appetitive and aversive CSs were presented simultaneously. Under this choice condition, the monkeys approached target for appetitive outcome with a high rate (>95%). We recorded neuronal activities from anterior VS while the monkeys performed the tasks. We found that the VS neurons showed a variety of activities (to CSs or outcomes) not only related to the appetitive stimuli, but also to aversive ones. Interestingly, neurons responding to aversive stimuli were largely different from neurons responding to appetitive stimuli. Moreover, the activities of neurons in each group differed depending on the two tasks. These results show that the VS represent aversive as well as appetitive information and that behavioral context influences activity of the VS neurons.

P1.198

Behavioral study of confidence judgments and checking in monkeys

Stoll, F. (Bron)[1,2], Procyk, E. (Bron)[1,2]
[1]*Inserm U846, Stem Cell and Brain Research Institute, Bron, France,* [2]*Univ. Claude Bernard Lyon I, Lyon, France*

When we make a decision, we may feel introspectively more or less confident that this decision was the correct one, even before receiving feedback. If uncertainty is too high, we may prefer collecting additional information before confirming or revising our choice. These confidence judgments are self-assessments of the quality one´s own performance, and are a crucial aspect of metacognition. Alteration of checking or verification behaviour might lead to obsessive and compulsive behaviours, but the underlying neurobiological processes of metacognition are poorly understood.
Here, we used 2 new protocols designed to investigate behavioural and neurophysiological aspects of confidence judgments and checking in monkeys.
In experiment 1, two monkeys had to perform a 2 forced-choice categorization task, and then were asked to either ***confirm*** or ***review*** the trial. By using the ***review*** option, monkeys collected more information about the stimulus and were able to either repeat or revise their first categorization choice. This procedure allowed us not only to assess the subjective confidence of animals, but also to study how these judgments are used in order to adapt their behaviour.
In experiment 2, three monkeys were offered the opportunity to track the appearance of a bonus reward in parallel to the execution of a categorization task. By touching a second lever at the onset of a trial, monkeys could check the evolution of a gauge which indicated approximately when the bonus would be available. If the monkey checked at full gauge, he immediately received a bonus reward, but a partially full gauge had no consequence.
Behaviour during learning showed that monkeys were to some extent able to use their confidence and adapt their behaviour accordingly. In the second protocol, we also elicited an adaptive checking behaviour modulated by the distance to the bonus reward.

P1.199

Reward risk coding in the human orbitofrontal cortex: an intracranial EEG recording study

Li, Y. (Lyon)[1,2], Vanni-Mercier, G. (Lyon)[1,2], Mauguière, F. (Lyon)[2,3], Dreher, J.-C. (Lyon)[1,2]
[1]*CNRS, UMR 5229, Institut des Sciences Cognitives, Lyon, France,* [2]*Université Lyon 1, Lyon, France,* [3]*Neurological Hospital, Lyon, France*

The risk of an outcome measures the unpredictability of the outcome (maximal for reward probability=0.5). Yet, it is still unknown whether population of neurons recorded with local field potentials (LFPs) explicitly encode risk information in the human orbitofrontal cortex (OFC). To investigate this question, six epilepsy patients were stereotaxically implanted with depth electrodes in the OFC (0.8 mm multicontact cylinders, 2 mm long), as part of a presurgical evaluation. Unbeknownst to the subjects, five types of slot machines with different reward probabilities (P=0; 0.25; 0.5; 0.75 and 1) were presented randomly. The subjects' task was to estimate on each trial the reward probability of each slot machine at the time of its presentation, based upon all the previous outcomes of the slot machine until this trial. The trials were self-paced and were composed of four phases:

(1) Presentation of slot machines (S1);
(2) Delay period phase during which the 3 spinners rolled around successively (500 ms each);
(3) Outcome phase during which the 3d spinner stopped (0.5 s);
(4) Reward/No reward delivery (bill presented or not for 1 s).
At the behavioral level, the RT followed an inverted U-curve relationship with reward probability ($F(4,20) = 8.15$, $p < 0.001$). Moreover, a main effect of reward probability (P) was observed on the percent of correct estimates of winning probability ($F(4,20) = 69.18$, $p < 0.001$), showing a U-shape relationship with reward probability. At the LFPs level, regardless of subsequent winning or not, a robust reward risk-sensitive OFC component started from the late phase of reward expectation, peaking at 25.47 ± 40.85 ms (unrewarded trials) and 89.11 ± 48.93 ms (rewarded trials), respectively. These signals emerged from the medial and lateral OFC. Importantly, the amplitudes of these ERPs followed an inverted U-curve relationship with reward probability, being maximal when reward risk is highest (P=0.5), and minimal when that is lowest (P=0 and P=1). This study characterizes the temporal dynamics of reward risk processing in the human OFC, and explains the fundamental neural mechanisms underlying the role of this brain region in a number of functions, such as attention-based learning.

P1.200

The ventral midline thalamus contributes to strategy-shifting in a memory task engaging cortico-hippocampal interactions

Cholvin, T. (Strasbourg)[1], Loureiro, M. (Strasbourg)[1], Cassel, R. (Strasbourg)[1], Cosquer, B. (Strasbourg)[1], Geiger, K. (Strasbourg)[1], Kelche, C. (Strasbourg)[1], Pereira de Vasconcelos, A. (Strasbourg)[1], Cassel, J.-C. (Strasbourg)[1]
[1]*Université de Strasbourg, UMR 7364 - LNCA, Strasbourg, France*

Electrophysiological and neuroanatomical evidence for reciprocal connections with the medial prefrontal cortex (mPFC) and the hippocampus make the reuniens and rhomboid (ReRh) thalamic nuclei a putatively major functional link for regulations of cortico-hippocampal interactions. In a first experiment using a new water escape device for rodents, the double-H maze (e.g. Cassel et al., Behav Brain Res 230, 2012, 33-342), we demonstrate in rats that a bilateral muscimol (MSCI) inactivation (0.70 vs. 0.26 and 0 nM) of the mPFC or the dorsal hippocampus (dHip) induces major deficits in a strategy shifting/spatial memory retrieval task during a misleading probe trial occurring 24h after the end of acquisition. By way of comparison, only dHip inactivation impaired recall at a 24h-delay in a classical spatial memory task in a Morris water maze. In the second experiment, we show that ReRh inactivation using 0.70 nM MSCI, which reduced performance without obliterating memory retrieval in the water maze, produces an as large strategy shifting/memory retrieval deficit as mPFC or dHip inactivation in the double-H maze. Thus, behavioral adaptations to task contingency modifications requiring a shift from a response memory towards a memory for place might operate in a distributed circuit encompassing the mPFC (as the potential set-shifting structure), the hippocampus (as the spatial memory substrate), and the ventral midline thalamus, and therein the ReRh (as the coordinator of this processing). The results of the current experiments provide a significant extension of our understanding of the involvement of ventral midline thalamic nuclei in cognitive processes: they point to a role of the ReRh in the regulation of dynamic interactions between the mPFC and the hippocampus and further elucidate the functional connectivity underlying behavioral flexibility.

P1.201

A behavioral characterization of executive functions in the dystrophin-deficient *mdx* mouse model of Duchenne muscular dystrophy

Chaussenot, R. (Orsay)[1], Laroche, S. (Orsay)[1], Vaillend, C. (Orsay)[1]
[1]*Université Paris-Sud, Centre de Neurosciences Paris Sud (CNPS) - CNRS UMR 8195, Orsay, France*

Duchenne muscular dystrophy (DMD) is associated with cognitive deficits partly attributed to the loss of the cytoskeletal protein, dystrophin (Dp427). Indeed, beyond its expression in muscles, Dp427 is normally associated with $GABA_A$ receptors in the postsynaptic densities of central inhibitory synapses in brain regions involved in cognitive functions, such as in cortex, hippocampus and cerebellum. Recent clinical studies pointed to specific deficits in attention, working memory and adaptive behaviors, which could be detected at a subclinical level even in patients with normal IQ, suggesting that defective executive functions could be a basis of the cognitive impairment in DMD. To test the

current hypothesis that Dp427 loss may be sufficient to induce mild deficits in executive functions, we undertook the first behavioral study of executive functions in Dp427-deficient *mdx* mice.
First, we showed that auditory brainstem responses and the modulation of the startle reflex by auditory cues were not impaired in *mdx* mice, suggesting that gating of sensory inputs was largely unaffected. However, acquisition of auditory-cued fear conditioning was deficient, suggesting an amygdala-dependent impairment. Performance was not modulated by random presentation of visual distractors and the deficit could be progressively compensated by repeated presentations of the conditioned stimulus. This, however, was associated with deficits in long-term memory retention that hindered reliable analyses of behavioral flexibility in this context. We therefore investigated the expression of flexible behavioral responses using various working-memory and reversal paradigms in spatial learning tasks. While initial learning performance was not impaired in *mdx* mice in these tasks, both working memory performance and behavioral flexibility were also unaffected.
Our study suggests that Dp427 loss in mice had no significant impact on executive functions, or that alterations were masked by influence of non-cognitive factors and presence of memory deficits. Future experiments using non-spatial paradigms might help confirming this hypothesis.

P1.202
Vulnérabilité à l'alcoolodépedance dans un modèle animal de schizophrénie

Jeanblanc, J. (Amiens)[1], Coune, F. (Amiens)[2], Jeanblanc, V. (Amiens)[3], Legastellois, R. (Emeryville)[4], Balguérie, K. (Amiens)[2], Naassila, M. (Amiens)[5]
[1]INSERM ERI 24, Université de Picardie Jules Verne, Amiens, France, [2]INSERM ERI 24, Amiens, France, [3]Université de Picardie Jules Verne, Amiens, France, [4]Gallo Research Center, Emeryville, United States, [5]INSERM ERI 24, Groupe de Recherche sur l'alcool et les Pharmacodépendances, Amiens, France

Les troubles liés à l'alcool se retrouvent chez 10 % de la population générale alors que cette proportion peut monter à plus de 50% chez les schizophrènes. Par ailleurs, il est intriguant de remarquer que les structures cérébrales impliquées dans chacune des pathologies, l'ensemble du circuit méso-cortico-limbique, sont parfaitement superposables. Dans cette étude nous avons tenté de mettre en évidence un phénotype de consommation excessive d'alcool dans le modèle de schizophrénie appelé NVHL (Neonatal Ventral Hippocampal Lesion).
Nous avons tout d'abord montré qu'une pré-exposition à l'alcool au cours de l'adolescence est nécessaire pour pouvoir observer à l'âge adulte une perte de contrôle de la consommation d'alcool dans un paradigme de libre choix en accès intermittent (ethanol 20%). Ensuite dans une tâche d'auto-administration opérante, nous avons montré que les rats NVHL sont plus motivés à consommer de l'alcool, présentent une résistance à l'extinction et rechutent 2 fois plus haut que les rats témoins. Enfin, nous avons testé l'effet d'un des rares traitement de l'alcoolodépendance à savoir la naltrexone sur les rats NVHL. Nous avons observés une réduction importante de la consommation d'alcool aussi bien chez les témoins que chez les rats NVHL.
L'ensemble de ces données montrent qu'une expérience précoce de la consommation d'alcool est nécessaire pour observer des troubles liés à l'alcool chez le rat NVHL adulte. L'étude des mécanismes moléculaires et cellulaires est en cours pour tenter de mieux comprendre la vulnérabilité à l'alcoolodépendance dans la schizophrénie.

P1.203
Motor resonance facilitates movement execution: an ERP and kinematic study

Ménoret, M. (Bron)[1], Curie, A. (Bron)[1,2], des Portes, V. (Bron)[1,2], Nazir, T.A. (Bron)[1], Paulignan, Y. (Bron)[1]
[1]Laboratoire sur le langage, le cerveau et la cognition-L2C2, UMR 5304, Institut des Sciences Cognitives, Bron, France, [2]Hospices Civils de Lyon, Service de Neuropédiatrie - HFME, Bron, France

Action observation, simulation and execution share neural mechanisms that allow a common motor representation. It is known that when these overlapping mechanisms are simultaneously activated by action observation and execution, motor performance is influenced by the observation and vice versa. To understand the neural mechanisms underlying this influence and to measure how variations in brain activity impact motor behaviour, we coupled kinematics and electrophysiological recordings of participants during the co-action/observation of congruent or non-congruent actions or during execution alone. We found increased velocity of the movement and a trajectory deviation of the executed action during the observation of congruent actions compared to the observation of non-

congruent actions or execution alone. This facilitation was distinguishable from the motor-related potentials of the participants; the motor-related potentials were transiently stronger in the congruent condition around the onset of the executed movement, which occurred 300 ms after the onset of the observed movement. This facilitation seemed dependent not only on spatial congruency but also on the optimal temporal occurrence of the observation and execution events.

P1.204

From learning set to task set in macaque monkeys

Faraut, M. (Bron)[1,2], Wilson, C. (Bron)[1,2], Procyk, E. (Bron)[1,2]
[1]INSERM U846, Stem Cell and Brain Research Institute, Bron, France, [2]Université de Lyon, UCBL, Lyon, France

In our open-ended and changing environment, it is necessary to behave in a flexible manner to produce optimal outcomes given changing contingencies. Behavioural flexibility requires us to understand the environment quickly and efficiently and adapt when it is changed. Primates can adapt behaviour flexibly in this way, but questions remain about how this flexibility is implemented, and what the neural correlates of such processes are.
Here we present the start of a project to investigate behavioural flexibility in macaque monkeys, using a test of their ability to adapt to changing task information in an environment that doesn´t always give the right answer.
3 monkeys learned a task of Task Set Manipulation, a monkey equivalent of the task of Collins & Koechlin (2012). In the training phases of the task, monkeys learned by trial and error the association between 2 stimuli and 3 targets using feedback to adapt their responses. Once they reached a behavioural criterion, 2 new stimuli were presented and monkeys entered a new exploration phase. Monkeys thus moved between phases of exploration and exploitation of the stimulus-reward environment. A stochastic reward environment was created by giving invalid feedback to monkeys in 10% of trials. This means that transitions between exploration and exploitation can only be triggered by a continuous checking of environmental information, and not solely by a single feedback, promoting flexibility of behaviour.
We show that monkeys used different learning strategies and adapted their responses to the stochastic environment. Monkeys demonstrated the formation of a stable learning set, a strategy that allows efficient learning of problems, acquired over a number of sessions. For example, over 400 problems, monkey P´s performance showed a reduction of mean errors to criterion of 93% between the first 50 and the last 50 problems, demonstrating a significant learning set. Finally, we show the level of transfer of this learning set to a version of the task in which mappings but not stimuli are changed. This task requires monkeys to learn to move between different task-sets as opposed to different problems, and therefore, as the project continues, we will use it to test the ability of monkeys to acquire and use task-sets.

P1.205

Small molecule activator of CBP/p300 acetyltransferases promotes neurogenesis and extends memory duration in adult mice

Cassel, R. (Strasbourg)[1], Chatterjee, S. (Strasbourg)[1,2], Mizar, P. (Bangalore)[2], Neidl, R. (Strasbourg)[1], Selvi, B.R. (Bangalore)[2], Mohankrishna, D.V. (Bangalore)[2], Vedamurthy, B.M. (Bangalore)[2], Schneider, A. (Strasbourg)[1], Bousiges, O. (Strasbourg)[1], Mathis, C. (Strasbourg)[1], Cassel, J.-C. (Strasbourg)[1], Eswaramoorthy, M. (Bangalore)[2], Kundu, T.K. (Bangalore)[2], Boutillier, A.-L. (Strasbourg)[1]

[1]Université de Strasbourg, UMR7364 - LNCA, Strasbourg, France, [2]Transcription and Disease Laboratory, Jawaharlal Nehru Centre for Advanced Scientific Research, Bangalore, India

If the brain functions of specific acetyltransferases such as the CREB-Binding protein (CBP) or p300 have been well documented with the use of mutant transgenic mice model, studies based on their direct pharmacological activation are still missing due to the lack of cell permeable activators. Here we present a small molecule (TTK21) activator of the histone acetyltransferases CBP/p300, which, when conjugated to glucose-based carbon nanosphere (CSP), passes the blood brain barrier, induces no toxicity, reaches different parts of the brain and crosses cell membranes. After intraperitoneal administration in mice, CSP-TTK21 significantly acetylated histones in the hippocampus and frontal cortex. Remarkably, CSP-TTK21 treatment promoted the formation of long and highly branched doublecortin positive neurons in the subgranular zone of the dentate gyrus and reduced BrdU incorporation, suggesting that CBP/p300 activation favors maturation and differentiation of adult neuronal progenitors. This is further supported by elevated mRNA levels of the neuroD1 differentiation marker and BDNF, a neurotrophin shown to be required for the terminal differentiation of new neurons in the adult hippocampus. Finally, CSP-TTK21 treatment was tested on spatial memory formation. We found that CBP/p300 activation during training, while not improving retention of a recent memory, resulted in a significant extension of memory duration. This report is the first evidence for CBP/p300-mediated histone acetylation in the brain by an activator molecule, along with demonstrations of beneficial implications on brain functions. We propose that direct stimulation of acetyltransferase function could stand as a valuable alternative of HDAC inhibitors, and could have important fallouts in terms of therapeutic options for brain diseases.

P1.206

Anorexia depends on the AKAP/PKA complex under the influence of serotonin 4 receptors in the nucleus accumbens

Pratlong, M. (Montpellier)[1,2], Laurent, L. (Montpellier)[1], Boussadia, B. (Montpellier)[1,2], Compan, V. (Montpellier)[1,2]
[1]Institut de Génomique Fonctionnelle, CNRS UMR 5203 - INSERM U.661, Dépt. de Neurobiologie, Montpellier, France, [2]Univ. Nîmes, Nîmes, France

Anorexia is a deadly mental disease related to restrictive diet despite requirements for energy. By modeling anorexia, we have previously caused by the first example of a rewarding molecular facet of anorexia: stimulating serotonin 4 receptors (5-HTR_4) or ecstasy favors anorexia caused by an increase in the levels of a peptide of addiction (CART: cocaine- and amphetamine-regulated transcript) within the nucleus accumbens (NAc), a part of the reward system. Here, we describe a new neural mechanism underlying anorexia-like behavior because increased levels in CART induced by the stimulation of 5-HTR_4 (BIMU8) within the NAc is enhanced when the binding between AKAP (A-kinase anchoring protein) / PKA complex is inhibited by the concomitant injection of St-Ht31 peptide. Results include changes in cAMP levels in the NAc following treatments and indicate a parallel enhancement of anorexia following the combined BIMU8/St-Ht31 treatment. Collectively, findings provide a new neural mechanism underlying anorexia in involving the AKAP/PKA complex with the 5-HTR_4/AMPc/PKA/CART signaling pathway within the NAc. Accordingly, an abnormal regulation of cAMP levels in the NAc by AKAP/PKA following the stimulation of 5-HTR_4 may likely trigger eating disorders such as, Anorexia nervosa.

P1.207

Decoding hand movement directions from intracranial EEG using power and cross-frequency coupling

Combrisson, E. (Lyon)[1], Perrone, M. (Grenoble)[2], Soto, J.L. (Sao Paulo)[3], Kahane, P. (Grenoble)[2], Lachaux, J.-P. (Lyon)[1], Jerbi, K. (Lyon)[1]
[1]INSERM U1028 UMR 5292, University Lyon 1, Lyon Neuroscience Research Center, Brain Dynamics and Cognition, Lyon, France, [2]Neurology Dept and GIN U836 INSERM-UJF-CEA, Grenoble, France, [3]Telecommunications and Control Engineering Department, University of Sao Paulo, Sao Paulo, Brazil

Intracranial EEG (iEEG) recordings in epilepsy patients have shown that movement execution leads to significant power modulations in multiple frequency bands in motor cortex (e.g. Crone et al. Brain, 1998). A number of studies have reported that iEEG recordings from the human motor cortex show directional tuning (Leuthardt et al. J Neural Eng, 2004; Mehring et al. J Physiol Paris, 2004). While

most decoding studies rely on power as decoding feature, a few recent studies have begun to explore the utility of Phase-Amplitude Coupling (PAC) as a new promising feature (Yanagisawa et al. J Neurosci. 2012; Wei et al. J Neural Eng 2007).
The aim of our study was to investigate the feasibility of using power and PAC to classify movement directions during planning and execution condition. To this end, several subjects implanted with iEEG in multiple brain structures performed a motor task. After an initial rest period (fixation), subjects were asked to prepare a reaching movement in one of four possible directions (up, down, right or left). Next, a Go signal appeared and subjects performed a center-out movement. We extracted two kinds of features: power and PAC. To define the optimal frequency bands of each, we developed an automatic algorithm which detects the most discriminant bands. We first evaluated the performance of each type of feature separately, and then we tested whether combining both features could further improve the classification. Our study revealed that power and PAC features provide performances with significant accuracies (>70%). In contrast to recent studies, we found that PAC can be a powerful tool to discriminate directions across conditions. Most of the discriminant power features were detected in the alpha and gamma bands, while the PAC provided best classifications when computed in the theta-gamma range. Furthermore, we found that multi-feature classification can significantly improve decoding performances. In conclusion, our study reveals that both power and PAC in fronto-parietal structures enable direction differentiation of the ongoing or up-coming movement. Our results are discussed in the context of novel paths for Brain-Computer Interfaces.

P1.208

How silent is silent reading? Intracerebral evidence for top-down activation of temporal voice areas during reading

Perrone-Bertolotti, M. (Lyon)[1], Kujala, J. (Lyon)[1], Vidal, J.R. (Lyon)[1], Hamame, C.M. (Marseille)[2], Ossandon, T. (Santiago)[3], Bertrand, O. (Lyon)[1], Minotti, L. (Grenoble)[4], Kahane, P. (Grenoble)[4], Jerbi, K. (Lyon)[1], Lachaux, J.-P. (Lyon)[1]
[1]INSERM U1028-CNRS UMR5292, Brain Dynamics and Cognition Team, Lyon Neuroscience Research Center, University Claude Bernard, Lyon, France, [2]Laboratoire de Psychologie Cognitive, CNRS (UMR 6146), Aix-Marseille Universite, Marseille, France, [3]Departamento de Psiquiatría, Facultad de Medicina y Centro Interdisciplinario de Neurociencia, Pontificia Universidad Catolica de Chile, Santiago, Chile, [4]CHU Grenoble and Department of Neurology, INSERM U704, F-38043, Grenoble, France

As you might experience it while reading this sentence, silent reading often involves an imagery speech component: we can hear our own “inner voice” pronouncing words mentally. Recent functional magnetic resonance imaging studies have associated that component with increased metabolic activity in the auditory cortex, including voice-selective areas. It remains to be determined, however, whether this activation arises automatically from early bottom-up visual inputs or whether it depends on late top-down control processes modulated by task demands. To answer this question, we collaborated with four epileptic human patients recorded with intracranial electrodes in the auditory cortex for therapeutic purposes, and measured high-frequency (50 -150 Hz) “gamma” activity as a proxy of population level spiking activity. Temporal voice-selective areas (TVAs) were identified with an auditory localizer task and monitored as participants viewed words flashed on screen. We compared neural responses depending on whether words were attended or ignored and found a significant increase of neural activity in response to words, strongly enhanced by attention. In one of the patients, we could record that response at 800 ms in TVAs, but also at 700 ms in the primary auditory cortex and at 300 ms in the ventral occipital temporal cortex. Furthermore, single-trial analysis revealed a considerable jitter between activation peaks in visual and auditory cortices. Altogether, our results demonstrate that the multimodal mental experience of reading is in fact a heterogeneous complex of asynchronous neural responses, and that auditory and visual modalities often process distinct temporal frames of our environment at the same time.

P1.209

D2 dopamine receptor agonist promotes a positive reinforcement in partial bilateral posterior ventral tegmental area of 6-OHDA lesioned rats

Ouachikh, O. (Clermont Ferrand)[1], Hafidi, A. (Clermont Ferrand)[2], Durif, F. (Clermont Ferrand)[3]
[1]Chu de Clermont-Ferrand, Univ. D'auvergne,ea 3845, Clermont Ferrand, France, [2]Université Blaise pascal clermont2, Clermont Ferrand, France, [3]CHU Gabriel Montpied, Clermont Ferrand, France

Dopamine dysregulation syndrome in Parkinson's disease (PD) has been attributed to dopamine replacement therapy (DRT) and/or a lesion of the dopaminergic system. The dopaminergic neuronal loss targets the substantia nigra and the ventral tegmental area (VTA). We hypothesize that DRT is responsible for the potential rewarding effect in PD by acting on the neuronal reward circuitry. Therefore this study explored the potential reward of DRT in partial bilateral VTA-lesioned animals. The posterior (p) VTA, which project to the nucleus accumbens (NAc) shell, constitutes the major neuronal circuitry implicated in addictive disorders. Using the conditioned place preference (CPP) behavioral paradigm, we investigated the motivational effects of dopamine receptor agonists (DARAs), and compared these effects to cocaine, which is well known to have a positive reinforcement effect, in rat with a partial bilateral lesion of the pVTA. Amongst the DARAs used only the D2R and D3R agonists (bromocriptine, PD128907 and pramipexole) and not DR1 subtype (SKF81297), induced a significant CPP in pVTA-lesioned animals, with the higher score obtained for bromocriptine. DARAs did not induce behavioral sensitization in Sham animals.
Immunochemical analysis of D2R expression was performed in the Nac shell of drug-free of both pVTA-lesioned and sham rats. Confocal D2R immunostaining analysis showed a significant increase in the number of D2R per cell in the NAc shell of pVTA lesioned rats compared to sham. This result demonstrated, for the first time, that DARAs effect is due to the DR2 overexpression in the NAc shell of pVTA-lesioned rats.
Moreover, cocaine, which is known to increase dopamine effect by blocking its synaptic recapture, induced behavioral sensitization in Sham group and not in dopamine deprived group. This result highlighted the importance of pVTA-NAc pathway in positive reinforcements. Altogether this study showed that neuronal degeneration in the pVTA, which caused a dopamine receptor sensitization in the nucleus accumbens shell, may explain how DRT promotes the appearance of DDS in PD.

P1.210

Joint influence of working memory load and time-on-task on rare target processing: an event-related study

Roy, R.N. (Grenoble)[1], Bonnet, S. (Grenoble)[1], Charbonnier, S. (Grenoble)[2], Campagne, A. (Grenoble)[3]
[1]CEA, LETI/STD/DTBS/LE2S, Grenoble, France, [2]GIPSA-lab, Grenoble, France, [3]Laboratoire de Psychologie et de NeuroCognition - UMR 5105, Grenoble, France

Both working memory load (WML) and time-on-task (TOT) are known to influence target processing, as well as its neural correlates. Yet, to our knowledge, no study has assessed their joint effect on neither the behavioral nor the electro-physiological level. In this study, we evaluated to what extent an increase of both WML and TOT affects the cortical processing of rare targets (versus distractors) by using a modified Sternberg paradigm that includes an oddball-like task. Twenty healthy participants (11 men, 25.3 years old) performed the experiment at 9 a.m. for 1.5 hour. In each trial, they had to memorize a list of digits presented sequentially on a computer screen. Then, they had to selectively react to the appearance of a geometrical shape (detection item), either a distractor, or a target (25% of the trials). Finally, they had to answer whether a probe digit was in the memorized list or not. Two levels of WML and TOT were considered (2/6 digits, short/long TOT), with a total of 144 trials per level. Detection items and the 2 levels of WML were pseudo-randomly presented. Response times (RT) and accuracies were recorded, as well as participants' answers to a mental fatigue questionnaire.

In addition, we measured the event-related potentials (ERPs) locked to the processing of the detection items. Both behavioral performance and the amplitude of ERPs were statistically analyzed using ANOVAs and Tukey post-hoc tests. Regarding behavioral results, an increase in WML reduced participants' accuracy and increased their RT to the probe item. Furthermore, TOT decreased participants' accuracy to detect targets and increased their RT in the low load condition. As for detection items' ERPs, preliminary analyses revealed that targets elicited components of higher amplitude at all electrode sites. Moreover, an increase in WML and TOT elicited a decrease in amplitude of the early components P1 and N1 and an increase in the amplitude of P2 over occipital, parieto-occipital and frontal sites, mostly for target items, in the low load condition. Interestingly, target/distractor P1, N2 and P2 discriminability was reduced at occipital and parieto-occipital sites when WML and TOT increased. Implications regarding cognitive resource allocation will be discussed.

P2.001

Neurotransmitters are modified in various brain areas of transgenic mice for APP or/and Dyrk1A, two chromosome 21 genes involved in Alzheimer disease and Down syndrome

London, J. (Paris)[1], Rouch, C. (Paris)[2], Assayag, E. (Paris)[2], Bui Chin, L. (Paris)[2], Medjaoui, H. (Paris)[2], Souchet, B. (Paris)[2], Luquet, S. (Paris)[2], Magnan, C. (Paris)[2], Dairou, J. (Paris)[2], Delabar, J.M. (Paris)[2]
[1]*Université Paris-Diderot, EAC-CNRS7059 BFA, Paris, France,* [2]*Univ Paris Diderot, Sorbonne Paris Cité, Adaptive Functional Biology, EAC CNRS 4413, Paris, France*

Background: Expression of *APP* (Amyloid Precursor Protein) and *Dyrk1A* (Dual-specific tyrosine phosphorylation-regulated kinase) genes is increased in the brain of patients with Alzheimer disease (AD) or Down syndrome (DS). Deficits in serotonine, dopamine or noradrenaline systems have been evidenced in post-mortem brain tissue samples from AD and DS patients and also in DS blood platelets and cerebrospinal fluid; these deficits may be related to neuronal loss, and to dendrite and spine alterations leading to synapse loss. Very few data on neurotransmitters in neural tissues of AD and mouse models are available. The aim of our study was to determine if and how the levels of monamine transmitters and their metabolites might be affected by *APP* or/and *Dyrk1A* overexpression in transgenic mice.
Results: Neurotransmitters expression was measured by electrochemical detection following by HPLC in hypothalamus, thalamus, hippocampus and striatum from 5 months old Tg*YAC hAPP* (TgA) males mice, Tg *mBACTgDyrk1A* (TgD) male and female mice and wild type mice. The main modification observed in TgA, TgD mice and TgA/TgD mice were:
a) in the serotonin system: a significant decrease of 5HT in the hypothalamus and hippocampus of TgD males and females mice and also in the same areas of TgA mice; an important increase in the serotonin turnover (5HIAA/5HT) in most structures of the trhee types of mice.
b) in the dopamine system: an important decrease was observed only in the hypothalamus of TgD mice with an increase in DOPAC/DA and HVA/DA.
c) in the noradrenergic system: whereas NorE is decreased in thalamus and hippocampus but not in hypothalamus in the TgD mice only, the adrenaline content is dramatically decreased in hypothalamus only but for the three types of mice.
These results will be extended soon to the measurements of the COMT and MAO content and activites of these enzymes involved in these pathways.
Conclusion: Overexpression of APP or/and Dyrk1A genes involved in both AD and DS conditions induced modified expression of neurotransmitters in various brain areas. As some monoamines can be assayed in the human blood, their measurement might be useful to follow the progression of the disease and the effects of pharmacological intervention.

P2.002

Molecular determinants for synaptic targeting of glutamate receptors at hippocampal mossy fiber synapses

Fièvre, S. (Bordeaux)[1], Carta, M. (Bordeaux)[1], Coussen, F. (Bordeaux)[1], Mulle, C. (Bordeaux)[1]
[1]*CNRS-Université Bordeaux Segalen, IINS, UMR 5297, Institut Interdisciplinaire de NeuroScience, Bordeaux, France*

Trafficking and stabilization of ionotropic glutamatergic receptors (AMPA, NMDA and kainate receptors (KARs)) at synaptic sites involves interactions with different partners including scaffolding proteins and

auxiliary subunits. These interactions occur through different receptor domains, including the C-terminal domain (CTD). In heterogous culture systems or in cultured dissociated neurons, surface trafficking of kainate receptors involves the 39 last amino acids of the CTD. KARs are specifically expressed in the *stratum lucidum* where mossy fibers contact CA3 pyramidal cells (Mf-CA3 synapses), whereas they are absent from other glutamatergic synaptic inputs to the same neuron. The mechanisms for such a constrained subcellular segregation is not known. At Mf-CA3 synapses, KARs comprise the GluK2, GluK4 and GluK5 subunits. Genetic ablation of GluK2 induces the loss of synaptic KAR-mediated currents. To investigate the role of the CTD of GluK2 in the synaptic trafficking and stabilization of KARs at mossy fiber-CA3 synapse we used a molecular replacement strategy in organotypic hippocampal cultures combined with electrophysiology and focal glutamate uncaging. Reexpression of GluK2wt in CA3 pyramidal cells of GluK2$^{-/-}$ mice specifically restores KAR-mediated currents at Mf-CA3 synapses and not at other glutamatergic inputs. By reexpression of mutant and truncated GluK2 subunits, we have identified a region in the CTD of GluK2 necessary for the synaptic trafficking and stabilization of KARs at Mf-CA3 synapses. We are currently performing glutamate uncaging coupled to patch clamp recordings to determine a functional map of KARs along CA3 pyramidal cells dendrites. Our study provide evidence for the molecular mechanisms underlying the stringent segregation of glutamate receptor subtypes at specific glutamatergic synapses in the hippocampal circuit.

P2.003

Transcriptional regulation of CRMP5 gene: Sox5 acts in vitro as a positive transcription factor

Naudet, N. (Lyon)[1], Chounlamountri, N. (Lyon)[1], Watrin, C. (Lyon)[1], Cavagna, S. (Lyon)[1], Honnorat, J. (Lyon)[1], Besançon, R. (Lyon)[1]
[1]*Centre de Recherche en Neurosciences de Lyon U1028 UMR5292 Univ Lyon1, Neurooncology Neuroinflammation Team, Faculty of Medicine Laennec, Lyon, France*

CRMP5 (Collapsin Response Mediator Protein 5) is highly expressed in developing brain, but poorly in adulthood. It plays a role in neuronal development. It has been previously shown in a development model with primary cells from fresh embryonic mouse hippocampus that changes in CRMP5 expression during early phases of development is associated with neuronal polarization. Our study focused on the transcriptional regulation of the CRMP5 gene.
Promoter activity was investigated by transfection and luciferase reporter assays; we show that the 2000 bases upstream of the CRMP5 gene promote its activity. Our results obtained using fractionated constructs of CRMP5 promoter show that one region enhances gene transcription; this region could potentially bind the transcriptional factor Sox5.
In one hand, mutation of the putative Sox5 binding site leads to a sharp decrease of CRMP5 promoter activity, in the other hand overexpression of Sox5 stimulates CRMP5 promoter activity, indicating that Sox5 could be a major transcription factor in CRMP5 promotion.
Overexpression of Sox5 into immortalized embryonic mouse hippocampal cell line leads to an increase of endogenous CRMP5 expression.
Our study supports that Sox5 promotes intrinsic regulations of CRMP5 gene.

P2.004

Epigenetic modifications after voluntary and chronic ethanol intake in the C57BL/6J mice hippocampus

Stragier, E. (Paris)[1], Massart, R. (Paris)[1], Hamon, M. (Paris)[1], Lanfumey, L. (Paris)[1]
[1]*Centre de psychiatrie et neurosciences, Pitié Salpêtrière, INSERM UMR 894, Paris, France*

Although ethanol at high dosage is well known to induce neurotoxic effects on hippocampal neurogenesis, chronic voluntary and moderate ethanol intake has been shown to stimulate dentate gyrus (DG) cell proliferation in C57BL/6J mice. In order to unveil the molecular mechanisms underlying ethanol-induced neuroplasticity, we studied the expression of markers involved in cell differentiation and plasticity in the hippocampus of C57BL/6J mice after exposure to free ethanol consumption. In addition, ethanol-induced epigenetic modifications were investigated by quantifying the expression of epigenetic markers such as HDACs, which are implicated in gene transcription control and neuronal maturation, and histones, whose post-translational modifications modulate gene transcription. qRT-PCR, chromatin immunoprecipitation and immunohistochemistry approaches were used for relevant determinations in the hippocampus of C57BL/6J mice that had free access to ethanol versus tap water for 3 weeks.
Voluntary ethanol consumption was found to increase the hippocampal expression of both bHLH activator family (transcription factors implicated in neural proliferation and maturation) and BDNF and to decrease that of HDACs. In addition, MeCP2 expression was increased at both mRNA and protein levels in the DG and CA3, and upregulation of acetylated histone H4 (H4Ac) and trimethylated histone H3 (H3K4me3) was also noted in these hippocampal subareas in ethanol-exposed mice. Analysis of *bdnf* gene evidenced ethanol-induced modifications at transcription level associated with variations affecting H3Ac and H3K4me3 in *bdnf* promoter. Changes in *bdnf* exons expression occurred together with ethanol-induced increase in BDNF protein in the hilus and CA3 area of the hippocampus. Altogether, these results suggest that chronic moderate ethanol intake under free choice conditions promotes neurogenesis in the DG in C57BL/6J mice by stimulating the expression of neural factors and BDNF. Epigenetic mechanisms involving HDAC and histone modifications at the level of *bdnf* gene play probably a key role in ethanol-induced BDNF upregulation in the hippocampus.

P2.005

Synaptic deficiency of STOP KO neurons observed by transmission electron microscopy

Pernet-Gallay, K. (La Tronche)[1], Brocard, J. (La Tronche)[2,3], Delaroche, J. (La Tronche)[1,3], Coulomb, N. (Grenoble)[3,4], Andrieux, A. (Grenoble)[3,4]
[1]Grenoble Institut des Neurosciences, Plateforme Microscopie Electronique, La Tronche, France, [2]Grenoble Institut des Neurosciences, La Tronche, France, [3]Université Joseph Fourrier, Grenoble, France, [4]Grenoble Institut des Neurosciences, Grenoble, France

STOP is a brain enriched protein that has actin binding properties and that confers microtubules cold stability depending on its phosphorylation state by CamKII. STOP KO mice present behaviour disorders, alteration of some axonal tract and LTP deficiency in the hippocampus. To better understand the role of STOP in LTP we compared the morphology of hippocampus WT and STOP KO cultured neurons at 23 DIV by electron microscopy. We show that spine morphology of STOP KO neurons is normal and no differences are observed between WT and KO neurons at rest. However, chemical stimulation with 90 mM KCl induces an increase of the thickness of the PSD in WT synapses, due in part to the recruitment of CamKII, that is not observed in STOP KO synapses. In contrast, when neurons are stimulated with glutamate, there is an increase of the thickness of the PSD in both WT and STOP KO synapses, indicating that recruitment of post synaptic proteins occurs normally in STOP KO neurons. This result suggest that the synaptic defect in STOP KO neurons is located in the presynaptic side.

P2.006

Syk kinases are implicated in the establishment of neuronal connections during development

Noraz, N. (Lyon)[1], Benon, A. (Lyon)[2], Jaaoini, I. (Lyon)[2], Chounlamountri, N. (Lyon)[2], Charoy, C. (Lyon)[3], Ya, C. (Lyon)[2], Watrin, C. (Lyon)[2], Benetollo, C. (Lyon)[4], Boudin, H. (Nantes)[5], Honnorat, J. (Lyon)[2], Castellani, V. (Lyon)[3], Pellier-Monnin, V. (Lyon)[2]
[1]Centre de Recherche en Neuroscience de Lyon, INSERM 1028 CNRS UMR 5292, Equipe Neurooncologie & Neuroinflammation, Lyon, France, [2]Centre de Recherche en Neuroscience de Lyon (CRNL), UMR 5292 CNRS/U1028 INSERM, Lyon, France, [3]Centre de Génétique et de Physiologie Moléculaire et Cellulaire, Lyon, France, [4]Centre de Recherche en Neuroscience de Lyon, Plateforme neurogénétique fonctionnelle et optogénétique, Lyon, France, [5]Institut de transplantation et de recherche en transplantation, Nantes, France

The function of the tyrosine kinases of the Syk family is well characterized in the hematopoietic system. They relay the signals emanating from the immunoreceptors, pillars of immune responses. An increasing number of data involve immunoreceptors in neural function. However it has yet to be determined whether these proteins in fact transmit an immunoreceptor-like signal in neuronal cells. In a previous work, we showed that phosphorylated forms of Syk - representative of an active form of the kinase - are confined to specialized neuronal populations of migrating cells (Hatterer, 2011).
(i) As a first example, in the developing cerebellum, phospho-Syk is restricted to a specific area corresponding to post-mitotic granule cells migrating tangentially through the external granular layer (EGL). The implication of Syk in this function was established *ex vivo*, from EGL micro-explants, a model of tangential migration, using a Syk pharmacological inhibitor. The ephrinB2/EphB2 pathway was identified as responsible for the modulation of Syk state of activation in this system. Finally, the implication of the Syk kinase family in GC migration was confirmed *in vivo*, in $Syk^{+/-}$/$ZAP\text{-}70^{-/-}$ mice. Indeed, ectopic granule cells were observed in the molecular layer of these adult mutant animals.
(ii) As a second example, in the developing spinal cord, growing commissural axons of E11-13 embryos expressed the phosphorylated form of Syk kinase, either in pre-crossing at the midline or in post-crossing. Analyzes of $Syk^{-/-}$/$ZAP\text{-}70^{-/-}$ double KO open book preparations, revealed late pathfinding defects, pointing to a set of guidance molecules involved in post-crossing. In a growth cone collapse assay, on cultured commissural neurons established from WT and $Syk^{-/-}$/$ZAP\text{-}70^{-/-}$ double KO embryos, Syk was found as required for ephrinB3/EphB2-induced collapse. In cultured cells, Syk was constitutively phosphorylated and ephrinB3-induced collapse was associated with an inhibition of Syk phosphorylation. This was in agreement with the collapsing effect of a Syk pharmacological inhibitor observed on commissural neurons.
In conclusion, Syk kinases are required in ephrin-dependent developmental events contributing to the establishment of neuronal connectivity.

P2.007

Molecular mechanisms of spatial working memory and reference memory in the hippocampus: a western blot analysis

Fraize, N. (Lyon)[1], Hameih, M. (Lyon)[1], Touret, M. (Lyon)[1], Salin, P. (Lyon)[1], Malleret, G. (Lyon)[1]
[1]*Univ. Claude Bernard Lyon 1, Fac. de Médecine RTH Laënnec, CNRS UMR 5292 Réseaux cycle veille-sommeil, Lyon, France*

Working Memory (WM) and Reference Memory (RM) are two processes aimed to store information. WM is a specific form of short-term memory that refers to the ability to retain information within a single trial. This information can then be stored into long-term/reference memory. RM refers to the long-term storage of information that remains constant over time, that is gradually acquired over many training sessions and is widely believed to be dependent on long term potentiation (LTP) of the synaptic efficacy. However, not all information that comes from working memory needs to be transferred into long-term memory. Insignificant data is better to be erased in order not to overload the brain with irrelevant things. It has been demonstrated that WM is very sensitive to proactive interference (PI) which is a phenomenon whereby information learned in the past interferes with the learning of more recently presented material. Consequently, forgetting this old information would be necessary to perform everyday tasks requiring WM abilities. Work from our team suggest that long term depression (LTD) of synaptic efficacy may serve to weaken previous memory traces, thus preventing them to interfere with new ones. In order to assess post-synaptic changes in receptors trafficking and signaling involved in LTP or LTD, we performed a selective WB analysis in the different areas of the hippocampus in control and test rats submitted to 10 days of training in a RM or WM task with or without interference. Our results show that, in the dentate gyrus (DG), the WM task with high level of interference induces a selective increase in the phosphorylation of the CamKinaseII (CamKII) as compared to controls and rats tested in RM or WM task with low interference. This increase was not observed in CA1 and CA3 area of the hippocampus. CamKII is a key protein involved in long-term synaptic plasticity and is activated by a synaptic increase in calcium concentration. Thus, our results suggest that long-term synaptic plasticity occurs selectively in the DG during the WM task with high interference, and that, in this task that requires a high level of cognitive flexibility, the memory trace must be quickly formed as well as quickly erased.

P2.008

Neurochemical and haematological analysis of haemorrhage in the neurohypophysis of male *Wistar* rat

Rouaiguia, N. (Alger)[1], Mamine-Dorbani, L. (Alger)[1], Ayad, G. (Alger)[1]
[1]*Université des sciences et de la technologie USTHB , Faculté des sciences biologiques FSB, LBPO/ Equipe de neurochimie, Biologie, Alger, Algeria*

Interactions between hormonal systems and neurotransmitters in the hypothalamo-neurohypophyseal system (HNS) are important factors in the maintenance of hydromineral balance. We are interested in the study of neurochemical plasticity by measuring changes in catecholindolaminergic activity in the neurohypophysis of male *WISTAR Rat* submitted to hypovolemia of 10% of total blood volume (tbv) and their implication in the control of homeostasis. After hypovolemia of 10% of tbv followed by 10mn of post-haemorrhagic recovery, we noted a decrease in hematocrit and natremia that demonstrated haemodilution, probably caused by the antidiuretic activity of vasopressin (AVP). Thus, statistical analysis showed an increase in kaliemia and plasma osmolarity. The results obtained during the study of systemic parameters, gave a reliable indication of the relative change in plasma volume. In the second part of our work we showed that hypovolemia induced profound alterations in neurochemical study which concerns the determination of catecholindolamines rates (DA dopamine, norepinephrine NA and serotonin 5-HT) and their metabolites (DOPAC, HVA, 5HIAA) in neurohypophysial tissue. After HPLC-DEC separation and analysis, DA and 5-HT increased when rate of NA decreased. The increase in the DA and the accumulation of DOPAC result from the increased activity of tuberohypophyseal dopaminergic neurons. Although, the rate of serotonin and its metabolite 5-HIAA were not significantly affected by the hemorrhage. Serotonin turnover is altered indicating a localized effect of hypovolemia in the catabolism serotonin. We suggested that this decline could be resulted from an accelerated transfer of this neurotransmitter into the blood.
In conclusion, our results suggested that changes in neurochemical and haematological parameters could result from a failure of hydromineral homeostasis which activates several neuroendocrine mechanisms including the release of catecholindolamines, ANG II and VP. These pituitary hormones work with the autonomic nervous system to decrease water and sodium loss, adjust the distribution of water between intra and extracellular fluid compartments.
Keywords: ***Catecholindolamines, neurohypophysis, AVP, hypovolemia, hydromineral homeostasis.***

P2.009

Membrane lipids tune synaptic transmission by direct modulation of potassium channels

Carta, M. (Bordeaux)[1], Lanore, F. (Bordeaux)[1], Rebola, N. (Bordeaux)[1], Szabo, Z. (Bordeaux)[1], Lourenço, J. (Bordeaux)[1], Nadler, A. (Heidelberg)[2], Schultz, C. (Heidelberg)[2], Blanchet, C. (Bordeaux)[1], Mulle, C. (Bordeaux)[1]
[1]*Interdisciplinary Institute for NeuroScience, UMR 5297, Bordeaux, France,* [2]*EMBL, Heidelberg, Germany*

Voltage-gated potassium (Kv) channels are involved in action potential repolarization in excitable cells. Exogenous application of membrane-derived lipids, such as arachidonic acid (AA), regulates the gating of Kv channels. Whether membrane-derived lipids released under physiological conditions impact on neuronal coding through this mechanism is unknown. We have investigated this possibility at mossy fiber to CA3 synapse in the hippocampus, where Kv channels in the presynaptic terminal have been shown to regulate glutamate release. He have found that that AA released in an activity-

dependent manner from postsynaptic hippocampal CA3 pyramidal cells acts as a retrograde messenger which induces robust facilitation of mossy fiber synaptic transmission. AA acts by broadening presynaptic action potentials through the direct modulation of Kv channels. This form of short-term plasticity can be triggered by natural patterns of spike discharge in the postsynaptic cell, and sets the threshold for the induction of the presynaptic form of long-term potentiation (LTP) at hippocampal mossy fiber synapses. Hence, direct modulation of presynaptic Kv channels by activity-dependent release of lipids serves as a physiological mechanism for the tuning of synaptic transmission.

P2.010

MuSK-ColQ interaction and signalisation in synaptogenesis of the neuromuscular junction

Dobbertin, A. (Paris)[1], Legay, C. (Paris)[1]
[1]*Université Paris Descartes, CNRS UMR 8194, Paris, France*

Collagen Q (ColQ) is a specific collagen expressed by muscles at the neuromuscular junction (NMJ), where it plays a crucial role by anchoring and accumulating acetylcholinesterase (AChE) in the synaptic basal lamina. The accumulation of the AChE-ColQ complex at the synapse requires the interaction of ColQ COOH terminus with muscle-specific receptor tyrosine kinase (MuSK), a key protein for NMJ formation. In particular, MuSK mediates the actions of the proteoglycan, agrin, on postsynaptic differentiation.
In addition to its structural function, ColQ has important regulatory functions at the synapse by controling acetylcholine receptor (AChR) clustering and synaptic gene expression. At least part of ColQ effects, are mediated by ColQ-MuSK interaction. Moreover, cell surface levels of MuSK are decreased *in vitro* and *in vivo* in cells lacking ColQ, suggesting that ColQ controls MuSK sorting or stabilisation in the muscle membrane.
In an attempt to understand how ColQ exerts its regulatory functions, we are investigating the properties of ColQ-MuSK interaction and the molecular events induced by the binding of ColQ to MuSK. To identify the domains of MuSK required for association with ColQ, we generated MuSK mutants deleted of the extracellular Ig-like domains 1, 2, 3, 4 or the cysteine-rich domain and we show that ColQ-MuSK interaction involves Ig-like domains 1 and 4. We also studied if ColQ modulates agrin-induced activation and phosphorylation of MuSK. Using ColQ-transfected and ColQ-deficient muscle cell lines, we show that ColQ inhibits agrin-induced MuSK phosphorylation. These results indicate that ColQ regulates MuSK activation and agrin function. The underlying molecular mechanisms as well as the influence of ColQ on the signaling pathways initiated by MuSK are under current investigations.

P2.011

Widespread expression of GluR delta-1 (GluD1) in the rodent brain

Hepp, R. (Paris)[1], Nomura, S. (Paris)[1], Hay, A. (Paris)[1], Dauphinot, L. (Paris)[2], Poirel, O. (Paris)[3], Potier, M.-C. (Paris)[2], El Mestikawy, S. (Paris)[3], Lambolez, B. (Paris)[1], Tricoire, L. (Paris)[1]
[1]*Université Pierre et Marie Curie, UMR7102, Paris, France,* [2]*CRICM, UPMC/CNRS UMR7225/INSERM UMRS975, Paris, France,* [3]*Université Pierre et Marie Curie, INSERM UMRS 952, CNRS UMR 7224, Paris, France*

The delta family of ionotropic glutamate receptors consists of the glutamate δ1 (GluD1) and glutamate δ2 (GluD2) receptors. GluD2 is strongly expressed in cerebellum where it regulates synapse structure and function. In contrast, the expression pattern of GluD1 in the central nervous system (CNS) is still a matter of debate. In this study the expression of GluD1 was examined in the rodent brain and

especially the cerebral cortex by in situ hybridization, single cell RT-PCR, quantitative PCR and western blotting using a new polyclonal antibody developed in the laboratory. In situ hybridization performed on adult mouse brain sections showed a widespread GluD1 mRNA expression. Labeling was stronger in forebrain structures such as hippocampus, striatum and cerebral cortex compared to hindbrain structures and cerebellum. Single cell RT-PCR performed on cortical neurons revealed that GluD1 mRNA is expressed in both excitatory pyramidal neurons and inhibitory interneurons. Cells co-expressing both GluD1 and GluD2 were also detected (20-30%). This shows that GluD1 is the major GluR delta subunit expressed in the cortex. Quantitative PCR showed that GluD1 mRNA expression in cerebral cortex doubles during the first postnatal month to reach the adult level. To examine the developmental time course of GluD1 expression at the protein level, we generated rabbit polyclonal antibodies raised against a C-terminal region of GluD1 fused to GST. In western blot, a band for GluD1 at the expected size was detected in extracts of GluD1 expressing HEK cells but not in GluD2 expressing cells indicating that the sera did not cross react with GluD2. Besides, this band was absent in brain extract from GluD1 knock-out mice. Using these antibodies, we found that GluD1 expression increased 4 fold during the two first postnatal weeks to reach adult levels. Cortical GluD2 expression exhibited a similar developmental time course both at the mRNA and protein levels. These data show that GluD1 is widely expressed in the rodent brain and that its expression is upregulated during the postnatal development.

P2.012

AP3m1 is required for synaptic scaling

Steinmetz, C.C. (Waltham)[1], Rutlin, M. (Waltham)[2], Shima, Y. (Waltham)[2], Sugino, K. (Waltham)[2], Lefort, S. (Waltham)[2], Nelson, S. (Waltham)[2], Turrigiano, G. (Waltham)[2]
[1]Brandeis University, Biology Dept, Waltham, United States, [2]Brandeis University, Waltham, United States

Synaptic scaling (SS) is a form of homeostatic plasticity that restores neuronal activity to 'baseline' levels in response to altered activity (Turrigiano *et al*., 1998). SS of excitatory synapses has been demonstrated in a variety of central neurons both *in vitro* and *in vivo,* and has been found to be transcription-dependent (Ibata et *al*., 2008). In order to identify novel transcripts that are up-regulated during SS we used a lentiviral transgenic mouse expressing the fluorescent protein, mCitrin. By immuno-staining and current clamp recording, we determined that in layer 4 of the primary visual cortex (L4V1) the expression of mCitrin is restricted to pyramidal neurons. We verified that mCitrine neurons in L4V1 express SS *in vivo* by blocking retinal activity for 2 days with monocular TTX injections and by recording mEPSCs at P14 from deprived and non-deprived mCitr+-neurons. Using an unbiased screen, we found that the mu1 subunit of the adaptor complex AP3 (AP3m1) is reliably up-regulated in L4V1 pyramidal neurons when visual activity drops and synapses are scaled up in strength. AP3m1 is a member of the clathrin-adaptor protein complex family AP1-4 and is important in trafficking vesicles between compartments within the trans-Golgi network. To determine whether AP3m1 is required for the expression of synaptic scaling we used an *in vitro* loss-of-function approach. We showed that down-regulating AP3m1 by shRNA blocks scaling up in synaptic strength induced by TTX in cultured cortical pyramidal neurons. So far, these data suggested that AP3m1 is critical for scaling up. Currently, we are investigating which sub-cellular compartment AP3m1 localizes to, whether AP3m1 co-localizes with AMPAR, and are performing gain of function experiments. This study should significantly increase our understanding of the molecular mechanisms of activity-dependent AMPAR trafficking as well as generate new insights into the normal mechanisms that maintain stability of neuronal circuit function.

P2.013

Toward a complete Purkinje neuron translatome with subcellular resolution

Launey, T. (Wako-shi)[1], Kratz, A. (Tsurumi)[2], Beguin, P. (Wako-shi)[3], Kaneko, M. (Wako-shi)[3], Chimura, T. (Wako-shi)[3], Suzuki, A.M. (Tsurumi)[2], Bertin, N. (Tsurumi)[2], Vigot, R. (Wako-shi)[3], Carninci, P. (Tsurumi)[2], Plessy, C. (Tsurumi)[2]
[1]RIKEN Brain Science Institute, Launey Research Unit, Wako-shi, Japan, [2]RIKEN Yokohama, Omics Science Center, Tsurumi, Japan, [3]Brain Science Institute RIKEN Wako, Launey Research Unit, Wako-shi, Japan

Development and synaptic plasticity in complex neurons requires local protein synthesis. To clarify the mechanisms controlling protein synthesis in the Purkinje cell and in particular in its dendrites, we combined several technologies to specifically extract ribosomes from this neuron. As the main component of the protein synthesis machinery, the ribosome is transiently bound to the messenger RNAs being translated into proteins. By encoding a ribosome-binding probe ("TRAP") into a virus vector targeted to PCs, we succeeded to capture several thousand ribosome-bound PC RNAs (translatome). Differential fractionation allowed us to discriminate cytoplasmic polysome from ER-associated ribosomes. High-sensitivity NanoCAGE and deep-sequencing were then used to identify all the isolated RNAs, either as protein-coding sequences or as regulatory RNAs. Using microdissections to separate the PC dendrites from its cell body, we could identify a group of several hundred RNAs that appear to be transported into dendrites for remote translation. Interestingly, in addition to transcripts of protein known to be involved in synaptic plasticity, we also found that these ribosome-bound RNAs include a large fraction of transcripts with no known function, representing either novel genes or regulatory RNA sequences.

P2.014

Embryonic microglia regulate forebrain wiring

Squarzoni, P. (Paris)[1], Oller, G. (Paris)[1], Pont-Lezica, L. (Paris)[2], Hoeffel, G. (Singapore)[3], Rostaing, P. (Paris)[2], Bessis, A. (Paris)[2], Ginhoux, F. (Singapore)[3], Garel, S. (Paris)[1]
[1]Institut de Biologie de l'Ecole Normale Supérieure, CNRS UMR 8197-INSERM U1024, Brain Development and Plasticity, Paris, France, [2]Institut de Biologie de l'Ecole Normale Supérieure, CNRS UMR 8197-INSERM U1024, Paris, France, [3]Singapore Immunology Network (SIgN), 8A Biomedical Grove, IMMUNOS Building Nos. 3-4, BIOPOLIS, 138648, Singapore, Singapore

Forebrain functioning relies on complex neural circuits that are built during embryogenesis. Defects in this process, triggered by either genetic mutations or maternal factors such as prenatal inflammation, can lead to neurologic or psychiatric disorders. Microglia, the resident macrophages of the brain, were recently shown to exert immune-independent functions during adult neurogenesis and postnatal synaptic pruning. Microglia invade the brain during early neurogenesis, thereby raising the intriguing possibility that they may participate in forebrain wiring or mediate deleterious effects of prenatal inflammations.
To address this major issue, we examined microglial location in the mouse embryonic forebrain and found that they concentrate in "hotspots", which are not associated with cell death, but that are crucial for axonal navigation and neuronal migration. Following these observations, we assessed the impact of either an absence/reduction in embryonic of microglia or their improper activation on forebrain embryogenesis. Using both genetic and pharmacological models of microglia depletion and/or inflammation, we observed specific axonal and neuronal defects, restricted to the hotspots in which microglia accumulate in normal conditions. Hence, our work shows for the first time that microglia contribute to normal brain embryogenesis and can act as deleterious mediators of maternal inflammation. This study provides novel insights into the mechanisms governing normal and pathological brain wiring and reveals a remarkable interplay between the development of the nervous and immune systems.

P2.015
VEGF modulates NMDA receptor function and synaptic localization in the hippocampus

DeRossi, P. (Lyon)[1], Chounlamountri, N. (Lyon)[1], Benetollo, C. (Lyon)[2], Honnorat, J. (Lyon)[1], Salin, P.-A. (Lyon)[3], Meissirel, C. (Lyon)[1]
[1]Centre de Recherche en Neuroscience de Lyon U1028 UMR5292 UnivLyon1, Neurooncology Neuroinflammation Team, Faculty of Medicine Laennec, Lyon, France, [2]Plateau Neurogénétique Fonctionnelle & Optogénétique, Lyon, France, [3]Centre de Recherche en Neuroscience de Lyon U1028 UMR5292 UnivLyon1, Sleep Team, Faculty of Medicine Laennec, Lyon, France

The vascular endothelial growth factor (VEGF) is well known to play a critical role during vascular development but recent evidence indicates that VEGF also regulates various neuronal processes in the nervous system, such as neurogenesis, hippocampal synaptic plasticity, learning and memory. However, little is known about the underlying molecular mechanisms through which VEGF regulates hippocampal synaptic transmission and plasticity. Recently, we showed a novel interaction between the glutamate receptor NMDA (NMDAR) and the VEGF receptor VEGFR2 during neuronal migration in the developing cerebellum. As NMDAR have been widely implicated in synaptic transmission and plasticity, we hypothesized that VEGF signaling might regulate NMDAR function at hippocampal synapses. Our results revealed that VEGF as well as its receptor VEGFR2 are expressed by pyramidal cells in the CA1 and CA3 regions of the hippocampus indicating that VEGF is an endogenous ligand for VEGFR2 in pyramidal cells. In addition, whole-cell patch clamp experiments in acute hippocampal slices showed that VEGF potentiates NMDAR mediated synaptic transmission. Using a paired-pulse stimulation, we demonstrated that this VEGF-dependent enhancement involves a postsynaptic modulation of NMDAR function, mediated by GluN2B expressing NMDAR. To better understand the molecular mechanisms underlying this postsynaptic modulation, we explored the effect of VEGF on the surface expression and synaptic targeting of NMDAR. Cell-free system exploration highlighted an interaction between the extracellular domains of VEGFR2 and the GluN2B subunit of NMDAR. Further, high-resolution imaging revealed that NMDAR and VEGFR2 co-activation in hippocampal cell culture induces an increase in GluN2B expressing NMDAR at synaptic sites and affects synapse number. Altogether, our results indicated a VEGF-dependent mechanism for regulating NMDAR localization and function at hippocampal synapses, suggesting that this molecular mechanism could be relevant for long-term synaptic plasticity, learning and memory.

P2.016
Involvement of STAT3 in the maintenance of monkey neural stem cells

Wianny, F. (Lyon)[1,2], Waltispurger, J. (Lyon)[1,2], Bourillot, P.-Y. (Lyon)[1,2], Dehay, C. (Lyon)[1,2]
[1]Inserm U846, Stem Cell and Brain Research Institute, Lyon, France, [2]Université de Lyon, Lyon 1, UMR-S 846, Lyon, France

Neural stem cells (NSCs) can be expanded *in vitro* as a self-renewing and multipotent population. The derivation of these cells in the non-human primate represents an invaluable tool for studying neuronal developmental processes, for drug screening and for pre-clinical validation of transplantation therapies. In the mouse, STAT3 (Signal Transducer and Activator of Transcription 3) is known to sustain NSC self-renewal and survival in response to the activation of the Notch pathway. To study the role of STAT3 in the maintenance of NSCs derived from monkey embryonic stem (ES) cells, we generated a Rhesus ES cell line stably expressing the fusion protein between the entire coding region of STAT3 and the modified ligand binding domain of the estrogen receptor. This mutant form of STAT3 is active upon 4-hydroxytamoxifen (4HT) treatment. We found that STAT3 activation (1) increased the proportion of NSCs during the initial steps of derivation, as shown by an increased proportion of Pax6

and Sox2 expressing cells and (2) increased the overall number of NSCs in long term cultures. By contrast, suppression of STAT3 signaling in NSCs using a lentivirus encoding a STAT3 shRNA led to massive cell death. The few surviving cells adopted a neuronal phenotype. Inhibition of Notch activation in NSCs reduced the pool of NSCs and induced neuronal differentiation. This effect could be partially bypassed by the activation of STAT3. These results suggest that STAT3 plays a central role in the maintenance of monkey NSCs.
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P2.017

Mitotic spindle asymmetry: a novel mechanism to generate asymmetrical division in cortical precursors is regulated by Wnt/PCP signaling

Delaunay, D. (Bron)[1], Knoblauch, K. (Bron)[1], Dehay, C. (Bron)[1]
[1]*Institut Cellule Souche et Cerveau, INSERM U 846, Bron, France*

Asymmetric cell division (ACD), whereby a progenitor gives rise to two dissimilar daughter cells, is tightly regulated during corticogenesis. Although several mechanisms are known to be involved in ACD, their relative contributions remain incompletely understood. Here we report that spindle size asymmetry (SSA) constitutes a novel ACD related mechanism during corticogenesis. We show in the mouse cortex that SSA occurrence and magnitude parallels ACD. We also show that Wnt7a and Vangl2, two members of the Wnt/Planar cell polarity pathway, influences the magnitude of the SSA and are essential to maintain spindle symmetry in cortical precursors both *in vitro* and *in vivo*. We found that the effect of the Wnt7a/Vangl2 pathway is relayed at the cell cortex by an adaptator protein that links the plasma membrane to the Cytoskeleton. Finally, *in vivo*, we found that the loss of Vangl2 around mid-neurogenesis drastically increase the SSA, ultimately leading to a loss of the late born neuron population. Therefore, we report here a novel type of division-controlling mechanism under the control of Wnt/PCP pathway: the mitotic spindle asymmetry.
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P2.018

Long-term depression of inhibition in CA2 allows activation of CA2 neurons by CA3

Nasrallah, K. (Paris)[1], Piskorowski, R. (Paris)[2], Chevaleyre, V. (Paris)[2]
[1]*Université Paris Descartes, Paris, France,* [2]*Université Paris Descartes, CNRS UMR 8118, Paris, France*

For decades, studies have developed a linear model of information transfer to explain hippocampal function, which has excluded area CA2. In this model, information travels through the hippocampus *via* a tri-synaptic loop (EC-DG-CA3-CA1): axons originating from layer II of the entorhinal cortex (EC) activate neurons in the dentate gyrus (DG) that project to CA3 neurons that then activate CA1 pyramidal cells. Studies in which CA3-CA1 transmission was abolished reveal that this tri-synaptic pathway may be insufficient to explain hippocampal function. Consequently, hippocampal circuitry, and namely the role of CA2 in the hippocampal network, has to be revisited. CA2 may be playing a significant role in hippocampal function, participating in information flow through the hippocampus. Indeed, CA2 forms a di-synaptic loop (EC-CA2-CA1), independent of CA3-CA1 transmission. These neurons are also connected by CA3 input, and like CA3 neurons, they project to CA1. There is a very strong level of inhibition in CA2 that prevents the activation of CA2 pyramidal cells by CA3 neurons. Consequently, information is propagated through the tri-synaptic circuit instead of a parallel quadric-

synaptic pathway (EC-DG-CA3-CA2-CA1). However, a pharmacological block of inhibition allows CA3 neurons to activate CA2 neurons. Although CA2 is characterized by a lack of plasticity at SC-CA2 excitatory synapses, we have recently found that there exists an activity-dependent long-term depression (I-LTD) at CA2 inhibitory synapses due to delta-opioid receptor activation. Furthermore, preliminary results reveal that this decrease of inhibition is sufficient to allow CA3 pyramidal neurons to activate CA2 neurons. Moreover, CA1, which constitutes the output of the hippocampus, is connected by both CA2 and CA3. We perform extracellular and whole cell recordings in area CA1 of hippocampal slices from 5-7 week-old mice.We have found that a decrease in inhibition in CA2 directly translates to an increase in firing in CA1 pyramidal cells. To investigate the consequences of the modulation of the inhibition/excitation balance in CA2, we examine how the activity of CA1 neurons is altered before and after induction of I-LTD in CA2.

P2.019
Learning induced LTP in mice

Remaud, J. (Toulouse)[1], Roumier, F. (Toulouse)[1], Pech, S. (Toulouse)[1], Francès, B. (Toulouse)[1], Dahan, L. (Toulouse)[1]
[1]*Univ. Paul Sabatier - Bât. IVR3 - B3, CNRS UMR 5169 (CRCA), Mémoire, Plasticité, Vieillissement, Toulouse, France*

Long-term potentiation (LTP) is a process by which the strength of the synaptic connection between neurons increases when the synapse is repeatedly activated. It can be “artificially” triggered by electrical stimulation protocols such as high frequency or theta burst stimulations (TBS). There is now a strong body of evidence demonstrating common molecular mechanisms underlying LTP and long term memory, and LTP is currently the best candidate for a neural correlate of memory.
However, only few studies succeeded in showing that LTP actually occurs in the hippocampus during learning. We recorded evoked field potentials (fEPSP) in area CA1 of the hippocampus in freely moving mice before and after a session of contextual fear conditioning. Here we report that this kind of electrophysiological measurement allows monitoring the “natural” LTP triggered by the training session. Moreover we show that conditioning-induced LTP partially occludes TBS-induced LTP, suggesting that natural and artificial LTP share some mechanisms.
This study provides the first electrophysiological evidence of a robust LTP induced by a single session of learning in mice.

P2.020
Pre and Post synaptic NMDA effects targeting Purkinje cells in the mouse cerebellar cortex

Bossu, J.L. (Strasbourg)[1], Dupont, J.L. (Strasbourg)[1], Poulain, B. (Strasbourg)[1], Doussau, F. (Strasbourg)[1], Valera, A. (Strasbourg)[1]
[1]*INCI, UPR 2356, Strasbourg, France*

N-methyl-D-aspartate (NMDA) receptors are associated with many forms of synaptic plasticity. Their expression level and subunit composition undergo developmental changes in several brain regions. In the mouse cerebellum, beside a developmental switch between NR2B and NR2A/C subunits in granule cells, functional postsynaptic NMDA receptors are seen in Purkinje cells of neonate and adult but not juvenile rats and mice. Nevertheless whereas NMDA receptor subunits are detected on parallel fiber terminals, a presynaptic effect of NMDA on spontaneous release of glutamate has not been demonstrated. Using acute slices, and whole cell patch clamp recordings of Purkinje cells we demonstrate a facilitatory presynaptic effect of NMDA on glutamate release by parallel fibers.

However, we show that this presynaptic effect is only observed for slices prepared from juvenile but not from adult mice. The mechanisms of action of NMDA on presynaptic terminals were investigated. Several pathways such as a depolarization leading to activation of voltage gated calcium channels and or a calcium entry via the NMDA receptors were under study. Using specific antagonists we plane to determine the subunits composition of the presynaptic NMDA receptor

P2.021

Expression, distribution and dystrophins roles in angiogenesis in rats subjected to water stress

Sifi, M. (Alger)[1], Dorbani-Mamine, L. (Alger)[1]
[1]*Université des sciences et technologie Haouari Boumedienne, Faculté des Sciences Biologiques, Alger, Algeria*

The hypothalamic-neurohypophyseal region is extremely sensitive to changes in physiological and hormonal, it is our choice model. This system undergoes neurochemical and morphological alterations remarkable reversible, associated with well-defined physiological stimuli such as dehydration. This system consists of magnocellular neurons whose cell bodies are grouped mainly in the supraoptic hypothalamic nuclei (NSO) and paraventricullaires (NPV). The axons of these neurons through the median eminence and neural lobe projecting into the pituitary where they secrete two neurohormones, vasopressin (AVP) and oxytocin (OT) involved in the regulation of mineral balance and vasoconstriction . AVP, in addition to its roles in homeostasis hydromineral, is known to induce constriction of afferent arterioles to hypothalamic nuclei, due to water stress. Associated with hypoxia, stress triggers angiogenesis (Alonso et al, 2005). At every level crossing magnocellular neurons, they are closely associated with glial cells (Swanson and Sawchenko 1983; Hatton, 1990).
Our focus is on the study of the involvement of dystrophins (Dps) and their associated proteins (DAP) in the phenomenon of angiogenesis, both in neuronal, glial and endothelial cells under conditions of stress ingestion of hypertonic saline solutions of different durations. By analogy with the roles of (Dp) and (DAPs) that are assigned in the muscle cell, the Dp with DAPs are supposed to play an essential role in cellular plasticity in the process of secretion of AVP and transduction signal.
Keywords: dystrophins, angiogenesis, neurohypophyseal axis hypothalamus, water stress

P2.022

Corridor guidepost cells for thalamic axons contribute to fear circuits

Deck, M. (Paris)[1], Tinterri, A. (Paris)[1], Mailhes, C. (Paris)[1], Kessaris, N. (London)[2], Bielle, F. (Paris)[1], Garel, S. (Paris)[1]
[1]*Institut de Biologie de l'Ecole Normale Supérieure, INSERM U 1024, CNRS UMR 8197, Brain Development and Plasticity, Paris, France,* [2]*University College London - Wolfson Institute for Biomedical Research, Department of Cell and Developmental Biology, London, United Kingdom*

Guidepost cells are positioned at critical decision points along the path of axons and provide them with guidance information, in the form of secreted cues or via axonal contact. While the formation of major forebrain projections relies on guidepost neurons, there is to date little information either on how they act or on their destiny. We recently identified a neuronal population, corridor cells, that acts as guidepost for thalamic axons, the main input for the neocortex. These cells exist in non-mammalian vertebrates, but they exert an axonal guidepost function only in mammals. This observation intriguingly suggests that they may also contribute to brain structures independently of their guidepost activity. To follow the final placement and fate of corridor cells in the adult brain, we took advantage of

several mouse lines, including *Islet1-cre, Nkx2.1-cre, Dlx1-floxVenusflox* mice. This experimental strategy allowed us to genetically trace corridor cells in mouse embryos and to investigate at the cellular and molecular level their morphology and fate at different developmental stages. Our results surprisingly show that corridor cells contribute to nuclei of the central extended amygdala. This superstructure of the basal ganglia is composed of several nuclei and is involved in the circuitry of reward and fear. Overall our work shows that guidepost cells are not only transient actors in brain wiring and suggest an evolutionary conserved role in fear related network that is independant from their acquired guidepost function.

P2.023

Astrocyte transcriptome from Mecp2-deficient mice: relevance to the pathogenesis of Rett syndrome

Delepine, C. (Paris)[1,2], Nectoux, J. (Paris)[1,2,3], Letourneur, F. (Paris)[1,2], Chelly, J. (Paris)[1,2,3], Bienvenu, T. (Paris)[1,2,3]
[1]Laboratoire de Génétique des Maladies Neurodéveloppementales, (UMR 8104), Université Paris Descartes, Institut Cochin, Paris, France, [2]Inserm U 1016, Paris, France, [3]Assistance Publique - Hôpitaux de Paris, Hôpital Cochin, Laboratoire de Biochimie et Génétique Moléculaire, Paris, France

Mutations in the gene encoding the transcriptional modulator methyl-CpG binding protein 2 (MeCP2) cause the neurodevelopmental disorder Rett syndrome which is one of the most frequent causes of intellectual disability with genetic origin in women. Although MeCP2 levels are roughly five-fold lower in astrocytes than in neurons, recent studies showed that Mecp2 loss in astrocytes contributes to Rett-like symptoms and restoration of Mecp2 can rescue some of these defects. Moreover, mutant astrocytes have a non-cell autonomous effect on neuronal properties, probably resulting from aberrant secretion of soluble factor(s). To identify these soluble factors, we have compared the gene expression profiles of wild type and mutant astrocytes from Mecp2 308/y mice using Affymetrix mouse 2.0 microarrays. The results obtained were confirmed by quantitative real-time RT-PCR. A false discovery rate (FDR) of 0.05 was applied to the lists of differentially expressed genes between wild-type and mutated samples. 2152 genes passed the stringent FDR filter of 0.05 including 1784 coding transcripts. However, only 80 also had an expression fold change >1.25 in Mecp2 308/y cells versus Mecp2 +/ y cells. Interestingly, among these genes, several genes encode secreted proteins such as myocilin, lipocalin 2, SHH or Wnt7b, implicated in neuronal maturation. Others contribute to major astrocytic functions such as the exocytotic release of glutamate (Slc17a8), glutamate metabolism(GAD1) and glucose metabolism (Adcy8, Acot4). We need to confirm these data at the proteomic level in astrocyte and conditional medium from symptomatic Mecp2-deficient mice. Therefore, insights into astrocyte secretion are critically important for understanding physiological responses and pathological mechanisms in Rett syndrome.

P2.024

Striatal plasticity in a rat model of Parkinson's disease induced by a 6-OHDA lesion in the subtantia nigra and α synuclein overexpression in the cerebral cortex

Muriel, M.-P. (Paris)[1], Ortega Perez, I. (Paris)[2], Decressac, M. (Lund)[3], Mattsson, B. (Lund)[3], Björklund, A. (Lund)[3], Hirsch, E. (Paris)[4]
[1]Hôpital de la Pitié-Salpétrière, INSERM UMRS 975-CNRS UMR 7225-CRIM, Institut du cerveau et de la moelle épinière, Paris, France, [2]CRICM, UPMC/CNRS UMR 7225, INSERM UMRS 975, Experimental Therapeutics of Neurodegeneration, Paris, France, [3]Wallenberg Neuroscience Center, Department of Experimental Medical Sciences, Lund, Sweden, [4]CRICM UMR_S INSERM 975, CNRS 7225, UPMC Univ, Experimental Therapeutics of Neurodegeneration, Paris, France

Parkinson´s disease is characterized by the progressive loss of nigral dopamine neurons. This loss of dopaminergic neurons provokes a dysfunction of the cortico striatal pathway. The disease is also characterized by presence of α-synucleine deposits in the degenerating neurons. While at early stages of the disease α-synucleine deposits are restricted to the lower part of the brain they are invading the cerebral cortex at end stages of the disease. So far, most of the animal models of the disease reproduce the nigrostriatal pathology but not the cortical pathology. The aim of this study was to study develop a model of end stage Parkinson's disease and to study the cortico-striatal plasticity in this model. To develop the model we have injected α synuclein using viral vector in motor cortex of rats with or without unilateral lesion 6-OHDA lesion of the substantia nigra. The site of injection, the loss of dopamine neurons and the localization of α synuclein were assessed using immunocytochemistry. Cortical lesion combined with nigral lesion induced a decrease in the size of corticostriatal fibers and morphological changes in corticostriatal synapses. A quantitative analysis showed an increase in the number and length of perforated asymetric synapses in rats with double nigral and cortical lesion. These morphological modifications of the corticostriatal terminals obtained by double nigral and cortical lesions in rat suggest that this model replicates closely the end stage human pathology of Parkinson disease.

P2.025

Modulation of NMDA receptor function by calsyntenin-1 regulates dendritic spine maturation in CA1 hippocampal pyramidal cells

Ster, J. (Zurich)[1], Steuble, M. (Zurich)[2], Orlando, C. (Zurich)[1], Diep, T.-M. (Zurich)[2], Raineteau, O. (Zurich)[1], Sonderegger, P. (Zurich)[2], Gerber, U. (Zurich)[1]
[1]Brain Research Institute, University of Zürich, Swiss Federal Institute of Technology Zürich, Zurich, Switzerland, [2]University of Zürich, Institute of Biochemistry, Zurich, Switzerland

Calsyntenins are members of the cadherin superfamily of cell adhesion molecules that act as vesicular docking proteins for kinesin-1-dependent plus-end-directed transport along microtubules. Genetic linkage studies in humans and gene deletion experiments in nematodes indicate a role for calsyntenins in learning and memory. We generated a calsyntenin-1 knock-out (Cst-1 KO) mouse to characterize the roles of this protein in synaptic function. We found that the absence of calsyntenin-1 enhanced NMDA receptor-dependent LTP at Schaffer collateral - CA1 pyramidal cell synapses, but not between CA3 pyramidal cells in juvenile mice (P15-P21). The increase in LTP was associated with prolonged kinetics of NMDA receptor-mediated synaptic responses, which reflected increased expression of GluN2B subunit-containing receptors. Quantitative confocal microscopy in hippocampal slices from juvenile Cst-1 KO mice revealed an increase in immature filopodia-like dendritic protrusions at the expense of thin-type dendritic spines. These results are consistent with a critical role for calsyntenin-1 in synapse formation and maturation during the early postnatal period.

P2.026

Early methyl donor deficiency associated with alteration of the miR-124 and Stat3 signaling pathways impairs embryonic brain development

Kerek, R. (Vandoeuvre-les-Nancy)[1], Geoffroy, A. (Vandoeuvre-les-Nancy)[1], Bison, A. (Vandoeuvre-les-Nancy)[1], Pourié, G. (Vandoeuvre-les-Nancy)[1], Guéant, J.-L. (Vandoeuvre-les-Nancy)[1], Bossenmeyer-Pourié, C. (Vandoeuvre-les-Nancy)[1], Daval, J.-L. (Vandoeuvre-les-Nancy)[1]
[1]Fac. de Médecine - Nancy Université- BP 184, INSERM U 954, Nutrition génétique et expositions aux risques, Vandoeuvre-les-Nancy, France

The methyl donors vitamin B12 and folate (vitamin B9) regulate the one-carbon metabolism that plays a key role in transmethylation reactions. Methyl donor deficiency constitutes a risk factor for various diseases, and has been correlated with developmental disturbances such as neural tube defects or abnormalities that can lead to spontaneous abortion.
We have studied the consequences of a periconceptual and gestational deficiency in methyl donors on the development of rat embryos. Additionally, we used an in vitro cell model of neuronal progenitors (H19-7 hippocampal cell line). The morphometric study of E20 fetuses showed a significant decrease in both size and weight of deficient embryos. Growth retardation also concerned the brain, with reduced weight and atrophy of various neurogenic brain structures (hippocampus, subventricular zone).
The study by TaqMan RT-q-PCR of the expression levels of a panel of microRNAs (miRs), which are posttranscriptional regulators of gene expression, showed a significantly increased expression of miR-124 under deficiency conditions in H19-7 cells and in rat fetuses at E16 and E20. In situ hybridization confirmed the increase of miR-124 in mature form in the cytoplasm, while Western blot analyses showed the downregulation of some of its targets, especially those involved in the Stat3 signaling pathways. Stat3 has been implicated in programming gene expression in biological events such as embryonic development, apoptosis, organogenesis and cell growth regulation. Its presence is important for normal brain development and neuroprotection. The expression of Stat3 and its phosphorylation were decreased under methyl donor deficiency, along with reduced proliferation of hippocampal progenitors, inappropriate initiation of differentiation and active cell death by apoptosis. Results show that methyl donor deficiency led to the deregulation of epigenetic mechanisms involving miR-124. This was associated with lower expression and activation of Stat3 and thus of its target genes involved in cell proliferation and survival, processes that could contribute to the altered brain development observed in our animal model.
Keywords: Folate, vitamin B12, development, differentiation, epigenetics.

P2.027

Impact of the mitochondrial fusion protein OPA1 on neuronal oxidative metabolism and synaptic transmission

Miquel, M.-C. (Toulouse)[1,2], Bertholet, A. (Toulouse)[3,4], Millet, A. (Toulouse)[3], Guillermin, O. (Toulouse)[3], Davezac, N. (Toulouse)[3], Daloyau, M. (Toulouse)[3], Tounkara, K. (Toulouse)[3], Chavis, P. (Marseille)[5], Belenguer, P. (Toulouse)[3]
[1]Univ. Paul Sabatier, CNRS UMR 5547-CBD - Bât 4R3B3, Dynamique Mitochondriale, Toulouse, France, [2]Université Pierre & Marie Curie - UPMC, Paris, France, [3]UMR 5547 CNRS Université Paul Sabatier, Toulouse, France, [4]University of California (UCSF), San Francisco, United States, [5]INMED, Marseille, France

Multiple findings have suggested that disruptions of mitochondrial functions and dynamics contribute to neurodegenerative diseases. Mitochondrial functions in neurons include regulation of calcium and redox signaling, developmental and synaptic plasticity and the arbitration of cell survival and death. Mitochondrial dynamics controls the organelle's morphology *via* forces of fusion and fission that are regulated by large dynamin-related GTPases. Loss or mutations of the inner membrane GTPase OPA1 are responsible for type 1 dominant optic atrophy (ADOA1), by mechanisms not fully understood. While links emerge between defects in mitochondrial fusion and neurodegeneration, the processes involved are still largely unknown.
To understand the mechanisms by which alterations of mitochondrial dynamics could contribute to mitochondria dysfunction, eventually leading to neuronal dysfunctions, we studied the effects of OPA1 down-regulation in primary neurons. In cortical neurons, RNA interference of the fusion protein OPA1 led to fragmented mitochondria that became less abundant along the dendrites. Furthermore, this inhibition resulted in reduced expression of mitochondrial respiratory complexes, decreased mitochondrial membrane potential, and diminished ROS levels. The onset of synaptogenesis was markedly impaired *via* reductions in pre- and post-synaptic structural protein expression and synapses number without first affecting the dendritic arborization. With longer time in culture, OPA1 extinction

led to a major restriction of dendritic growth, together with synaptic proteins reduction (Bertholet et al., 2013). Consequences on axonal morphology mitochondria are under study. Finally, electrophysiological recordings showed that OPA1 depletion induced changes in synaptic transmission.
Altogether, our findings suggest a new role for OPA1 in neuronal and synaptic maturation through maintenance of proper mitochondrial oxidative metabolism and distribution, highlighting the role of mitochondrial dynamics in neuronal functioning and providing new insights not only into mitochondrial dynamics-linked neurodegenerative diseases like ADOA1 but to other neurodegenerative pathologies.

P2.028

CK2 phosphorylation of IQCJ-SCHIP1 regulates its interaction with ankyrin G and its accumulation at the AIS

Dargent, B. (Marseille)[1], Debarnot, C. (Marseille)[1], Fache, M.-P. (Marseille)[1], Pipéroglou, C. (Marseille)[1], Garcia Del Cano, G. (Araba)[2], Goutebroze, L. (Paris)[3], Papandréou, M.-J. (Marseille)[1]
[1]Aix Marseille Université, CNRS, CRN2M UMR 7286, Marseille, France, [2]University of Basque Country, Department of Neurosciences, Araba, Spain, [3]UMR-S 839 INSERM UPMC, Institut du fer à Moulin, Paris, France

The axonal initial segment (AIS) is a domain of neurons that plays a central role in the electrogenesis and neuronal polarity. This domain presents a complex molecular organisation that includes sodium and potassium channels, growth and transcriptional factors and cytosqueletal associated proteins. Among these, ankyrin G (Ank G) orchestrates the formation and maintenance of the AIS. In particular, sodium channel location depends on its interaction with AnkG. This association is regulated by channel phosphorylation by CK2, a kinase that also segregates at the AIS (1).
We previously showed that IQCJ-SCHIP1 is enriched in the AIS. This protein that binds calmodulin in the absence of calcium, interacts with AnkG in vitro, but its role is presently unknown (2). Here, our study focuses on the analysis of the mechanisms involved in IQCJ-SCHIP1 segregation at the AIS. We first used an shRNA approach to silence AnkG expression on cultured hippocampal neurons. We showed that this treatment abrogates both AnkG and IQCJ-SCHIP staining at the AIS, suggesting that SCHIP location depends on its interaction with AnkG. We thus further analysed the interaction of IQCJ-SCHIP with AnkG. Using a pull-down assay, we showed that AnkG-GFP expressed in COS-1 cells associated with GST-IQCJ-SCHIP1 phosphorylated by CK2 but not with non-phosphorylated GST-IQCJ-SCHIP1. The effect of CK2 phosphorylation on the interaction between IQCJ-SCHIP1 and Ank G was further analyzed by Surface Plasmon Resonance (SPR). Phosphorylated IQCJ-SCHIP1 interacted with Ank G with a Kd; 10^{-9}M while no binding was observed without CK2 treatment. SPR approaches using IQCJ-SCHIP1 mutants truncated in the C-terminus tail revealed that a segment including several potential CK2 phosphorylation sites is directly involved in the interaction with AnkG. Finally, treatment of cultured neurons with TBB, an inhibitor of CK2, decreased IQCJ-SCHIP staining at the AIS by about 40% but did not affect AnkG staining, demonstrating that IQCJ-SCHIP location depends on CK2 activity.
Altogether these data demonstrate that IQJC-SCHIP1 segregates at the AIS through its interaction with AnkG and that this interaction is strongly regulated by its phosphorylation by CK2.
1: J Cell Biol. 2008, 183:1101
2: J Neurosci. 2008 28:6111

P2.029

Sensory map transfer to the neocortex relies on pre-target ordering of thalamic axons

Lokmane, L. (Paris)[1], Proville, R. (Paris)[1], Narboux-Nême, N. (Paris)[2], Györy, I. (Freiburg)[3], Keita, M. (Paris)[1], Mailhes, C. (Paris)[1], Léna, C. (Paris)[1], Gaspar, P. (Paris)[2], Grosschedl, R. (Freiburg)[3], Garel, S. (Paris)[1]
[1]IBENS, Paris, France, [2]Institut du Fer à Moulin INSERM S 839, Paris, France, [3]Department of Cellular and Molecular Immunology, Max Planck Institute of Immunobiology, Stuebeweg, Freiburg, Germany

Sensory maps, such as the representation of mouse facial whiskers, are conveyed throughout the nervous system by topographic axonal projections that preserve neighboring relationships between adjacent neurons. In particular, map transfer to the neocortex is ensured by thalamocortical axons (TCA), which are topographically organized in response to intrinsic cortical signals. However, TCA already show a topographic order early in development, as they navigate towards their target. Here, we show that this pre-ordering of TCA is required for the transfer of the whisker map to the neocortex. Using a genetic strategy that specifically perturbs the development of an intermediate target, the basal ganglia, we scrambled TCA topography en route to the neocortex without affecting the thalamus or neocortex. Notably, embryonic somatosensory TCA were shifted towards the visual cortex and showed a relative intermixing along their trajectory. We found that somatosensory TCA rewired post-natally to reach the somatosensory cortex, but failed to form a topographic anatomical or functional map. Our study reveals that sensory map transfer relies not only on positional information in the projecting and target structures but also on pre-ordering of axons along their trajectory, thereby opening novel perspectives on brain wiring.

P2.030

Knockdown of the neuronal beta tubulin subunit TUBB3, implicated in human in malformations of cortical development, results in a delay of radial migration in mouse

Saillour, Y. (Paris)[1,2], Bruel-Jungerman, E. (Paris)[3,4,5], Rucci, J. (Paris)[1,2], Poirier, K. (Paris)[1,2], Francis, F. (Paris)[3,4,5], Chelly, J. (Paris)[1,2]
[1]INSERM 1016, Paris, France, [2]Institut Cochin, Université Paris Descartes, Paris, France, [3]INSERM UMR-S 839, Paris, France, [4]Université Pierre et Marie Curie, Paris, France, [5]Institut du Fer à Moulin, Paris, France

Over the last years, the critical role of the cytoskeletal network in cortical development has been established. The importance of microtubules was further emphasized with the association of mutations in genes encoding alpha-tubulin (*TUBA1A*, *TUBA8*), and beta-tubulin (*TUBB2B*) in lissencephalies and polymicrogyria (Keays 2007, Poirier 2007, Jaglin 2009, Abdollahi 2009) and *TUBB5* in microcephaly with cortical gyration abnormalities. We have also reported the involvement of *TUBB3* missense mutations in polymicrogyria and cortical simplification in 6 different families. We investigate here the association between TUBB3 dysfunction and these malformations of cortical development (MCDs) by analysing the consequences of *Tubb3* downregulation on cortical radial neuronal migration in mice induced by *in utero* RNA interference.
Our studies show that *in utero* expression of a *Tubb3* 3'UTR-shRNA induced a significant arrest of cells within the sub-ventricular zone/intermediate zone during the mouse cortex development (E18.5). We observed that the majority of the inactivated blocked cells display multipolar or round shapes. The overexpression of a wild type human TUBB3 cDNA significantly rescues the shRNAi-induced neuronal migration defects while MCD-related TUBB3 missense mutation cDNA overexpression does not rescue the migration defect. However overexpression of TUBB3 mutants affects the final position of neuronal cells in the cortical plate suggesting therefore a potential dominant negative effect of TUBB3 mutations.
Secondly, we tested the functional redundancy of the beta tubulin subunits and show that TUBB2B and TUBB4 only partially rescue the sh-*Tubb3* induced migration phenotype suggesting a specific function of *Tubb3* in the neuronal migration which cannot be replaced by the other tested beta tubulins.
This study confirms the crucial role of TUBB3 in radial migration and highlights the functional specificity of the beta tubulin subunits.

P2.031

The GTPase RhoQ/TC10 is essential for dendritic spine morphogenesis in cerebellar Purkinje neurons

Schmidt, S. (Montpellier)[1], Jaudon, F. (Montpellier)[1], Wehrlé, R. (Paris)[2], Vodjani, G. (Paris)[3], Doulazmi, M. (Paris)[2], Vandermoere, F. (Montpellier)[4], Dusart, I. (Paris)[2], Debant, A. (Montpellier)[1]
[1]CRBM CNRS UMR 5237, Montpellier, France, [2]CRICM UMR_S INSERM 975, CNRS 7225, UPMC Univ, Paris, France, [3]UPMC University, Paris, France, [4]IGF, Montpellier, France

By regulating actin cytoskeleton dynamics, Rho family GTPases are implicated in various aspects of neuronal differentiation, but their function during postnatal Purkinje cell development remains poorly understood. Purkinje neurons are crucial elements in the synaptic network of the cerebellum and they have the most spectacular dendritic tree among central nervous system neurons. Despite considerable interest in the development of these cells, the signalling networks involved in their postnatal differentiation remain elusive.
In order to identify novel regulators of postnatal Purkinje cell differentiation among proteins of the Rho GTPase signaling pathway, we have performed a gene expression profiling of all mammalian Rho GTPases, using real-time quantitative PCR on mRNA from FACS-purified Purkinje cells, at different postnatal stages. We selected the Cdc42-family GTPase RhoQ (TC10) as a strong candidate, as its expression increased dramatically during the period corresponding to the morphogenesis of dendrites and spines. Depletion of RhoQ in cerebellar organotypic cultures led to a complete absence of spines along the dendrites, which could be rescued by reintroducing RhoQ in the cells, implicating RhoQ as an essential actor of spinogenesis. We further identified the complex composed of bPIX/GIT1/PAK/Shank as downstream effectors of RhoQ in the cerebellum. This complex has been shown essential for the formation of dendritic spines in other neuronal types. We have therefore established an essential and novel function for RhoQ and the bPIX/GIT1/PAK/Shank complex in the regulation of postnatal Purkinje cell development and dendritic spine morphogenesis.

P2.032

Responses mediated by group I metabotropic glutamate receptors at recurrent synapses between CA3 pyramidal cells

Rosenberg, N. (Zürich)[1]
[1]University of Zurich, Zürich, Switzerland

Several studies have reported the presence and described functional roles of group I metabotropic glutamate receptors (mGlu1 and mGlu5) at synapses formed by mossy fibers on hippocampal CA3 pyramidal cells. A previous study utilizing an indirect approach concluded that group I mGlu receptors do not mediate synaptic responses at recurrent synapses between neighboring CA3 pyramidal cells. Using the whole cell patch clamp technique and extracellular stimulation, we demonstrate for the first time the presence of mGlu receptor-mediated current at CA3-CA3 pyramidal cell synapses (42 ± 9 pA for 30 µA stimulation, n = 8). During application of the specific mGlu5 antagonist (MPEP, 10 µM), we observed a minor but significant decrease of the current (-21 ± 0.26 %, n = 6) whereas administration of the mGlu1 antagonist (YM298198, 1µM) exhibited a sharp reduction of the current at these synapses (-40 ± 0.49 %, n = 6). Next, we investigated whether these receptors at CA3-CA3 pyramidal cell synapses contribute to synaptic plasticity. We found that pharmacological activation of group I mGlu receptors induces LTD of unitary responses in CA3 pyramidal cell connected pairs.

In summary, our results provide evidence for the presence of both receptors, mGlu1 and mGlu5, at recurrent collateral synapses with a stronger expression of mGlu1 and an involvement of group I mGluR in LTD.

P2.033

CK2 accumulation at the axon initial segment is dependent on sodium channel Na_v1

Hien, Y.E. (Marseille)[1], Montersino, A. (Marseille)[1], Castets, F. (Marseille)[1], Leterrier, C. (Marseille)[1], Filhol-Cochet, O. (Grenoble)[2], Vacher, H. (Marseille)[1], Dargent, B. (Marseille)[1]
[1]Aix Marseille Université, CNRS, CRN2M UMR 7286, Marseille, France, [2]INSERM, U 873, CEA, iRTSV/LTS, Grenoble, France

In neurons, action potential generation requires a high concentration of voltage-gated sodium channel Nav1 at the axon initial segment (AIS). The precise accumulation of Nav1 results from a direct interaction between the ankyrin-binding motif of Nav1 and ankyrin G (ankG). Notably this interaction is regulated by protein kinase CK2 phosphorylation. CK2 is highly enriched at the AIS where it most likely phosphorylates the ankyrin-binding motif of Nav1 channels. The pharmacological inhibition of its activity induces a loss of both Nav1 and AnkG accumulation. To further strengthen the role of CK2 in regulating AIS protein interactions/accumulations, we addressed the question of how CK2 is concentrated at the AIS.
Using a shRNA approach, we observed that Nav1 inhibition induces a significant reduction of CK2 concentration at the AIS, and also that Nav1-depleted neurons have a decreased ankG staining at the AIS. Thus, to distinguish the between the effects of Nav1 and ankG depletion on CK2 concentration at the AIS, we used a Kv2.1-Nav1 chimeric channel that reduces Nav1 accumulation at the AIS without depleting ankG. Hippocampal neurons transfected with Kv2.1-Nav1 show a robust reduction of CK2 staining at the AIS, implying that CK2 forms a complex with Nav1. To test this possibility, GST pull-down assays were performed using GST-CK2 and the intracellular loop II-III of Nav1.2 fused to GFP (GFP-Nav1.2 II-III). Our results show that GST-CK2 is specifically able to bind to GFP-Nav1.2 II-III. Additionally, to better understand the role of CK2-phosphorylation of Nav1 channels, we generated phosphospecific antibodies against the serine 1112 of Nav1.2. This serine is located in the Nav ankyrin-binding motif, and was shown to be phosphorylated by CK2 in vitro. We now demonstrate that Nav1 are natively phosphorylated at serine 1112 in rat brain as well as in cultured hippocampal neurons, and that this phosphorylation follows Nav1 expression levels during neuronal development. In conclusion, we demonstrated that CK2 accumulation at the AIS depends on Nav1 channels, with which they form tight complexes. The existence of such complexes likely leads to Nav1-phosphorylation during neuronal development and, consequently, Nav1-membrane anchoring by enhanced binding to ankG.

P2.034

Implication of the dementia-related protein Chmp2B in synaptic plasticity

Chassefeyre, R. (Grenoble)[1], Martinez-Hernandez, J. (Grenoble)[1], Bertaso, F. (Montpellier)[2], Bouquier, N. (Montpellier)[2], Couté, Y. (Grenoble)[3], Blot, B. (Grenoble)[1], Pernet-Gallay, K. (Grenoble)[1], Sadoul, R. (Grenoble)[1], Fagni, L. (Montpellier)[2], Goldberg, Y. (Grenoble)[1]
[1]Grenoble Institut des Neurosciences, Grenoble, France, [2]Institut de Génomique Fonction - Univ. de Montpellier 1 & 2, Montpellier, France, [3]Institut de Recherches en Technologies et Sciences pour le vivant U 1038 INSERM/CEA/UJF, Grenoble, France

The Charged Multivesicular body Protein 2B (Chmp2B) is a subunit of the ubiquitous ESCRT-III complex, a cytoplasmic entity responsible for membrane deformation and scission in endosomal sorting and other cellular activities. Dominant mutations in human CHMP2B underlie a rare familial form of the neurodegenerative disease fronto-temporal dementia (1). In cultured rat hippocampal neurones, knock-down of Chmp2B impairs the morphological maturation of dendritic spines, without affecting neuronal survival (2). Overexpressed wild-type Chmp2B assembles into structures that adhere to and deform the plasma membrane, suggesting that Chmp2B could function as a membrane scaffold (3).
Here we present evidence that Chmp2B is involved in synaptic plasticity. Chmp2B immunoreactivity is strongly enriched in specific brain regions, including cortex, hippocampus, and olfactory bulb. In hippocampal neurones in culture or in situ, the protein is distributed throughout the somatodendritic domain, including the dendritic spines. Immuno-electron microscopy indicates that Chmp2B concentrates just beneath the spine plasma membrane, around the post-synaptic density. Biochemical fractionation, co-immunoprecipitation and proteomic analysis demonstrate that synaptic Chmp2B is part of a large supramolecular assembly which contains other subunits of ESCRT-III, as well as NMDA receptors and a range of other synaptic proteins. Consistent with published results, in cultured hippocampal neurones transfected with a control shRNA plasmid, stimulation of post-synaptic NMDA receptors by exogenous glycine induced long-term potentiation of miniEPSC amplitude and frequency, and also caused a stable increase in median spine volume measured by confocal microscopy. Both of these effects were abrogated in neurones transfected with an shRNA plasmid targeting Chmp2B. These results suggest that Chmp2B and ESCRT-III may be required for coupling NMDA receptors to plasticity-related effectors, the identity of which is currently being investigated.
(1) Skibinski et al., Nat. Genetics (2005), 37(8):806-8.
(2) Belly et al., J. Cell Sci. (2010), 123:2943-54.
(3) Bodon et al., J. Biol. Chem. (2011), 286(46):40276-86.

P2.035
Role of IL1RAPL1 in synaptic function

Ramos, M. (Paris)[1], Nole, C. (Paris)[1], Chelly, J. (Paris)[1], Dorseuil, O. (Paris)[1], Billuart, P. (Paris)[1]
[1]*Institut Cochin, INSERM U 1016, Paris, France*

Interleukin-1 receptor accessory protein like 1 (*IL1RAPL1*) gene, a member of interleukin-1 receptor family expressed in the brain, has been associated with intellectual disability and autism spectrum disorder. The mouse model of IL1RAPL1 loss of function shows altered learning and memory processes, a reduction of dendritic spine density in hippocampus, and a deficit in long-term plasticity. To better define the pathophysiology of this genetic condition and to understand the specific role of IL1RAPL1 in synapse formation, signaling and function, different IL1RAPL1 mutations have been identified from patients and their consequences have been analyzed.
Using mouse hippocampal neurons we examined the effect of IL1RAPL1 mutants over-expression on synapse formation and maturation. Preliminary results show that some mutations impair the increase in excitatory synapse number observed by IL1RAPL1 WT overexpression.
We have previously shown that in neurons, IL1RAPL1 is required for IL-1β-triggered JNK activation, but not for activation of p38 or NFκB. In addition, our group described the interaction of IL1RAPL1 with PSD-95, a major component of excitatory synapses. This interaction targets PSD-95 to synapses and regulates its phosphorylation through JNK activation. As PSD-95 and JNK are known to regulate membrane trafficking of AMPA receptor subunits, we are currently exploring the role of IL-1β and IL1RAPL1 signaling in AMPAR regulation.
This study improves the understanding of synapse establishment and function, and will help to correlate specific IL1RAPL1 mutations with the cognitive impairments observed in patients.

P2.036

A developmental program sets left-right asymmetry of phrenic motoneurons and connectivity

Charoy, C. (Villeurbanne)[1], Chaix, Y. (Villeurbanne)[1], Morle, L. (Villeurbanne)[1], Durand, B. (Villeurbanne)[1], Skidmore, J. (Ann Harbor)[2], Martin, D. (Ann Harbor)[2], Falk, J. (Villeurbanne)[1], Castellani, V. (Villeurbanne)[1]
[1]CNRS, Université Claude Bernard Lyon1, CGphiMC, Villeurbanne, France, [2]University of Michigan Medical Center, Pediatrics and Communicable Diseases and Human Genetics, Ann Harbor, United States

The diaphragm is a respiratory muscle composed of a central tendinous region and two lateral muscles innervated by ipsilateral pools of cervical motoneurons forming the left (L) and right (R) phrenic nerves. Although apparently symmetric, both lateral muscles and phrenic nerves indeed exhibit left-right (L-R) asymmetric features, which we observed are highly stereotyped and present in the embryo from the onset of target innervation. We explored the developmental program controlling these asymmetries, concentrating on the symmetry-breaking Nodal signaling, setting L/R asymmetry of the visceral organs in the early embryo. Analysis of mouse lines in which the Nodal signaling is disturbed demonstrated the implication of this cascade and provided evidence that the L-R asymmetry of the diaphragm nerve pattern is encoded by the motoneurons. Additional approaches including microarray and ex vivo culture models were undertaken to further explore the molecular mechanisms governing the setting of the L-R asymmetry. Our data support the existence of yet unreported left-right identity of phrenic motoneurons.

P2.037

Characterization of paraventricular monoaminergic cells: evolutionary and developmental perspectives

Yamamoto, K. (Gif sur Yvette)[1], Affaticati, P. (Gif sur Yvette)[1], Fontaine, R. (Gif sur Yvette)[1], Bureau, C. (Gif sur Yvette)[1], Vernier, P. (Gif sur Yvette)[1], Demarque, M. (Gif sur Yvette)[1]
[1]CNRS Gif-sur-Yvette, Neurobiologie & Développement (NED), Gif sur Yvette, France

Tyrosine hydroxylase (TH), the rate-limiting enzyme for dopamine (DA) synthesis, is often used as a marker for dopaminergic cells. TH immunoreactivity (TH-IR) generally overlaps with DA-IR except one cell population, the paraventricular organ (PVO), found in all non-mammalian vertebrate classes. These cells are DA-IR but negative for TH-IR, hence often referred to as DA-accumulating cells.
The TH gene was duplicated in the early stage of vertebrate evolution. The recent characterization of the second TH gene, TH2 led us to re-examine the properties of these cells. We report that PVO cells in chick and zebrafish express TH2, suggesting that they indeed synthetize DA. This information has been overlooked probably because commercially available anti-TH antibodies do not recognize the TH2 protein. In zebrafish we also found that these cells express VMAT2, vesicular monoamine transporter, confirming that they could use DA as a neurotransmitter.
Serotonin (5HT) is another monoamine neurotransmitter. Although it is often considered to be separated system with DA, all the components of metabolic pathways are shared between DA and 5HT, except the first synthetic enzyme (TH and tryptophan hydroxylase; TPH). The TPH gene was also duplicated in the early stage of vertebrate evolution, bearing TPH1 and TPH2. While TPH2 is the major paralog expressed in the brain and TPH1 is expressed only in PNS in mammals, TPH1 is expressed in hypothalamic nuclei in other vertebrates.
Using in situ hybridization and immunohistochemistry, we found that some of the PVO cells are both dopaminergic and serotoninergic, in chick and zebrafish. Thus it is likely that the presence of DA and 5HT co-containing cells is ancestral to the divergence of the vertebrate lineage.
Taking advantage of VMAT2-GFP transgenic line, we examined the development of these cells in zebrafish. The expression of VMAT2 in PVO cells started around 24 hours post-fertilization. Interestingly these cells do not express Hu, a panneuronal marker, nor Elavl3, mRNA coding the Hu protein. Further experiments are planned to examine the peculiar properties of these cells. To

determine how this dual neurotransmitter phenotype is regulated over time and in response to change in electrical activity is of particular interest.

P2.038

A kif5/synaptosomal associated protein of 23 kda (snap-23) complex mediated vesicular transport of metabotropic glutamate receptors mGluR1a in glial and neuronal cells

Homburger, V. (Montpellier)[1], Raynaud, F. (Montpellier)[1], Belhamici, Y. (Montpellier)[1], Thibert, E. (Montpellier)[1], Carrette, J. (Montpellier)[1], Bockaert, J. (Montpellier)[1], Fagni, L. (Montpellier)[1]
[1]*CNRS-UMR5203, Institut de Génomique Fonctionnelle, Montpellier, France*

Metabotropic glutamate receptors (mGluRs) are expressed in glial cells and neurons. In polarized neurons, mGluR7a and mGluR1a are localized in pre- and post-synapse, respectively. To reach their final destination, they should be transported into distinct post-Golgi carriers. Different microtubule-based motors and adaptor proteins contribute to the appropriate trafficking of ionotropic glutamate receptors in both axons and dendrites, but for mGluRs this issue is still unresolved. Our work focused on molecular motors and adaptors that are responsible for mGluR1a transport.
Transport and exocytosis were assessed in glial and neuronal cells expressing mGluRs tagged with a N-terminal fluorescent protein (YFP or mcherry), selectively cleavable from outside the cell by thrombin. MGluR1a post-Golgi traffic was synchronized by temperature-induced block of trans-Golgi network exit. We showed that mGluR1a post-Golgi transport used mobile vesicles as motorized carriers along microtubules. Kinesin kif5 was identified by mass-spectrometry as a motor involved in this transport. Snap-23, a ubiquitous soluble N-ethylmaleimide-sensitive factor attachment protein receptor (SNARE) protein was also identified as a linker between mGluR1a and kinesin kif5. These results were confirmed by cell surface receptor restoration experiments using dominant-negative kif5 and snap-23 mutants. Immunoprecipitation and dual-color, time-lapse imaging experiments showed the presence of mGluR1a, snap-23 and kif5 on the same vesicular complex and their co-transport. Moreover, mGluR1a receptor C-terminus was required for mGluR1a vesicular transport as indicated by mutations and deletion experiments. BRET experiments also suggested a direct interaction between mGluR1a and snap-23, but not with kinesin kif5.
In conclusion, this work revealed that mGluR1a post-Golgi carriers contained a pre-assembled complex bearing a kinesin (kif5) for motorized transport and a SNARE protein (snap-23) for vesicle fusion. Further work will be required to precise and define protein interaction domains.

P2.039

Crosstalk between GPCRs: functional selectivity mechanism of 5-HT2A receptor under the control of mGlu2 receptor

Prézeau, L. (Montpellier)[1], Bologna, J.-C. (Montpellier)[1], Faraki, S. (Montpellier)[1], Becamel, C. (Montpellier)[1], Pin, J.-P. (Montpellier)[1], Marin, P. (Montpellier)[1]
[1]*Institut de Génomique Fonction - Univ. de Montpellier 1 & 2, Cnrs Umr 5203-Inserm U661, dépt. de Pharmacologie Moléculaire, Montpellier, France*

These last years, two new concepts have emerged in the field of G-protein coupled receptor (GPCR): 1- functional selectivity (and biased agonism) means that GPCRs couple specific sets of signaling pathways depending on the ligand they are activated by; 2- a GPCR crosstalk is the regulation of a GPCR activity and signaling by another receptor, involving a physical association or not.
Here, we described that a crosstalk between two GPCRs, the serotoninergic 5-HT2A and the glutamatergic mGlu2 receptors, led to a functional selectivity of 5-HT2A signaling under the control of the activity of mGlu2.

5-HT_{2A} is involved in mental disorders (schizophrenia, anxiety, or depression), and is the target of antipsychotic and antidepressant antagonist compounds. 5-HT2A is activated by two types of agonists, hallucinogens (LSD or DOI) and compounds devoid of hallucinogenic effects (Lisuride, ergotamine). All agonists induce a Gq pathway, but hallucinogens activate also Gi/o and src pathways leading to specific expression of a set of transcription factors.
mGlu2 agonists display antipsychotic activity too, and decrease DOI-induced hallucinogenic behaviors in rodents. It has been proposed that this would be explained by mGlu2 / HT2A receptor complexes in frontal cortex pyramidal neurons. Actually, co-expression of mGlu2 enhanced the 5-HT2A-driven DOI-induced Gi activation, while activation of mGlu2 suppressed DOI effect. These data suggested that mGlu2 control 5-HT2A DOI-induced Gi responses.
However, it was not known whether the effect of another ligand like the endogenous ligand 5-HT would be regulated the same way, and whether the Go pathway would also be controlled by mGlu2. Moreover, we intended to address the involvement of mGlu2 coupling in its regulatory action.
We showed that 5-HT, like DOI, was able to activate Gi and Go pathways even in absence of mGlu2. Moreover, we showed a differential action of mGlu2 on 5-HT2A coupling depending the 5-HT2A ligand: both 5-HT and DOI-induced Go activation was abrogated by mGlu2 activation, whereas only the DOI-induced Gi activation was blocked, not 5-HT's one. Thus, depending on its activation state, mGlu2 induced biased agonism (5-HT vs DOI) and functional selectivity towards the 5-HT2A ability to activate the Gi and Go pathways.

P2.040

Melanocortin receptor type 4 modulates voltage operated calcium channels by G protein signaling

Agosti, F. (La Plata)[1], López Soto, E.J. (La Plata)[1], Rodriguez, S.S. (La Plata)[1], Perello, M. (La Plata)[2], Raingo, J. (La Plata)[1]
[1]IMBICE CIC-CONICET (Instituto Multidisciplinario de Biología Celular), Electrophysiology Lab., La Plata, Argentina, [2]IMBICE CIC-CONICET (Instituto Multidisciplinario de Biología Celular), Neurophysiology Lab., La Plata, Argentina

Melanocortin receptor type 4 (MC4R) is a G protein coupled receptor highly expressed in specific sites of the brain. MC4R binds to α-MSH and its activation impact in many aspects of neuron function, including synaptic activity. Moreover, MC4R exhibits basal activity. Presynaptic neuronal voltage operated calcium channels (VOCC) control neurosecretion. Two different genes encode the presynaptic VOCCs CaV2.1 and CaV2.2. These channels are highly sensitive to G protein coupled receptors mediated modulation. Here we investigate how MC4R basal and evoked activity impact on CaV2.1 and CaV2.2 function and the pathways involved. We performed whole cell patch clamp in transfected HEK293 cells co-expressing CaV2.1 or CaV2.2, and a MC4R-containing plasmid with the soluble GFP sequence (to detect transfected cells). We found that MC4R co-expression specifically reduced CaV2.1 basal calcium currents in a 38%, relative to the control condition expressing only CaV2.1, while it did not alter CaV2.2 basal current levels, suggesting a specific effect of MC4R basal activity on CaV2.1 function. When we applied the MC4R agonist melanotan-II (MTII) to HEK293 cells co-expressing CaV2.2 and MC4R, we found a concentration dependent inhibitory effect on CaV2.2 currents (EC50~36 nM). When we evaluated MTII effect on 5 cells co-expressing CaV2.1 and MC4R that displayed calcium currents (ranging 60-100 pA) we found no effect of MTII at saturating dose (100 nM). When we co-express RGS2 (a GTPase protein that prevent Gq and Gs actions) we found that MC4R evoked activity is mediated by Gq and/or Gs proteins, and MC4R basal activity is independent of Gq and Gs proteins. Our results suggest that MC4R activity has a differential effect on the two major presynaptic calcium channels: CaV2.1 is inhibited by basal MC4R activity and CaV2.2 is sensitive to agonist-evoked MC4R activity. Supported by CONICET (EJLS, MP and JR), CIC-PBA (FA and SR), PICT2010-1589 (JR) and PICT2010-1954 (MP).

P2.041

Advancing the frontiers. Border control at the motor neuron domain by ephrinB2

Laussu, J. (Toulouse)[1], Davy, A. (Toulouse)[1]
[1]Centre de Biologie du Développement, Toulouse, France

The mammalian central nervous system (CNS) is composed of hundreds of different neuronal subtypes that must be produced in the appropriate number and at the appropriate time and place. Alteration in neuronal production, either in terms of number or subtype, leads to modifications in neuronal network wiring that may adversely impact sensory perception, motor function or behavior. Eph:ephrin signaling is involved in various aspects of the nervous system development, with a prominent role in topographic mapping and axon guidance. Here we explored the implication of this signaling pathway in controlling the production of motor neurons innervating the limb. First, using a GFP reporter mouse line we show that ephrin-B2 is expressed in the specific population of progenitors which gives rise to these motor neurons. Second, we used the Cre-lox technology to specifically excise EfnB2 in motor neuron progenitors and show that targeted loss of EfnB2 entails a significant decrease in the number of these progenitors and their progeny. Lastly, we provide evidence that conditional loss of pMN progenitors is not due to cell death or premature differentiation but correlates with changes in the patterning of the ventral neural tube. Altogether, our results indicate that ephrinB2 is required in pMN to produce the correct number of motor neurons and oligodendrocytes and highlight the complex role of Eph:ephrin signaling in the developing nervous system.

P2.042

Control of synaptic transmission by tissue non-specific alkaline phosphatase in the cerebral cortex

Nowak, L.G. (Toulouse)[1], Rosay, B. (Toulouse)[1], Czégé, D. (Budapest)[2], Világy, I. (Budapest)[2], Frances, B. (Toulouse)[3], Millán, J.L. (La Jolla)[4], Fonta, C. (Toulouse)[1]
[1]CNRS - Université de Toulouse, Laboratoire Cerveau et Cognition, Toulouse, France, [2]Eötvös University, Budapest, Hungary, [3]CNRS - Université de Toulouse, Centre de Recherches sur la Cognition Animale, Toulouse, France, [4]Burham Institute, La Jolla, United States

Alkaline Phosphatase (AP) activity has been revealed within the brain tissue of mammals, including human and non-human primates (Fonta et al. 2004; Négyessy et al. 2011).
We previously showed that AP activity is extracellularly localized in the synaptic cleft and at the nodes of Ranvier. We also found that AP activity is regulated by neuronal activity, as demonstrated by sensory deprivation experiments (Fonta et al. 2004). These findings strongly suggest that AP is involved in neurotransmission and cerebral cortex functions. We recently reported that AP activity of the neuronal cells can be attributed to the Tissue Non-specific Alkaline Phosphatase (TNAP) gene expression rather than to other Alkaline Phosphatase genes (Brun-Heath et al. 2011). In view of the neurological symptoms accompanying human TNAP deficiencies - epilepsy in particular -, we aimed at further investigating the functions of TNAP in the brain.
First, we examined the role of TNAP in controlling neurotransmitter synthesis. TNAP supposedly regulates the intracellular concentration of PLP (vitamin B6), which is a cofactor for numerous enzymes involved in neurotransmitter synthesis. We used the TNAP-KO (Akp2-/-) mice model that mimics the severe form of human hypophosphatasia - including epilepsy and early postnatal death - to measure the neurotransmitter content by HPLC. Our results show that the concentration of GABA was markedly reduced and that of serine slightly decreased in the Akp2-/- mice. Thus, TNAP deficiency would alter the functioning of PLP-dependent enzymes such as glutamate decarboxylases and racemases, and thus GABA and serine synthesis.
Second, we examined the role of TNAP in controlling synaptic transmission by performing electrophysiological recording in slices of normal mouse somatosensory cortex maintained in vitro. Blocking TNAP activity with levamisole modified short-term synaptic plasticity, significantly reducing

synaptic depression and/or increasing synaptic facilitation. These electrophysiological data are compatible with the putative role of TNAP in regulating the extracellular concentration of ATP and adenosine through its ectonucleotidase function.
Altogether, our results strengthen an essential role of TNAP in synaptic transmission in the mammalian brain.

P2.043

Evidence that neurons of the supramammillary nucleus activated during paradoxical sleep are glutamatergic

Billwiller, F. (Lyon)[1], Clement, O. (Lyon)[1], Charrié, P. (Lyon)[1], Luppi, P.H. (Lyon)[1]
[1]*Université Claude Bernard Lyon 1, Faculté de Médecine RTH Laennec, CNRS UMR 5292, Lyon, France*

Together with muscle atonia and rapid eye movements (REM), paradoxical sleep (PS) is characterized by cortical and hippocampal activation. The pathways responsible of this activation are still a matter of debate. It has been proposed that pontine PS-on neurons activate the intralaminar thalamic nuclei and the basal forebrain cholinergic neurons which in turn activate the cortex. The supramammillary nucleus (SuM) might also play a role in such activation, in particular that of the hippocampus. Indeed it projects to the medial septum and the dentate gyrus and plays a role in the genesis of hippocampal theta rhythm (Kirk and McNaughton, 1991). It has been shown that the SuM contains a large number of glutamatergic neurons. Further, the SuM projection to the dentate gyrus is issued from glutamatergic neurons (Boulland et al, 2009; Soussi et al, 2010). Besides, we recently demonstrated by means of Fos labeling that the SuM contains a large number of neurons specifically activated during PS hypersomnia (Sapin et al, 2010).
In view of these data, we hypothesized that the neurons of the SuM activated during PS are glutamatergic. To test this hypothesis, we used a combination of Fos immunostaining and *in situ* hybridization of vesicular glutamate transporter 2 (vGLUT2) mRNA in three groups of rats: control (n=4), deprived of PS for 72h (n=4) and allowed to recover during 150 min after such deprivation (n=4). In rats subjected to PS recovery, we found that the majority of the Fos-labeled neurons located in the SuM also express the vGLUT2 mRNA. In contrast, no or only a few double-labeled neurons were observed in control and PS deprived animals. These results indicate that the majority of neurons activated during PS in the SuM are glutamatergic. It might therefore be hypothesized that hippocampal activation and theta ryhtms occurring during PS might be induced by a glutamatergic projection from PS-on neurons localized in the SuM.

P2.044

Interleukin-6-induced S100B secretion is inhibited by haloperidol and risperidone

Lunardi, P. (Porto Alegre)[1,2], de Souza, D.F. (Porto Alegre)[1], Wartchow, K. (Porto Alegre)[1], Hansen, F. (Porto Alegre)[1], Guerra, M.C. (Porto Alegre)[1], Nardin, P. (Porto Alegre)[1], Gonçalves, C.A. (Porto Alegre)[3]
[1]*Universidade Federal do Rio Grande do Sul, Biochemistry, Porto Alegre, Brazil,* [2]*Université Paris Descartes, Neuron-glia Interactions, Paris, Brazil,* [3]*Universidade Federal do Rio Grande do Sul, Biochemistry /ICBS, Porto Alegre, Brazil*

Background: Although inflammation may be a physiological defense process, imbalanced neuroinflammation has been associated with the pathophysiology of brain disorders, including major depression and schizophrenia. Activated glia releases a variety of pro-inflammatory cytokines that

contribute to neuronal dysfunction. Elevated levels of S100B, a glia derived protein, have been observed in the serum and CSF of schizophrenic patients suggesting a glial role in the disease. We herein evaluate whether S100B secretion could be directly modulated by the main inflammatory cytokines associated with the pathophysiology of schizophrenia - IL-1β, TNF-α, IL-6 and IL-8 - in C6 glioma cells and acute hippocampal slices, as well as the possible involvement of MAPK pathways in these responses. We also investigate the effects of these cytokines on GFAP and oxidative stress. Finally, we explore the effects of typical and atypical antipsychotic drugs on glial cytokine-induced S100B release.
Discussion: Our results suggest that S100B secretion is increased by pro-inflammatory cytokines via MAPK and that oxidative stress may be a component of this modulation. These results reinforce the idea that the S100B protein is involved in the inflammatory response observed in many brain diseases, including schizophrenia. Moreover the antipsychotics, haloperidol and risperidone, were able to inhibit the secretion of S100B following IL-6 stimulation in C6 glioma cells.

P2.045

Effects of hypergravity on hippocampal transcriptome compared to those observed in mice under corticosterone injection-mimicked stress

Morel, J.-L. (Bordeaux)[1], Porte, Y. (Bordeaux)[1]
[1]*Institut des Maladies Neurodegeneratives, UMR 5293, Bordeaux, France*

Centrifugation is a widely used procedure to study altered gravity on Earth, allowing the understanding of how a physical constraint sustained during the whole life is able to condition the mammalian physiology. Previously, a consortium of laboratories funded by the French spatial agency (CNES) and ANR showed that exposition to 3G level of hypergravity during 21 days induces global modification of the general physiology (bone, muscle, immune, vestibular and cerebrovascular functions) in C57Bl6J mice. In these experiments, 3G exposure increased plasma concentration of corticosterone, a stress hormone known for modulating memory, including in pathological cases. The measurement of memory capacities by standard memory is yet not possible in the centrifuge. However, literature largely described a large panel of proteins (channels, second messengers, transcription factors, structural proteins) whose expression is modified during memory processing. On the contrary, it is well known that deficiencies in the expression of these same agents can alter memory, including in pathologies or normal cerebral aging. Then, thanks to Illumina technology, we compared the whole hippocampal transcriptome of three groups of C57Bl6/J mice to gain insights into the effects of hypergravity on cerebral functions. Namely, a group of 3 weeks 3G-centrifuged mice has been compared to (1) a group subjected to an acute injection of corticosterone at a dosis comparable to the plasma levels previously measured and (2) a group receiving a transdermal chronic infusion of corticosterone during 21 days. Our results showed that hypergravity induced modification of expression of 76 genes. Among these, 31 were selectively regulated by hypergravity (26 up- and 5 down-regulated). 39 were up-regulated by hypergravity and acute corticosterone injection. Only 1 gene was upregulated by hypergravity and chronic transdermic infusion of corticosterone. Finally, 5 genes were regulated by hypergravity and both corticosterone treatments). In conclusion, if the hypergravity and corticosterone-induced stress have common pathways to modify gene expression in hippocampus, the hypergravity could also affect expression of hippocampal genes per se.

P2.046

Dynamics of the muscarinic m2 receptor (m2R) in living hippocampal neurons

Lambert, L. (Paris)[1], Fafouri, A. (Paris)[2], Herzog, E. (Paris)[1], Csaba, Z. (Paris)[2], Dournaud, P. (Paris)[2], El Mestikawy, S. (Paris)[1], Bernard, V. (Paris)[1]

[1]*Université Pierre et Marie Curie, INSERM UMRS 952 - CNRS UMR 7224, Paris, France,* [2]*Université Denis Diderot, INSERM, UMR U676, Paris, France*

The m2 muscarinic acetylcholine receptor (m2R), a G protein-coupled receptors, is expressed at the neuronal membrane where it regulates neuronal functions. The proper trafficking of m2R to and from the neuronal membrane is essential for normal physiological responses. We have previously shown that the stimulation of m2R induces its endocytosis in neurons in vivo. However, the dynamic properties and the molecular mechanisms that govern endocytosis are still badly known. We especially addressed the question of the endocytotic pathway used by m2R at early stages after stimulation. When stimulated, a GPCR may use different endocytosis pathways, including clathrin- or a caveolin-dependant pathways. In neurons, no data concerning the type (s) of pathway (s) that is (are) used by m2R after stimulation is available. For that we have developed original tools to study m2R trafficking in living hippocampal neurons in primary cultures, including plasmids expressing m2R tagged with fluorochromes (GFP, Super Ecliptic Phluorin) at is N terminus, co-expression of m2R and subcellular compartment markers and live neuron imaging using a confocal spinning disk microscope and a video-microscope. All our constructions were validated ; e.g. we have especially checked that m2R internalization by carbachol was not affected by the presence of the fluorochrome. We have co-expressed m2R-GFP with clathrin or caveolin 1 tagged with red fluorescent protein and living neurons where observed under microscope and imaged. We have shown for the first time that, very soon after the beginning of endocytosis (6mn), m2R co-localizes partly with clathrin. The possibility for the m2R to internalize even after blocking of the clathrin-dependant pathway with drugs or molecular tools (phenylarside oxide and shRNA for clathrin heavy chain) suggests that there may exist another pathway of endocytosis. We are checking if the m2R may use the caveolin-dependant pathway to enter into the neuron. Alternatively, we cannot also exclude that another clathrin- and caveolin-independant pathway exist. Works are in progress to identify the post-endocytotic fate of m2R after stimulation, using markers of the different subcellular organelles.

P2.047

Ctr9, a protein of the transcription complex Paf1, regulates the dopamine transporter activity at the plasma membrane

Erdozain, A.M. (Paris)[1], De Gois, S. (Paris)[1], Slama, P. (Paris)[1], Pietrancosta, N. (Paris)[2], Louis, F. (Paris)[1], Bouvrais-Veret, C. (Paris)[1], Daviet, L. (Paris)[3], Giros, B. (Paris)[1]
[1]*Laboratoire de Physiopathologie des Maladies du Système Nerveux Central, INSERM U.952 - CNRS UMR 7224 - Universite Paris VI, Paris, France,* [2]*Laboratoire de Chimie et Biochimie Pharmacologiques et Toxicologiques, CNRS-UMR8601-Universite Paris V, Paris, France,* [3]*Hybrigenics, Paris, France*

Dopamine (DA) is a major regulator of sensorimotor and cognitive functions. The dopamine transporter (DAT) is the key protein that determines spatial and temporal activity of released dopamine in the synaptic cleft, by rapid reuptake of the neurotransmitter into presynaptic terminals. The importance of this reuptake process is substantiated by the profound consequences of its blockade using psychostimulant drugs (i.e. amphetamine, cocaine) or by genetic invalidation of the DAT gene (DAT-KO mice). To date, several evidences have suggested that transporter-interacting proteins may play a role in DAT function and regulation.
In order to discover new DAT-associated proteins, we used the yeast two-hybrid system to screen a rat brain library using the entire intracellular carboxyl terminal domain of DAT. We identified Ctr9, a tetratricopeptide protein, as a novel DAT-interacting partner and we confirmed this interaction by GST-pulldown assay. We then demonstrated that co-expression of Ctr9 with DAT in mammalian cells results in colocalization of the two proteins and in a dramatic increase of DAT-mediated dopamine uptake. Binding of Ctr9 to the transporter requires the first half of DAT carboxyterminus and more precisely the residues YKF. Mutation this amino acids not only leads to a loss of interaction with Ctr9, but also impairs DAT activity by retention of the transporter in the cytoplasm. Characterization of the role of Ctr9 gave us new insights on this protein. First we provided evidence that Ctr9 localization is

not restricted to to nucleus, as previously described. Second, by performing 3D-modelization of DAT-Ctr9 interaction, we uncovered three new tetratricopeptide domains in Ctr9 sequence.
Altogether, our datas suggest a role for Ctr9 in the regulation of the dopamine transporter activity.

P2.048

In vitro electrophysiological characterization and optogenetic manipulations of parvalbumin-expressing neurons in the globus pallidus

Abdi, A. (Bordeaux)[1], Miguelez, C. (Bordeaux)[1,2,3], Morin, S. (Bordeaux)[1], Bioulac, B. (Bordeaux)[1], Baufreton, J. (Bordeaux)[1]
[1]Université Bordeaux Segalen, Institut des Maladies Neurodégénératives, Bordeaux, France, [2]Faculty of Medicine and Dentistry, Department of Pharmacology, University of the Basque Country, Leioa, Spain, [3]Faculty of Pharmacy, Department of Pharmacology, University of the Basque Country, Vitoria-Gasteiz, Spain

The globus pallidus (GP) constitutes the central node in BG circuitery as it controls the pattern of activity of all other nuclei of the BG through its widespread projections. The GP is exclusively composed of GABAergic neurons which have been classified into different groups according to morphological, neurochemical, and electrophysiology criteria. Recent electrophysiological studies have challenged this classification in subtypes by reporting that GPe neuron properties are spread over a continuous space, making differentiation into subgroups impossible. Our objective was thus to clarify the existence of several neuronal subtype in GP using the correlation between electrophysiological properties with immunohistochemical profiling.
First, we performed immunohistochemistry for parvalbumin (PV), preproenkephalin (PPE) and the pan-neuronal marker NeuN. These experiments revealed, as previously shown by others, that PV and PPE do not co-localize in GP neurons and that PV-expressing and PPE-expressing GP neurons represents ~ 50% and ~ 35% of the total number of GP neurons respectively. A third population corresponding to 15% of GP neurons was only positive for NeuN. We then performed patch-clamp recordings in transgenic mice (PV-Ai9) in which the population of PV-expressing neurons is labelled by the fluorescent protein double-tomato (dt). Electrophysiological parameters such as spontaneous firing rate, driven activity, rebound properties were collected in dt+ and dt- neurons. We found that the spontaneous firing rate of dt+ neurons was significantly higher than dt- neurons. Rebound-burst parameters and driven activity were also different between the two types of neurons. We also found that the short-term plasticity profiles of striatally-evoked IPSCs were different between dt+ and dt- neurons. Finally we performed optogenetic manipulations of PV-expressing GP neurons using viral expression of ChR2 or NpHR in PV-Ai9 mice. Our results suggest that PV-expressing GP neurons have specific intrinsic and synaptic properties compared with the other neuronal populations of this nucleus supporting the conclusion of the existence of functionally-distinct neuronal subtypes in this nucleus.

P2.049

Role of microRNA miR124 in bone cancer pain mechanisms

Elramah, S. (Bordeaux)[1,2], Bastide, M. (Bordeaux)[1,2], Leste-Lasserre, T. (Bordeaux)[3], Brochard, A. (Bordeaux)[3], Nagy, F. (Bordeaux)[2], Landry, M. (Bordeaux)[2], Favereaux, A. (Bordeaux)[2]
[1]Université Bordeaux Segalen, Interdisciplinary Institut for Neurosciences, Bordeaux, France, [2]CNRS, UMR 5297, Interdisciplinary Institute for Neuroscience, Bordeaux, France, [3]INSERM U862, Institute François Magendie, Physiopathologie de l'addiction, Bordeaux, France

Bone cancer pain is the most disruptive event among cancer patients, with a devastating effect on their quality of life. MicroRNAs (miRNAs) are non-coding small RNA molecules, that negatively regulate gene expression by binding to the 3'UnTranslated Region (3'UTR) of target mRNAs leading to translational inhibition or degradation. Because long-lasting changes in cancer pain sensitivity are accompanied by altered gene regulation, miRNAs expressed in nociceptive pathways may influence the development and maintenance of pain. In the present study, we aimed
(i) at characterizing the mouse model of cancer pain, and
(ii) at studying the modulation of miRNA and gene expression involved in bone cancer pain mechanisms.
We used a murine model of bone cancer, which shares similarities with human cancer-induced bone pain. Assessment of the nociceptive state using Dynamic Weight Bearing (DWB) and Plantar tests, showed a decrease in operated paw weight bearing and withdrawal latency at day 14 that persisted until mouse sacrifice (day 21). Furthermore, we characterized the extent of cancer-induced bone destruction by 3D X-rays and gross anatomy. Results from mRNA microarrays and miRNA qPCR panel revealed changes in expression patterns of miRNAs and target mRNAs in mouse spinal cord of bone cancer model. This was further confirmed using quantitative RT-PCR, and candidate miRNA/genes have been identified. Here, we focused on the central nervous system-specific miRNA: miR124, and its targeted genes that showed significant regulation in pain condition. The affinity of miR124 and its candidate targets were further confirmed using luciferase assay.
The changes of miRNAs expression in the spinal cord have never been studied in bone cancer pain context. Here, we show that miRNAs are likely to play a major role in cancer pain, and might represent a potential therapeutic target.

P2.050

Phosphorylation of mafA in mice is vital at birth

Lecoin, L. (Orsay)[1], Rocques, N. (Orsay)[1], Ruffault, P.-L. (Gif sur Yvette)[2], Larcher, M. (Orsay)[1], Eychène, A. (Orsay)[1], Fortin, G. (Gif sur Yvette)[2], Champagnat, J. (Gif sur Yvette)[2], Pouponnot, C. (Orsay)[1]
[1]Institut Curie, CNRS U3347 INSERM U1021, Orsay, France, [2]Neurobiologie et Développement,UPR 3294,C.N.R.S.,IFR 144 Neuro-Sud Paris, Institut de Neurobiologie Alfred Fessard, Gif sur Yvette, France

MafA is a bZIP Leucine zipper transcription factor, the expression of which is restricted to small sub-populations of neurons in the central nervous system (olfactory bulb, preoptic area of the hypothalamus, facial motor nucleus, trigeminal sensory nucleus of the brainstem, dorsal horn and interneurons of the spinal cord) and in the peripheric nervous system (touch sensory neurons). By analyzing respiration in maf-A knock-out mice, we show that the mutants display fewer neonatal apneas than control animals. This observation shows that mafA-expressing neurons control the breathing pattern during the first hours after birth.
We have previously shown that the transcriptional activity and degradation of MafA is controlled by GSK3 phosphorylation. We have generated a knock-in of mafA in which the phosphorylated residues have been mutated into alanin and can no longer be phosphorylated by GSK3. In these mice, mafA protein over-accumulates in maf-A expressing neurons. Homozygous mutants lack neonatal apneas as KO, but, in addition display a progressively disorganized rhythm and all of them die 8 to 14 hours after birth. These data suggest that mafA transcription factor activity interferes with the respiratory rhythm during the first day of life and that its post-translational regulation is of crucial importance to allow physiological maturation of the neonatal respiratory rhythm.

P2.051

Regulation of activity dependant drebrin phosphorylation on S659 by the tumor suppressor PTEN

Kreis, P. (London)[1], Hendricusdottir, R. (London)[2], van Diepen, M. (Horsham)[3], Mack, T. (Berlin)[4], Ryves, J. (Cardiff)[5], Harwood, A. (Cardiff)[5], Leslie, N. (Dundee)[6], Parsons, M. (London)[2], Eickholt, B. (Berlin)[4]

[1]King's College London, MRC Center for Developmental Neurobiology, London, United Kingdom, [2]King's College London, London, United Kingdom, [3]Novartis Pharmaceuticals UK Limited, Horsham, United Kingdom, [4]Charité - Universitätsmedizin Berlin, Berlin, Germany, [5]Cardiff School of Biosciences, Cardiff, United Kingdom, [6]College of Life Sciences, University of Dundee, Dundee, United Kingdom

PTEN (Phosphatase and tensin homolog deleted on chromosome 10) is a lipid phosphatase that mainly antagonises the PI3K signalling pathway by transforming the phosphatidyinositol 3 phosphate lipid (PIP3) into phosphatidyinositol 2 phosphate (PIP2). As a tumour suppressor, PTEN has been largely studied for its role in cancer, however little is known about its function and regulation in the central nervous system. We have recently identified novel proteins that form complexes with PTEN in brain tissue by mass spectrometry. One binding partner was identified as drebrin and characterisation of the interaction by co-immunoprecipitation and using FLIM (fluorescence lifetime imaging microscopy) demonstrated that drebrin interacts directly and preferentially with the C2 domain of PTEN in particular a sequence of 22 amino acids named the D-loop. Drebrin is an actin binding protein largely known for its function in the formation of dendritic spines and in the regulation of neuronal transmission. Reduced levels of drebrin have been observed in the hippocampus of patients with Alzheimer's disease. In search for potential effects of the PTEN-drebrin interaction we identified a phosphorylation site on drebrin at serine 659 that is regulated by PTEN. We developed a phosphospecific antibody against this site and showed that in basal conditions reduction of the interaction between PTEN and drebrin or suppression of the phosphatase activity of PTEN increases the level of P-S-659-drebrin. Following neuronal activity, using high potassium or high frequence stimulation, the phosphorylation of drebrin is increased which correlates with a decrease in the binding of drebrin to PTEN. Furthermore loss of PTEN not only increases the basal level of S-659-phosphorylation but also prevents any additional activity dependant phosphorylation. In summary, we identified a novel mechanism by which PTEN acts as a guard keeper of the amount of phosphorylation in basal conditions and responds to synaptic activity through dissociation from a drebrin complex, leading to a de-repression of phosphorylation of drebrin at S659.

P2.052

Imaging of the tripartite synapse using STED microscopy

Panatier, A. (Bordeaux)[1,2], Oliet, S. (Bordeaux)[1,2], Nägerl, V. (Bordeaux)[2,3]

[1]Neurocentre Magendie, U 862, Bordeaux, France, [2]Université de Bordeaux, Bordeaux, France, [3]IINS-CNRS UMR 5297, Bordeaux, France

Research on astrocytes in the brain has in recent years led to a thorough reappraisal of their role in brain function, extending greatly beyond the classic view as mere providers of structural support to neurons. Indeed, the concept of the tripartite synapse, composed of neuronal and astrocytic elements, recognizes the important role of these glial cells in the regulation of information transfer at synapses in the central nervous system, in a highly dynamic and multifaceted way.
Despite much progress, a major obstacle for investigating the crosstalk between astrocytes and neurons at synapses is their enormous morphological complexity and small size. This is particularly true for secondary and tertiary astrocytic processes, which typically are much too small to be properly resolved by conventional light microscopy.
To overcome this limitation and to better understand how astrocytes interact with spine synapses, we used stimulated emission depletion (STED) microscopy in organotypic hippocampal slices. We imaged neurons and astrocytes in two colors using a home-built STED microscope, which has a spatial

resolution of at least 70 nm. To visualize neuronal and astrocytic morphology in the same tissue we crossed transgenic mouse lines where neurons (Thy1-YFPH) or astrocytes (GFAP-GFP) are labeled. In addition, we used patch pipettes to fill astrocytes with Alexa Fluor 488 dye and to record from them electrophysiologically.
Our experiments demonstrate that it is possible to visualize the complex and hyperfine morphology of astrocytes by STED microscopy, faithfully resolving astrocytic processes that are much too small for conventional light microscopy. Importantly, the two-color STED approach allows us to reveal and investigate the intimate morphological relationship between spines and astrocytic processes embedded in brain slices under near-physiological conditions.

P2.053

The p21-activated kinase PAK3 forms heterodimers with PAK1 in brain implementing trans-regulation of PAK3 activity

Barnier, J.-V. (Orsay)[1], Combeau, G. (Orsay)[1], Kreis, P. (Orsay)[1], Domenichini, F. (Orsay)[1], Amar, M. (Orsay)[1], Fossier, P. (Orsay)[1], Rousseau, V. (Orsay)[1]
[1]*Université Paris-Sud CNRS, Centre de Neurosciences Paris-Sud, UMR 8195 CNRS, Orsay, France*

p21-activated kinase 1 (PAK1) and PAK3 control cell movement and division. In the brain, they are well known to regulate dendritic spine formation and maturation, and to play a role in synaptic transmission and synaptic plasticity (Kreis & Barnier, 2009). PAK3, in particular, is linked to X-linked intellectual disability (ID) with the identification of five associated mutations. The *pak3* gene is expressed in neurons both as a GTPase-regulated PAK3a protein and as three splice variants retaining one or two additional inserts (named b and c) and displaying unusual constitutive kinase activity. PAK1 regulation is based on its homodimerization, forming an inactive complex of two PAK1 molecules. Here, we studied the mechanism of *pak3* regulation by analyzing the ability of PAK3 to dimerize.
We firstly analysed the regulation of the PAK1/PAK3 complex in co-transfected cells by co-immunoprecipitation assays and by two-hybrid analysis using different tagged constructs. We demonstrate that although PAK3a is able to homodimerize, it is more likely to form heterodimeric complexes with PAK1, through a strong N-ter/C-ter interaction. The b and c inserts present in the regulatory domain of PAK3 splice variants decrease the dimerization but the capacity of PAK3 to form heterodimers with PAK1 is retained.
We further showed that PAK1 and PAK3 are co-expressed in neurons by RT-PCR on single cells, are colocalized within dendritic spines by immunofluorescence, co-purify with post-synaptic densities in fractionation experiments, and co-immunoprecipitate in brain lysates. Using kinase assays, we demonstrate that PAK1 inhibits the activity of PAK3a but not of the splice variant PAK3b in a trans-regulatory manner suggesting that the PAK3 splice variants have additional regulatory mechanisms. .
Interestingly, we show that an ID mutation that abrogates the kinase activity of PAK3, impairs dimerization with PAK1 whereas the R67C mutation that decreases GTPase binding does not. Altogether, these results show that PAK3 and PAK1 signaling may be coordinated by heterodimerization. We also discuss the hypothesis that heterodimerization linking PAK1 and PAK3 pathways may in part explain why some ID mutations are more severe than others (Combeau et al., 2012, J. Biol.Chem., 287,30084-96).

P2.054

Evidence for a role of the PAK3 kinase in adult neurogenesis in a mouse model of intellectual disability

Domenichini, F. (Orsay)[1], Carraro, M. (Orsay)[1], Veyrac, A. (Orsay)[1], Poirier, R. (Orsay)[1], Agasse, F. (Orsay)[2], Laroche, S. (Orsay)[1], Coronas, V. (Poitiers)[3], Barnier, J.-V. (Orsay)[1]
[1]*Université Paris-Sud XI, Centre de Neurosciences Paris-Sud, UMR 8195 CNRS, Orsay, France,* [2]*Institut Curie, CNRS UMR 3306, Orsay, France,* [3]*Université de Poitiers, Institut de Physiologie et Biologie Cellulaires, Poitiers, France*

Mutations in the pak3 gene, coding for the p21-activated kinase 3 (PAK3), are responsible for non-syndromic intellectual disability in human (Allen et al., 1998). Kinases of the PAK family are important effectors of Rac1 and Cdc42 GTPases, involved in signalling pathways that control actin cytoskeleton and thus, in the regulation of dendritic spine dynamics and synaptic plasticity (Kreis & Barnier, 2009). Moreover, it has been shown that PAK kinases also participate in the regulation of cell proliferation. In the developing Xenopus embryo, PAK3 is implicated in the control of cell cycle withdrawal and in neuronal cell commitment (Souopgui et al., 2002). Many studies have shown that newborn neurons in the adult brain significantly contribute to synaptic plasticity and memory processes (Ming & Song, 2011). Moreover, cognitive deficits in certain disease, including models of syndromic intellectual disability (Lazarov et al., 2011), are associated with impaired adult neurogenesis.
We hypothesized that PAK3 could play a role in the regulation of adult neurogenesis and therefore that *pak3* mutation-related neurogenic defects could underlie certain features of intellectual disability. We studied neurogenesis in KO *pak3* pups in the subventricular zone and dentate gyrus by *ex vivo* approaches (neurosphere assay, monolayer cultures of different cell types and qPCR). We observed a significant increase of neural precursor proliferation without alteration of cardinal properties of neural stem cells. *In vivo*, in transgenic mice, we found a decrease of the number of adult-born cells in the dentate gyrus.
Altogether, these results strongly suggest that altered adult neurogenesis due to *pak3* mutations may at least in part underlie some of the cognitive deficits associated with intellectual disability.

P2.055

Modulation of glutamatergic synaptic transmission by 5-HT_{2A} receptors in rat prefrontal cortex

Becamel, C. (Montpellier)[1], Barre, A. (Montpellier)[1], Bockaert, J. (Montpellier)[1], Marin, P. (Montpellier)[1]
[1]*Institut de Génomique Fonction - Univ. de Montpellier 1 & 2, CNRS UMR 5203 - INSERM U 661, Dépt. de Neurobiologie, Montpellier, France*

The serotoninergic system plays an important role in regulating prefrontal cortex functions such as emotional control, cognitive behaviors and working memory. Among the G protein-coupled receptors activated by serotonin (5-HT), 5-HT_{2A} receptors raise particular interest. Indeed, they are the target of a large number of psychoactive drugs including atypical antipsychotics (antagonists or inverse agonists) and the majority of psychedelic hallucinogens act as agonists or partial agonists at 5-HT_{2A} receptors.
5-HT_{2A} receptors that are especially abundant in layers V of the prefrontal cortex, with a predominant expression in apical dendrites of pyramidal neurons, have been involved in numerous psychiatric diseases including psychoses such as schizophrenia.
Several studies have shown that activation of 5-HT_{2A} receptors in the prefrontal cortex results in an increase in spontaneous glutamatergic synaptic activity. However, the mechanism of 5-HT_{2A} receptor-mediated modulation of synaptic transmission in prefrontal cortex is still matter of debate. The purpose of this study is to characterize the role of 5-HT_{2A} receptors in the glutamatergic synaptic transmission. Here, we showed that activation of 5-HT_{2A} receptors modulates synaptic transmission of thalamo-cortical inputs by eliciting a reversible potentiation of NMDA evoked response. This effect is abolished in presence of extracellular Mg2+, a NMDA blocker, suggesting an involvement of presynaptic NMDA receptors that could lead to an increase of glutamate release. We confirmed this hypothesis by performing NMDA paired-pulse facilitation and AMPA mediated miniature EPSCs recording. Moreover, delivering GDP-β-S (a G protein signaling pathway blocker), directly in the post-synaptic neuron did not affect the effect of 5-HT_{2A} activation, as well as PLC and PKC blockers, suggesting the involvement of presynaptic 5-HT_{2A} receptors.

P2.056

Vascular endothelial growt factor and glutamate: a new crosstalk underlying glioblastoma invasice capacities

Bardin, M. (Lyon)[1,2], Lay, S. (Lyon)[1,2], Chounlamountri, N. (Lyon)[1,2], Benetollo, C. (Lyon)[2,3], Honnorat, J. (Lyon)[1,2], Meissirel, C. (Lyon)[1,2]
[1]INSERM U 1028 - CNRS UMR 5292, Centre de Recherche en Neurosciences de Lyon, Neurooncology and Neuroinflammation, Lyon, France, [2]Université Claude Bernard Lyon 1, Lyon, France, [3]INSERM U 1028 - CNRS UMR 5292, Centre de Recherche en Neurosciences de Lyon, Functional Neurogenomics and Optogenetics, Lyon, France

Vascular endothelial growth factor (VEGF) inhibitors are now the focus of anticancer therapy because VEGF is a critical regulator of tumor angiogenesis and high levels of VEGF in tumors are of bad prognosis. However, brain tumors and in particular glioblastoma (GBM), have been shown to eventually escape anti-VEGF treatments raising the question of alternative therapeutic targets. Recent data have demonstrated that GBM release glutamate, and those with high glutamate release have a distinct growth advantage in the host brain. GBM-derived glutamate activates a subset of glutamate receptors on tumor cells that promotes their migratory behavior and malignant phenotype, thereby contributing to the invasive properties of GBM.
Recently, a promoting effect of VEGF on cell migration has been extended to developing neurons and was shown to involve a cross-talk between VEGF and glutamate receptors. Therefore, we hypothesized that VEGF receptors might interact with glutamate receptors and regulate their function in GBM cells. Our aim is to investigate how VEGF and glutamate receptors cross-talk to exacerbate GBM growth and invasion.
To explore this possibility we have been using well known cellular model of GBM and already revealed that they expressed the VEGF receptors VEGFR1 and VEGFR2 as well as the subunits of the calcium-permeable AMPA type of glutamate receptors. We also assessed the physiological role of the VEGF and glutamate receptor crosstalk in GBM cell migration, by performing migration assays and selectively blocking VEGFR2 and /or calcium-permeable AMPA receptors in human GBM cells. Our results indicate that blocking the VEGF/VEGFR2 or glutamate/AMPA autocrine signaling pathways alters the migratory ability of human GBM cells.
Altogether, our current data suggest an unexpected crosstalk between VEGF and glutamate receptors in tumor cells that could be responsible for GBM progression and invasion. In the future, this new crosstalk may provide rationale for combining anti-VEGF and specific anti-glutamate receptor targeted therapies.

P2.057

Mechanism of glioblastoma proliferation by CRMP5 controling Notch signaling pathway

Moutal, A. (Lyon)[1,2], Bertrand, C. (Lyon)[1], Malleval, C. (Lyon)[1], Watrin, C. (Lyon)[1,2], Chounlamountri, N. (Lyon)[1], Besançon, R. (Lyon)[1,2], Honnorat, J. (Lyon)[1,2,3], Meyronet, D. (Lyon)[1,2,3], Thomasset, N. (Lyon)[1]
[1]U 1028 UMR 5292 Univ Lyon1, Centre de Recherche en Neuroscience de Lyon, Neurooncology Neuroinflammation Team, Lyon, France, [2]Universite Claude Bernard Lyon 1, Lyon, France, [3]Hospices Civils de Lyon, Hôpital Neurologique, Lyon, France

CRMP5 belongs to the Collapsin Response Mediator Protein (CRMP) family highly expressed in the developing brain, and in adult brain areas that retain neurogenesis where CRMP5 is involved in the generation and survival of newly generated neurons. In pathology, we previously identified CRMP5 as a marker of aggressivity in neuroendocrine lung tumors (Meyronet et al, 2008) while Liang et al (2005) using transcriptomic analysis of 20 Glioblastoma, revealed CRMP5 in a cluster of genes related to proliferation correlated with a poor overall survival. We confirmed these results at the protein level in a study of a continuous series of 84 GBMs and correlated higher CRMP5 expression with a significantly lower median survival (5,9 months) than those with low or negative expression (15,5 months) (p=0,0026). Interestingly, GBM with higher CRMP5 expression were characterized by a higher mitotic index.

At a cellular level, CRMP5 knockdown (siRNA) in the GBM cell line GL15 resulted in a proliferation decrease by 60% ($p=1.07.10^{-10}$) analysed by EdU incorporation. At a mechanistic level, we revealed a loss of expression of the Notch pathway target gene hes1 using qRT-PCR and Western Blotting. A direct effect of CRMP5 on Notch transcriptional activation was shown using reporter gene (luciferase) activation with a decrease in hes1 promoter activation by 50%.
To further assess CRMP5 action on Notch signaling pathway, we investigated Notch receptors activation at the ligand level, endocytosis level and degradation level. No effect was detected at ligand and endocytosis level. We demonstrated a loss of Notch1 and Notch2 receptor, under CRMP5 blockade, using Western blot analysis while corresponding mRNA where still detected by qRT-PCR. Using proteasome and lysosome inhibitors we showed Notch1 and Notch2 receptors lysosomal degradation under CRMP5 silencing.
Taken together, those results show that CRMP5 expression in GBM positively modulates Notch signaling pathway in relation with GBM proliferation. Further characterization of CRMP5 dependant Notch activation in glioblastoma will have tremendous implications for patient prognosis and elaboration of very specific new antitumor therapies.

P2.058

Polarity protein Crb3 controls Schwann cell elongation via the Hippo signaling pathway

Cotter, L. (Montpellier)[1], Fernando, R. (Montpellier)[1], Perrin-Tricaud, C. (Montpellier)[1], Berthelot, J. (Montpellier)[1], Bartolami, S. (Montpellier)[1], Tricaud, N. (Montpellier)[1]
[1]*Institut des neurosciences de Montpellier, Inserm U 1051, Montpellier, France*

Fast nerve conduction relies on the electrical isolation of axons via successive myelin segments. The myelin geometry _diameter and internodal length_ is therefore a critical parameter of the nerve conduction velocity. However the basic mechanisms that contribute to the optimum longitudinal and radial extension are only partially understood.
The myelinating Schwann cell (mSC) is an extremely polarized cell and polarity proteins have recently emerged as key players in the basic cellular mechanism of myelination. Here we show that Crb3 (mammalian homolog of drosophilia Crumbs), a small transmembrane polarity protein, which is located exclusively to the mSC microvilli participates to a mechanism that negatively regulate mSC length. Crb3 inhibits myelination by activating the Hippo pathway. This pathway transduces signals from the plasma membrane to the nucleus where it regulates the function of transcription factor YAP. YAP is required for SC myelination and our data suggest that axonal stretching during body growth stimulates YAP activity to elongate the myelin sheath.
In addition we show that Lama2 mice that mimic a human peripheral neuropathy with reduced internodal lengths have a decreased YAP nuclear activity. Restoring in these mice a higher amount of nuclear YAP by knocking down Crb3 partially rescues the internodal length. This suggests that defects in YAP activation or Crb3-hippo pathway may be the cause of some peripheral neuropathies**.**

P2.059

Organotypic hippocampal slice cultures and microglia

Lalisse, S. (Montpellier)[1], De Bock, F. (Montpellier)[1], Rassendren, F. (Montpellier)[1], Ulmann, L. (Montpellier)[1]
[1]*Institut de Génomique Fonctionnelle, Montpellier, France*

Microglia plays a critical role in determining the spatial and temporal extent of inflammation in different brain pathologies including epilepsy and neuropathic pain. Purinergic receptors, particularly ATP-

gated P2X channels regulate several facets of microglia activation notably through their implication in the release of pro-inflammatory mediators or trophic factors. Among the different P2X receptors, P2X4 seems to play a crucial role in activated microglia. Indeed, following peripheral nerve injury P2X4 expression is induced de novo in activated spinal microglia, where it controls the release of microglial BDNF and the subsequent increase of local network excitability. Similarly, following induction of a status epilepticus, P2X4 is up regulated in hippocampal microglia and P2X4-deficient mice display altered microglial activation and reduced CA1 pyramidal cell death after status epilepticus. In the hippocampus, P2X4 receptors are also expressed in pyramidal neurons where their activation coincides with episode of high electrical activities. The purpose of our work is to elucidate the respective roles of neuronal and microglial P2X4R in the hippocampus following induction of SE. Here, we use a mouse organotypic hippocampal slice culture (OHC) as an in vitro model of status epilepticus, which offers the possibility to manipulate microglia. Our strategy relies on the specific clodronate-mediated depletion of microglia in OHC from wild type mice and their replenishment with microglia from P2X4-deficient mice. To that aim we use the CX3CR1$^{eGFP/+}$ mice, in which microglia express the green fluorescent protein, and allows for an easy analyze of microglia. Here we present preliminary data showing that an efficient microglial depletion is obtained following a 7 days treatment of OHC with clodronate. We show that microglial depeletion has several side effects such as an apparent increase in cell death, activation of astrocytes, both likely due to the reduction of the phagocytic function of microglial and the accumulation of dead cells. We also provide evidence that replenishment of clodronate-depleted microglial OHC can be efficiently obtained.

P2.060

Neuronal nitric oxide synthase expression and activity in supraoptic and paraventricular nuclei of normal and Dp71 deficit mice

Benabdesselam, R. (Algiers)[1], Rendon, A. (Paris)[2], Hardin-Pouzet, H. (Paris)[3], Dorbani-Mamine, L. (Algiers)[1]
[1]FSB/USTHB, Equipe de Neurochimie/LBPO, Algiers, Algeria, [2]Institut de la Vision, INSERM UMR S 592 Laboratoire de Physiopathologie Cellulaire et Moléculaire de la Rétine, Paris, France, [3]UPMC, INSERM UMRS 952, CNRS UMR 7224, Labo de Physiopathologie des Maladies du SNC, Paris, France

Next to their osmoreceptive function, the supraoptic and paraventricular nuclei (SON/PVN) exert their major role in osmoregulation as the effector pole by adapting the synthesis and the release of vasopressin neuropeptide to the variation of the plasmatic osmolality. This needs a fine tuning of the magnocellular neuron (MCN) activity and among the different modulatory pathways, nitric oxide (NO) is of particular importance. Previous study of our group demonstrated that mice knock-out for Dp71 exhibit a perturbated osmoregulatory axis, characterized by a lower plasma osmolality under normal conditions. To go further in the understanding of the mechanisms underlying this result, in the SON and PVN, we analysed nNOS, and NADPH diaphorase, the histochemical marker of NOS activity, because these proteins represent an index of the osmoregulatory axis activation. Their expression and activity increase in response to osmotic stimulation, dehydration or hypovolemia. Indeed, NO is an important modulator of vasopressin synthesis. In normal mice, we found nNOS in magnocellular neurons in the dorsal part of the SON and in the lateral part of the PVN. In the SON and PVN of Dp71-null mice, nNOS expression was upregulated and in the same way, the NADPH-diaphorase staining was increased. Since NADPH-diaphorase activity correlates with the distribution of NOS expression and activity, it could be deduced that the enhancement of nNOS expression was accompanied with a rise of its activity and presumably with an increase of the NO production. Such a modification in the NO pathway activity could also, at least partially, explain the modified osmolality of the Dp71-null mice.
Keywords: Dp71, neuronal nitric oxide synthase, NADPH-diaphorase, supraoptic nucleus, paraventricular nucleus.

P2.061

Afterhyperpolarization in mitral cells: contribution of synaptic and non-synaptic components

Mael, D. (Villeurbanne)[1], Fourcaud-Trocmé, N. (Lyon)[2], Palouzier-Paulignan, B. (Lyon)[2], Garcia, S. (Lyon)[2], Kuczewski, N. (Lyon)[2]
[1]*Université Claude Bernard Lyon 1, Rhône, Villeurbanne, France,* [2]*CRNL - UCBL - CNRS - UMR 5292 - INSERM U 1028, Lyon, France*

The hyperpolarization that follows the action potential generation, classically known as afterhyperpolarization (AHP), participates in the regulation of the neurons firing pattern. The AHP is generally exclusively due to the activation of potassic currents; however in the Mitral Cells (MC) of the olfactory bulb (OB) two synaptic, glutamatergic and GABAergic, components do also participate in this phenomena. Indeed, following the action potentials generation, MC dendrites are capable to release the neurotransmitter glutamate that in turn is responsible for a direct MC auto-excitation and of a disynaptic dendritic feed-back inhibition through the activation of the inhibitory neurons. To date, the three components of the MC AHP (potassic, glutamatergic and GABAergic) have been studied separately. In the present study we attempt to determine the relative contribution of the three components to the afterhyperpolarization and their impact on the action potentials generation. To this aim MC were recorded from OB slices and the AHP produced by MC firing at different frequencies in the absence and in the presence of inhibitors of the synaptic transmission.

P2.062

Methodological contribution to the proteomic analysis of physiologic and IL1RAPL1 depleted mouse excitatory synaptosomes

Luquet, E.M. (Bordeaux)[1], Biesemann, C. (Göttingen)[2], Humeau, Y. (Bordeaux)[1], Herzog, E. (Bordeaux)[1]
[1]*Institut Interdisciplinaire de Neurosciences, CNRS UMR 5297, Bordeaux, France,* [2]*Georg August University Göttingen, Faculty of Biology, Göttingen, Germany*

Il1rapl1 absence is known to induce cognitive disorders. Il1rapl1 is thought to be a post-synaptic protein engaged in heterophilic transynaptic adhesion with the presynaptic protein PTP∂ and in intracellular interaction with the post synaptic density protein PSD95. The goal of our project is to identify the molecular impact of *il1rapl1* deletion on the excitatory synaptosomes. The up and down-regulated VGLUT1 synaptic proteins in il1rapl1 KO mice will be identified by the expression levels comparison of native VGLUT1 synapse proteins from the lateral amygdala with the ones from il1rapl1 KO mice. The specificity and efficacy of our proteomic approach on VGLUT1 positive synaptosomes will greatly depend on the purity of our samples. Our VGLUT1^{venus} knock-in mice will allow the purification of VGLUT1^{venus} synaptosomes through fluorescence activated synaptosome sorting. Both targeted screening of synaptic markers through semi-quantitative immunoblotting and a mass spectrometry based broad screening will be performed. In the present communication we will present an update on the methodologies implemented through out the project. Through the identification of proteins affected by *il1rapl1* deletion we expect to contribute significantly to the understanding of mechanisms leading to cognitive disorders.

P2.063

Decreased vitamin B12 availability induces ER stress through impaired SIRT1-deacetylation of HSF1

Ghemrawi, R. (Vandoeuvre-les-Nancy)[1], Pooya, S. (Vandoeuvre-les-Nancy)[1], Lorentz, S. (Vandoeuvre-les-Nancy)[1], Gauchotte, G. (Vandoeuvre-les-Nancy)[1], Arnold, C. (Vandoeuvre-les-Nancy)[1], Gueant, J. (Vandoeuvre-les-Nancy)[1], Battaglia-Hsu, S. (Vandoeuvre-les-Nancy)[1]
[1]*Fac. de Médecine - Nancy Université, INSERM U 954, NGERE, Vandoeuvre-les-Nancy, France*

Vitamin B12 (cobalamin) is a key determinant of S-adenosyl methionine-dependent epigenomic cellular regulations related to methylation/acetylation and its deficiency produces neurodegenerative disorders by elusive mechanisms. Sirtuin 1 deacetylase (SIRT1) triggers cell response to nutritional stress through endoplasmic reticulum stress. Recently, we have established a N1E115 dopaminergic cell model by stable expression of a transcobalamin-oleosin chimer (TO), which impairs cellular availability of vitamin B12, decreases methionine synthase activity and S-adenosyl methionine level and reduces cell proliferation. In contrast, Oleosin-Transcobalamin chimer (OT) does not modify the phenotype of transfected cells. Presently, the impaired cellular availability of vitamin B12 in TO cells activated irreversible endoplasmic reticulum stress pathways, with increased p-eIF-2α, p-PERK, p-IRE1α, ATF6, decreased chaperon proteins and increased pro-apoptotic markers, CHOP and cleaved caspase 3, through reduced SIRT1 expression and subsequent greater acetylation of HSF1. Adding either B12 or SIRT1 and HSF1 activators and overexpressing SIRT1 and HSF1 dramatically reduced the activation of endoplasmic reticulum stress pathways in TO cells. Conversely, impairing SIRT1 and HSF1 by siRNA, expressing a dominant negative form of HSF1 or adding a SIRT1 inhibitor led to B12-dependent ER stress in OT cells. Addition of B12 abolished activation of stress transducers and apoptosis and increased expression of protein chaperons, in OT cells subjected to Thapsigargin, a strong ER stress stimulator. AdoX, an inhibitor of methyltransferase activities, produced similar effects than decreased B12 availability on SIRT1 and endoplasmic reticulum stress, by a mechanism related to increased expression of hypermethylated in cancer 1 (HIC1). Taken together, these data show that cellular vitamin B12 has a strong modulating influence on ER stress in N1E115 dopaminergic cells. The impaired cellular availability in vitamin B12 induces irreversible ER stress by greater acetylation of HSF1 through decreased SIRT1 expression, while adding vitamin B12 produces protective effects in cells subjected to ER stress stimulation.

P2.064

Imaging glutamatergic synaptic vesicles mobility in amygdala acute slices of *il1rapl1* deficient mice

Zhang, X. (Bordeaux)[1], Normand, E. (Bordeaux)[1], Lepleux, M. (Bordeaux)[1], Humeau, Y. (Bordeaux)[1], Herzog, E. (Bordeaux)[1]
[1]*Interdisciplinary Institute for Neuroscience, CNRS UMR 5297, Bordeaux, France*

IL1RAPL1 absence is known to induce cognitive disorders. IL1RAPL1 is thought to be a post-synaptic protein engaged in heterophilic transynaptic adhesion with the presynaptic protein PTP∂ and in intracellular interaction with the post-synaptic density protein PSD95. IL1RAPL1 depletion or overexpression is associated with morphological and functional alterations at glutamatergic synapses in rodents. In previous studies we used VGLUT1^{venus} knock-in mice to show that synaptic vesicles (SV) travel between synapses in the axon and are shared among boutons in a super pool of SVs. Here our aim is to establish the live imaging of VGLUT1^{venus} material in acute slices and to combine it with electorphysiology of the lateral nucleus of the amygdala complex (LA). Then, our goal is to study the effect of glutamatergic SV mobility on neurotransmission of LA neurons in physiological conditions. Concomitantly, we will study *il1rapl1* deletion in order to understand the effects of the deletion on VGLUT SV mobility.

P2.065

FosB and ΔFosB gene expression studies in cocaine acute and chronic self-administration model

Drutel, G. (Bordeaux)[1], Leste Lasserre, T. (Bordeaux)[1], Fiancette, J.-F. (Bordeaux)[1], Renault, P. (Bordeaux)[1], Revest, J.-M. (Bordeaux)[1], Balado, E. (Bordeaux)[1], Deroche-Gamonet, V. (Bordeaux)[1], Piazza, P.V. (Bordeaux)[1]
[1]Université Bordeaux Segalen, Neurocentre Magendie - INSERM U.862, Physiopathologie de l'addiction, Bordeaux, France

Addiction is characterized by compulsive drug seeking and loss of control over drug intake. The development of drug addiction has been shown to involve alteration in gene expression and particularly genes encoding the Fos family of transcription factors like FosB ans ΔFosB. The present study examined the gene expression of these transcription factors in different brain region after acute and chronic intravenous cocaine self-administration using an animal model that discriminates rats showing an addiction-like behavior from rats that maintain a controlled drug use. We found similar mRNA expression pattern for FosB and ΔFosB in striatal subregions (nucleus Accumbens (NAc) and Caudate Putamen (CPu)). These both transcripts expression are largely increased 40 minutes after the last session of 7 or 18 days of cocaine self-administration. We can show that induction of expression of both genes is largely higher in CPu than NAc. In contrast after 24h00 of withdrawal their expressions are totally downregulated more specifically in NAc. Moreover we found mRNA tolerance for FosB and ΔFosB in both striatal regions. Previous studies have found that ΔfosB is increased after repeated cocaine exposure. Our results suggest also potential implication of ΔFosB after acute cocaine use. Finally we could not identify for both genes any expression differences between addict and non-addict-like animals after 2 months of self-administration. At the level of transcription regulation, these genes seem clearly involved in drug effect but not in development of addiction-like behavior.

P2.066
Saccadic eye movements in depressed elderly patients

Carvalho, N. (Besançon)[1,2], Monnin, J. (Besançon)[1,2,3], Noiret, N. (Besançon)[1,4], Nicolier, M. (Besançon)[1,3], Tio, G. (Besançon)[1,3], Haffen, E. (Besançon)[1,2,3], Laurent, E. (Besançon)[4], Vandel, P. (Besançon)[1,2,3]
[1]Clinical Psychiatry, University Hospital, Besançon, France, [2]EA 481, Neurosciences, University of Franche-Comté, Besançon, France, [3]CIC-IT 808 Inserm, University Hospital, Besançon, France, [4]EA 3188, Laboratory of Psychology, University of Franche-Comté, Besançon, France

Background: Depression in elderly is often unknown, trivial, and frequently considered as a consequence of aging. Cognitive inhibition, a major component of executive functions, has been found to be impaired in depressed elderly through neuropsychological assessment. Here we propose to assess cognitive inhibition through an eye tracking study, no data being available on saccadic performances in the specific population of depressed elderly.
Method: Twenty inpatients (mean age = 70.4) meeting DSM-IV criteria for major depressive disorder were compared to forty seven healthy controls (mean age = 66.7). They performed neuropsychological and psychiatric assessments and two eye movement tasks: a prosaccades task to obtain basic parameters of eye movements and an antisaccade task to evaluate the inhibition capacities.
Results: In comparison with healthy subjects, depressed patients showed impaired performances in both oculomotor tasks. Concerning the prosaccade trial, depressed patients had higher reaction times and error rates than healthy controls. In the antisaccade task, reaction times and error rates were also higher in depressed patients than healthy subjects, however, the two populations showed similar correction rates.
Conclusion: Our study showed a marked psychomotor retardation and a deficit to inhibit response production in depressed elderly, in line with current literature in young depressed patients. This study offers new insight on the cognitive impairment of aged major depressive patients by two simple eye movement tasks.

P2.067

New tools for investigating response inhibition in adults with ADHD

Suarez Del Chiaro, I. (Marseille)[1], Burle, B. (Marseille)[1], Hasbroucq, T. (Marseille)[1], Lopera, F. (Medellin)[2], Casini, L. (Marseille)[1]
[1]*Université d'Aix Marseille - Laboratoire de Neurosciences Cognitives - CNRS, Marseille, France,* [2]*Neurosciences Groupe University of Antioquia, Medellin, Colombia*

Several theoretical models suggest that the core deficit in children with Attention Deficit Hyperactivity Disorder (ADHD) relies on response inhibition. However, research concerning the persistence of this deficit in adulthood is lacking.
This has been studied by comparing performances obtained by adults with ADHD and control subjects in a Simon Reaction Time (RT) task. The inhibition of inappropriate responses elicited by irrelevant information was evaluated through 1/ the analysis of RT and accuracy distribution (slope of the delta plots) and 2/ the analysis of electromyographic activity (EMG), which provided the possibility to identify partial errors (subthreshold EMG burst associated with incorrect response and preceding the correct one).
The classic analysis of mean RT indicated a larger Simon effect in adults with ADHD suggesting difficulties in inhibiting the automatic activation produced by the non relevant timulus information. However, in contrast, distribution analyses as well as analysis of partial errors revealed that the ability to suppress the automatic response remains intact in these patients.

P2.068

Changes in brain metabolic profiles in genetic absence-epilepsy rats from Strasbourg (GAERS) as investigated by ^{1}H HRMAS NMR spectroscopy

Deransart, C. (La Tronche)[1], Chabrol, T. (La Tronche)[1], Maunoir-Regimbal, S. (La Tronche)[2], Cavarec, F. (La Tronche)[1], Depaulis, A. (La Tronche)[1], Fauvelle, F. (La Tronche)[2]
[1]*Grenoble Institut des Neurosciences, INSERM U. 836 - Equipe 9,Bât. Edmond J. Safra, La Tronche, France,* [2]*IRBA-CRSSA, La Tronche, France*

Purpose: Genetic Absence Epilepsy Rats from Strasbourg (GAERS) are an inbred strain of Wistar rats which display spontaneous and recurrent spike-and-wave discharges on their electroencephalogram at an onset age of 25 days postnatal. Previous data[1] suggest that these rats had a different metabolism when compared to non-epileptic control rats (NEC). In order to further explore this hypothesis, we have compared metabolic profiles between GAERS, NEC and Wistars in ventrobasal thalamus (VB), primary somatosensory (SSI) and motor cortex (M1), striatum (St), hippocampus (H) and cerebellum (Cb), by using High Resolution Magic Angle Spinning (HRMAS) NMR.
Methods: Animals were quickly killed by decapitation, structures removed on ice and rapidly frozen in liquid nitrogen.
^{1}H HRMAS NMR spectra of small intact biopsies (15 mg) were acquired at 400 MHz using a CPMG pulse sequence and 4 Kz spinning rate. Relative concentrations of 18 metabolites were obtained using jMRUI software: acetate, alanine, aspartate, total creatine, choline, γ-amino-butyric acid, glutamate, glutamine, glutathione, glycerophosphocholine, glycine, lactate, myo-inositol, *N*-acetylaspartate, phosphoethanolamine, phosphocholine, scyllo-inositol, taurine.
Results: Compared to NEC, GAERS had mainly lower level of phosphoethanolamine in H, M1, SSI and St, and higher level of glutamine in H, M1, St and VB. Interestingly, scyllo-inositol was significantly decreased in all structures in GAERS compared to NEC and Wistars.

Discussion: Experiments are running on 14 day-old rats in order to examine whether these changes have already appeared before the onset of the first spike-and-waves discharges. Whether decreased levels of scyllo-inositol and phosphoethanolamine may contribute to the epileptic phenotype of GAERS will be further investigated. The role of scyllo-inositol is not well known; either antiepileptic[2] or marker of bad prognostic in chronic alcoholism[3] or Alzheimer desease[4] have been reported. Scyllo-inositol could be protective at low concentrations.

1- Melo T.M. et al., J Cereb Blood Flow Metab 26:1496-506, 2006;
2- Nozadze M. et al. Seizure, 20:173-6, 2011;
3- Viola A. et al., MAGMA, 17:47-61, 2004;
4- Griffith H.R. et al., NMR Biomed. 20:709-716, 2007.

P2.069

The MTLE mouse as a model of human temporal lobe epilepsy: a comparative study of most commonly-used antiepileptic drugs

Duveau, V. (La Tronche)[1], Bouyssières, C. (La Tronche)[1], Dumont, C. (La Tronche)[1], Depaulis, A. (Grenoble)[2], Roucard, C. (La Tronche)[1]
[1]SynapCell, La Tronche, France, [2]Grenoble Institut des Neurosciences U 836, Grenoble, France

Epilepsy is one of the most common chronic neurological disorders. Among the different form of epilepsies, mesio temporal lobe epilepsy is one of the most prominent and is as well the most common form of focal drug resistant epilepsy.
There are nowadays a large number of antiepileptic drugs (AEDs) available on the market; however they are still ineffective in drug-resistant patients. There is therefore an urgent need to develop new therapeutic strategies in order to treat those patients.
The main morphological and electrophysiological features of MTLE patient have been resumed in a mouse model. In fact, a unilateral injection of kainic acid in the dorsal hippocampus provokes a cell loss as well as morphological reorganization observed in MTLE patient. Similarly to what is observed in patient, the establishment of the model follows three steps; first, an initial event (kainic acid injection), followed by a latent phase of 2 to 3 weeks and eventually the occurrence of spontaneous recurrent seizures. Thus, this model appears to be a relevant model for studying new therapeutic in epilepsy. However, little is known about the effect of the most commonly used AEDs on this model.
In this work, we investigated the effect of the most commonly used AEDs on the occurrence of focal spontaneous hippocampal paroxysmal discharges (HPD) in the MTLE mouse model.
We studied the dose response effect of acute injection of commonly used AEDs on the occurrence and duration of HPD by deep EEG recording.
We found that vigabatrin, levetiracetam, diazepam and pregabalin could suppress the number and the cumulated duration of HPD in a dose-dependent manner. Moreover, only high doses of valproate, lamotrigine and carbamazepine were effective in this model, but with strong side-effects.
We demonstrated that the MTLE mouse model is sensitive to most of the AEDs on the market but at different threshold of concentration.
In this work, we established for the first time, the pharmacological identity card of the MTLE mouse model.

P2.070

Parkinson's disease related motivational deficits revealed by a nigrostriatal dopaminergic lesion in a rat model, are reversed by pramipexole

Duran, T. (Grenoble)[1], Carcenac, C. (Grenoble)[1], Drui, G. (Grenoble)[1], Savasta, M. (Grenoble)[1], Carnicella, S. (Grenoble)[1]

[1]*GIN Inserm U836, Team 10 - Dynamic and Pathophysiology of Basal Ganglia, Grenoble, France*

Recent evidence supports the general view that Parkinson's disease is not merely a movement disorder but it also affects both cognitive and psychiatric functions. Amongst these non motor complications, apathy, which is defined as a lack of motivation and operationalized as a quantitative reduction of goal-directed behaviors, is a major source of disability. We recently developed a lesion-based model, with stereotaxic injection of 6-hydroxydopamine (6-OHDA) into precise areas of the rat SNc or VTA, in which degeneration of the DA mesocorticolimbic and nigrostriatal systems can be clearly separated. We found, in several operant tasks, that a partial denervation of the nigrostriatal, but not of the mesocorticolimbic, DA system induced a profound and selective motivational deficit during instrumental preparatory action. In order to further validate our experimental model, we investigate here the time-course of the effects of the nigrostriatal DA denervation on motivation and how pramipexole, a D2/D3 receptor agonist that has been shown to reverse apathy in PD, may reverse such deficit.
We showed in rats which have previously acquired operant sucrose self-administration, that SNc DA lesions induced a rapid and persistent reduction in operant performances. 12 days of subchronic intraperitoneal pramipexole administration fully reversed these deficits, that progressively reappeared after withdrawal from the DA treatment.
This longitudinal preclinical study therefore support the implication of the DA nigrostriatal system in PD-associated apathy. Moreover, by showing a good isomorphy and predictive validity, our model highlights the relevance of the dopaminergic D2/D3 receptors as potential targets for alleviating apathy in PD.

P2.071

Toggling serotonin 4 receptors in the nucleus accumbens provoked anorexia to binge-type eating

Hachem-Delaunay, S. (Montpellier)[1,2], Laurent, L. (Montpellier)[1,3], Pratlong, M. (Montpellier)[1,2], Jean, A. (Montpellier)[1], Martin, R. (Palo Alto)[4], Compan, V. (Montpellier)[1,2]
[1]*CNRS UMR 5203, INSERM U661, Univ. Montpellier I & II, Institut de Génomique Fonctionnelle, département de Nurobiologie, Montpellier, France,* [2]*Univ Nimes, Nimes, France,* [3]*Contralco, Gignac, France,* [4]*Roche Bioscience, Palo Alto, United States*

Eating adequately is essential to survival. However, highly stressed adolescents affected by *Anorexia nervosa* can restrict their caloric intake to dangerously low, and often mortal, levels. One complex facet of this illness is the transition between anorexia and overeating. Here, we tested whether the transition from the abusive consumption of food towards excessive food deprivation (anorexia-like behavior) depends on the constitutive activity of the serotonin receptor (5-HTR_4), within the nucleus accumbens (NAc), a structure of the brain's reward system. The constitutive activity in the absence of 5-HT is a spontaneous capacity of these G-protein coupled receptors (GPCRs) to activate their signaling pathways. This property was discovered *in vitro*, but its physiological consequences are unknown, with the exception of only one GPCR (melanocortin receptor). Following up on an ongoing study demonstrating that stimulation of the 5-HTR_4 (accumulation of active form: *R**) in the NAc activates an “addiction pathway” (AMPc/PKA/CART) to inhibit hunger, we found here that inhibition of the constitutive activity of 5-HTR_4 (accumulation of inactive form: *R*) following injection of a specific inverse agonist in the NAc, inhibits this pathway and provokes hyperphagia in fed and food-deprived male mice. In food-deprived mice, food binging further requires increases in the levels of the neuropeptide Y in the NAc. Transferring a gene (JetPEI^{TM}) encoding a “locked” 5-HTR_4 to 5-HT instead of the native 5-HTR_4, more constitutively active, in the NAc of wild-type and 5-HTR_4 knockout mice, triggers anorexia. Collectively, findings show that controls of the “accumbal addiction pathway” by *R* and *R** forms are opposed. Hence both extremes of the *R*/*R** equilibrium may drive paradoxical hypophagia (anorexia) to consumption of food (bulimia) because *R*/*R** state increases or decrease 5-HTR_4 activity. These findings represent a rare demonstration of the physiological relevance of the constitutive activity of GPCRs, and reveal new insights in the neurophysiology of feeding behavior.

P2.072

Ventral Pallidum and cortical structures involved in behavioural expression of anxiety: PET imaging study associated with local inhibitory dysfunction of external Pallidum territories in non-human primate

Galineau, L. (Tours)[1], Kas, A. (Paris)[2], Worbe, Y. (Paris)[3], Chaigneau, M. (Fontenay-aux-Roses)[4], Herard, A.-S. (Fontenay-aux-Roses)[4], Guillermier, M. (Fontenay-aux-Roses)[4], Delzescaux, T. (Fontenay-aux-Roses)[4], Féger, J. (Paris)[5], Hantraye, P. (Fontenay-aux-Roses)[4], Tremblay, L. (Bron)[6]
[1]UFR Sciences Pharmaceutiques-Univ. F. Rabelais, INSERM U.930 - Biophysique Médicale, Tours, France, [2]Service de Médecine Nucléaire, AP-HP, Groupe hospitalier Pitié-Salpêtrière, Paris, France, [3]Centre de Recherche de l'Institut du Cerveau et de la Moelle épinière, Paris, France, [4]MIRCEN, Fontenay-aux-Roses, France, [5]Institut du cerveau et de la moelle épinière, Paris, France, [6]Centre de Neuroscience Cognitive, UMR5229-CNRS, Université Lyon1, Bron, France

The external Globus Pallidus is involved in motor, cognitive and motivational functions through segregated sensorimotor, associative and limbic territories in a same relationship than all others Basal Ganglia structures. In a previous study, we demonstrated in monkey that microinjections of a GABA antagonist (bicuculline) in the different pallidal territories led to dyskinesia, hyperactivity state and stereotyped behaviors, respectively (Grabli et al. 2004). The stereotyped behavior produced from the Ventral Pallidum (VP), is a common feature of neurological or psychiatric disorders such as Tourette's syndrome or Obsessive Compulsive Disorders and had been thought to be a behavioral expression of an anxious state. In this study performed on same 4 monkeys, we compared the changes in cerebral metabolism consecutive to bicuculline injections in 3 different pallidal sites, either in limbic part (VP), in the sensorimotor or associative territories leading respectively to stereotyped behaviors (liking/biting fingers and grooming), abnormal movement or hyperactivity state. For this aim, we performed in vivo positron emission tomography using ^{18}F-2-fluoro-2-deoxy-D-glucose, as brain activity is closely related to brain glucose metabolism. Besides, the blood cortisol levels, a biomarker of anxiety, were measured in these animals for each condition. Our results showed distinct cerebral modifications in glucose consumption in the 3 experimental conditions with antero-ventral (frontal, insular and temporal cortex) and postero-dorsal (post-central sulcus and parietal cortex) increased activities for stereotyped behavior and abnormal movement, respectively, in addition to an intermediate profile for hyperactivity. The bicuculline injections in the VP led to significantly increased activities in the ventral prefrontal cortex, anterior insula and the amygdala, three cerebral structures known to be involved in aversive encoding information and anxiety disorders. In addition, significant elevated cortisol blood levels were observed in the stereotyped behavior condition vs no change for the two other conditions. Overall, our results strongly suggest the implication of VP in a cerebral network involved in the expression of behavioral disorders related to anxiety disorders.

P2.073

Bone marrow-derived M2 macrophages do not protect against ischemic brain damage *in vivo*

Desestret, V. (Lyon)[1,2], Riou, A. (Lyon)[2], Chauveau, F. (Lyon)[1,2], Cho, T.-H. (Lyon)[2], Devillard, E. (Lyon)[1], Marinescu, M. (Lyon)[2], Ferrera, R. (Lyon)[3], Rey, C. (Lyon)[4], Chanal, M. (Lyon)[1], Angoulvant, D. (Lyon)[3], Honnorat, J. (Lyon)[1], Nighoghossian, N. (Lyon)[2], Berthezène, Y. (Lyon)[2], Nataf, S. (Lyon)[1], Wiart, M. (Lyon)[2]
[1]Université de Lyon, Centre de Recherche en Neurosciences de Lyon (CRNL); CNRS, UMR 5292; INSERM, U1028; Université Lyon 1, Lyon, France, [2]Université de Lyon, Centre de Recherche en Acquisition et Traitement de l'Image pour la Santé (CREATIS); CNRS, UMR 5220; INSERM, U1044; INSA-Lyon; Hospices Civils de Lyon; Université Lyon 1, Lyon, France, [3]Université de Lyon, CarMeN;

INSERM, U1060; INRA, 1235; INSA-Lyon; Université Lyon 1, Lyon, France, [4]ProfilXpert-LCMT; CNRS, UMS 3453; INSERM, US7, Lyon, France

The inflammatory response following ischemic stroke is dominated by innate immune cells: resident microglia and blood-derived macrophages. The ambivalent role of these cells in stroke outcome might be explained in part by the acquisition of distinct functional phenotypes: classically (M1) and alternatively activated (M2) macrophages. Elucidating the impact of blood-derived macrophages on neuron viability after focal cerebral ischemia is of great interest, offering possible novel treatment strategies for stroke patients.
To shed light on the crosstalk between hypoxic neurons and macrophages, an *in vitro* model was set up in which bone-marrow-derived macrophages were co-cultured with organotypic hippocampal slice cultures (OHCs) subjected to oxygen and glucose deprivation (OGD). Dentate gyrus granular neurons were systematically affected by OGD. Macrophages provided potent protection against neuronal cell loss through a paracrine mechanism, and expressed M2 polarization.
These findings raised the possibility of using bone-marrow-derived M2 macrophages for cell-based intervention (CBI). Two million M2 macrophages (or vehicle) were administered intravenously during the subacute stage of ischemia (D4) in a model of transient middle cerebral artery occlusion. Therapeutic benefit was evaluated in rats with transient middle cerebral artery occlusion (tMCAO) and sham-operated rats on a two-weeks' serial sensorimotor testing. In parallel, the following MRI endpoints were longitudinally monitored: lesion size and brain swelling, blood-brain barrier integrity, and quantitative T2 values before and after injection of ultrasmall particles of iron oxide (USPIO), as neuroinflammation markers. No improvement in outcome, whether functional or lesional (as assessed by imaging and immunohistology), was observed in animals treated with M2 macrophages.
In conclusion, the present study failed to demonstrate any *in vivo* protective effect of M2 macrophages injected intravenously during the subacute stage of tMCAO, despite the neuroprotective effects observed *in vitro*. Immunomodulatory therapy, such as intravenous M2 macrophage supplementation, has to take into account the complexity and dynamics of post-ischemic cerebral inflammation and blood-brain barrier alterations.

P2.074

Neuroprotection of dopaminergic neurons in Parkinson disease models by small synthetic molecules, which cross the blood brain barrier

Le Douaron, G. (Paris)[1,2], Schmidt, F. (Paris)[1,2], Amar, M. (Paris)[1], Kadar, H. (Gif sur Yvette)[3], Séon-Méniel, B. (Châtenay-Malabry)[2], Ferrié, L. (Châtenay-Malabry)[2], Touboul, D. (Gif sur Yvette)[3], Brunelle, A. (Gif sur Yvette)[3], Michel, P.P. (Paris)[1], Raisman-Vozari, R. (Paris)[1], Figadère, B. (Châtenay-Malabry)[2]
[1]Hôpital de la Pitié-Salpétrière, INSERM UMRS 975-CNRS UMR 7225-CRICM, Institut du cerveau et de la moelle épinière, Paris, France, [2]CNRS-UMR 8076 BiOCIS, Université Paris-Sud, Châtenay-Malabry, France, [3]Institut de Chimie des Substances Naturelles, CNRS UPR 2301, Gif sur Yvette, France

Parkinson disease (PD) is a neurodegenerative disorder of ageing, characterized by invalidating motor symptoms resulting from the loss of dopaminergic (DA) neurons in the *substantia nigra*. New therapies are required to limit or halt progression of the disease. Recently our team synthesized hybrids of natural products (melatonin and fatty acid) with the aim of developing drugs possessing dual effects on DA neurons, namely neuroprotective and neuritogenic. We found a number of compounds exhibiting both activities in a PD model of midbrain cultures in which DA cell death is selective and progressive (*Bioorg. Med. Chem.* 2010, p.5103). The development of a first lead compound revealed its weak ability to cross the BBB. In order to increase this ability, which is essential for *in vivo* activity, we synthesized a 2nd generation of compounds, derived from amino-quinoxalines. After screening these molecules in the *in vitro* PD model, we were able to identify a compound, with neuroprotective activity and good physico-chemical properties that predict good BBB permeation based on Quantitative Structure-Activity Relationship study (QSARs). Two complementary analytical methods: HPLC-MS/MS and MALDI-TOF mass spectrometry imaging showed the presence of this compound in

brain tissue, and demonstrated its distribution on sagittal mouse brain sections, respectively. To evaluate the *in vivo* activity of our lead we used a MPTP lesioned mouse model, which is commonly used as *in vivo* animal model of PD. Finally, a 3rd generation library was synthesized and screened in order to ameliorate the neuroprotective activity of the quinoxaline lead compound. Our most recent results will be presented.

P2.075

A proteomic approach of how glioma stem cells control of their microenvironment

Chneiweiss, H. (Paris)[1], Lipecka, J. (Paris)[1], Thirant, C. (Paris)[1], Dias-Morais, A. (Paris)[1], Devaux, H. (Paris)[1], Varlet, P. (Paris)[1], Soncin, F. (Lille)[2], Virolle, T.V. (Nice)[3], Gavard, J. (Paris)[4], Junier, M.-P. (Paris)[1]
[1]Centre de Psychiatrie et Neurosciences, Inserm Umr 894 - Univ. Paris-Descartes, plasticité Gliale, Paris, France, [2]CNRS, Angiogenese, Lille, France, [3]Institut de Biologie Valrose (iBV) UMR CNRS 7277 - UMR INSERM 1091, Nice, France, [4]Inserm Institut Cochin, Paris, France

Glioblastomas are the most frequent adult primary brain tumors that still remain fatal despite major clinical efforts. As in other solid tumors, populations of glioblastoma stem-like cells (GSCs) endowed with tumor initiating and therapeutic resistance properties have been identified. GSCs and their progeny are intermingled with reactive stromal cells and endothelial cells to create a highly vascularized tumor. In one direction, endothelial cells and their secreted proteins are able to sustain GSC properties while, in turn, GSCs can promote neoangiogenesis, modulate endothelial cell functions and may even transdifferentiate into endothelial cells. We developed proteomic approaches (2D-DIGE and mass spectrometry using either MALDI MS/MS or orbitrap) to decipher the proteins involved in the GSC control of their microenvironment. For this purpose we compared cultured GCS with either the original tumoral tissue they were derived from, or normal human neural stem cells, or human brain microvessel endothelial cells, at the whole cell level or considering only their secretome. We first observed that the endothelial secretome is sufficient to maintain GSC stemness through a mTOR-dependent pathway. We then identified novel GSC-secreted molecules with pro-angiogenic properties (Semaphorin 3A, hepatoma-derived growth factor). This opens the path to the design of a concerted attack of glioblastoma vasculature that could overcome the development of resistance to single-targeted therapies while keeping away the toxicity of the treatments.

P2.076

Characterisation of copy number variations in early-onset bipolar disorders

Mansour, S. (Créteil)[1,2,3], Chevalier, L. (Créteil)[1], Nicolas, A. (Créteil)[1], Etain, B. (Créteil)[1,4], Ly, T. (Créteil)[1,3,5], Zelenika, D. (Evry)[6], Lathrop, M. (Evry)[6], Henry, C. (Créteil)[1,2,4], Kahn, J.-P. (Toul)[7], Bellivier, F. (Paris)[8,9], Leboyer, M. (Créteil)[1,2,4], Jamain, S. (Créteil)[1]
[1]INSERM U955 équipe 15, Hôpital Henri Mondor, Créteil, France, [2]Université Paris Est, Faculté de Médecine, Créteil, France, [3]Fondation FondaMental, Créteil, France, [4]AP-HP, Hôpital H. Mondor, A. Chenevier, Pôle de Psychiatrie, Créteil, France, [5]AP-HP, Hôpital H. Mondor - A. Chenevier, Plateforme de Ressources Biologiques, Créteil, France, [6]Commissariat à l'Energie Atomique, Institut Génomique, Centre National de Génotypage, Evry, France, [7]CHU de Nancy, Hôpital Jeanne-d'Arc, Département de Psychiatrie et de Psychologie clinique, Toul, France, [8]AP-HP, Hôpital Saint-Louis-Lariboisière-F, Service Universitaire de Psychiatrie, Paris, France, [9]Faculté de Médecine, Université Denis Diderot, Paris, France

Bipolar disorder is a severe and chronic mood disorder characterised by alternating episodes of mania and depression. The lifetime prevalence of bipolar disorder is about 1% in general population and the

illness is associated with a considerable morbidity and a high lifetime risk of suicide. Despite a complex inheritance, twin and family studies suggest a strong genetic contribution in vulnerability to bipolar disorder. An early onset form of the disease is associated with more severe symptoms and a higher familial risk. Recent studies demonstrated an enrichment of rare copy number variations (CNVs) in patients with bipolar disorder, difference that was emphasized in subjects with an early disease onset.
We conducted a CNV analysis in a cohort of 236 subjects with early-onset bipolar disorder, using HumanHap550 Illumina beadchips. Nine rare CNV (eight deletions and one duplication) were found to affect brain-expressed genes and were not previously described in public databases for genomic variants. Interestingly, all of them encode molecules involved in synapse formation or function. We validated seven out of the nine CNVs by quantitative PCR and determined the breakpoint for four of the eight deletions. We then sequenced the coding regions of the disrupted genes in 200 patients with early-onset bipolar disorder and identified several mutations, predicting amino acid changes and modification in protein structure or function.
In conclusion, our findings suggest that rare variations in genes encoding synaptic molecules might play a role in vulnerability to early onset form of bipolar disorder.

P2.077

L-Dopa restores the glutamate concentration in putamen in Parkinson's disease - a ^{1}H magnetic resonance spectroscopy study

Mazuel, L. (Clermont Ferrand)[1], Chassain, C. (Clermont Ferrand)[2], Jean, B. (Clermont Ferrand)[2], Cladière, A. (Clermont Ferrand)[2], Speziale, C. (Clermont Ferrand)[2], Durif, F. (Clermont Ferrand)[1,3]
[1]Univ Clermont 1, UFR Médecine, EA7280, Clermont Ferrand, France, [2]CHU Gabriel Montpied, Centre d'Imagerie par Résonance Magnétique, Clermont Ferrand, France, [3]CHU Gabriel Montpied, Service de Neurologie, Clermont Ferrand, France

Parkinson's disease (PD) is characterized by the loss of dopaminergic neurons inside the Substantia Nigra pars compacta (SNpc). The degeneration leads to an alteration of the dopaminergic nigrostriatal pathway that induces synaptic changes inside the striatum. Especially, studies on model animals of PD suggest an overactive glutamatergic pathway inside the striatum after dopaminergic depletion that is reversed after L-Dopa treatment (Chassain et al., 2008; 2010). Therefore, the aim of this study was to analyze, by a non-invasive way, the specificity of the metabolic brain profile of parkinsonian patients.A single-voxel ^{1}H NMR spectroscopy (MRS) was carried out at 3T (PRESS; TR = 1.5s; TE = 29ms; voxel size = 1cm^{3}). 10 healthy patients (6 men and 4 women) were compared to 10 patients with a PD. Patients and volunteers were matched for age and sex. The average duration of PD evolution was 8.5 ±6.8 years. The measurement of the metabolic profile was realized bilaterally inside the putamen. For parkinsonian patients, the effect of L-DOPA treatment (Modopar® dispersible, 200 mg, acute administration) was also studied by comparing patients in MED-OFF vs. MED-ON condition in the same day during 2 MRS sessions. The absolute concentrations of metabolites were determined using jMRUI software (Lyon, France). We observed a significant increase of glutamate (GLU) concentration inside the putamen of parkinsonian patients in MED-OFF compared to healthy volunteers (7.7 ± 1.8 vs. 10.18 ± 1.4 mmol/L; $p < 0.001$) which was reversed by L-DOPA (MED-ON : 9.1 ± 2.1 mmol/L vs. MED-OFF : 10.18 ± 1.4 mmol/L; $p < 0.05$). No significant changes of creatine concentration were observed between healthy volunteers and MED-OFF patients.Thus, for the first time, the present study reports an overactive glutamatergic pathway inside the putamen in parkinsonian patients. This may leads to the use of the GLU increase as a marker of the disease. Moreover, the normalization of the GLU levels after dopaminergic treatment could be used as a marker of the treatment efficiency.

P2.078

Prenatal prevention of epilepsy: maternal treatment prevents embryonic neuronal migration defects and epileptic activity caused by rat *Srpx2* silencing *in utero*

Salmi, M. (Marseille)[1], Bruneau, N. (Marseille)[1], Cillario, J. (Marseille)[1], Lozovaya, N. (Marseille)[1], Massacrier, A. (Marseille)[1], Buhler, E. (Marseille)[1], Cloarec, R. (Marseille)[1], Tsintsadze, T. (Marseille)[1], Watrin, F. (Marseille)[1], Tsintsadze, V. (Marseille)[1], Zimmer, C. (Marseille)[2], Villard, C. (Marseille)[3], Lafitte, D. (Marseille)[3], Cardoso, C. (Marseille)[1], Bao, L. (Shangai)[4], Muscatelli, F. (Marseille)[1], Pauly, V. (Marseille)[5], Khalilov, I. (Marseille)[1], Durbec, P. (Marseille)[2], Ben Ari, Y. (Marseille)[1], Burnashev, N. (Marseille)[1], Represa, A. (Marseille)[1], Szepetowski, P. (Marseille)[1]
[1]Mediterranean Institute of Neurobiology (INMED), INSERM UMR_S901, Aix-Marseille University, Marseille, France, [2]IBDML, CNRS UMR6216, Aix-Marseille University, Marseille, France, [3]PIT2 Platform, CRO2, INSERM UMR911, Aix-Marseille University, Marseille, France, [4]State Key Laboratory of Cell Biology, Institute of Biochemistry and Cell Biology, Shangai, China, [5]SPIM, AP-HM, Marseille University Hospital, Marseille, France

Altered development of the human cerebral cortex can cause severe malformations with often intractable focal epileptic seizures and may participate in common pathologies, notably epilepsy. This raises important conceptual and therapeutic issues. Ideally the patients would benefit from as early intervention as possible, before the long-term epileptic consequences appear later in life - hence the need to test the feasibility of such preventive strategies in the appropriate animal models. We had previously reported on the detection of two missense *SRPX2* (Sushi-Repeat Protein, X-linked 2) mutations in two related epileptic disorders of the speech cortex. Using an *in utero Srpx2* silencing approach, we have now created a rat model of altered development of the cerebral cortex, that was analyzed using a large combination of complementary approaches, including morphological analyzes, time-lapse videomicroscopy and electrophysiology recordings. Here we demonstrate that Srpx2 influences neuronal migration in the developing rat cerebral cortex, and that it increases alpha-tubulin acetylation. Following *in utero Srpx2* silencing, spontaneous epileptiform activity was recorded postnatally. The neuronal migration defects and the postnatal epileptiform consequences were prevented early in embryos by transient maternal administration of a tubulin deacetylase inhibitor. Early prevention of the neuronal migration defects and of the postnatal epileptic consequences as obtained here, could be of broad interest given the known convergence of multiple neuronal migration pathways and disorders on alpha-tubulins, and as future progress in developmental neuroimaging and in prenatal genetic diagnosis can be anticipated.
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All three first authors contributed equally to the study.

P2.079

Amyloid beta oligomers disrupt plasticity-related calcium events in dendritic spines

Rush, T. (La Tronche)[1], van den Berg, N. (La Tronche)[1], De Seranno, S. (La Tronche)[1], Frandemiche, M.L. (La Tronche)[1], Dollmeyer, M. (La Tronche)[1], Lanté, F. (La Tronche)[1], Buisson, A. (La Tronche)[1]
[1]Grenoble Institut des Neurosciences, INSERM U836 - UJF, La Tronche, France

Alzheimer's disease (AD) is the leading cause of dementia affecting an estimated 34 million people. AD is characterized by the production of amyloid beta, the appearance of tau- positive neurofibrillary tangles and the malfunction and loss of dendritic spines. Disruption of synaptic function and plasticity correlates better with disease progression than the gross loss of synapses observed later in AD. Soluble, amyloid beta oligomers (Aβo) are of specific interest as they appear prior to fibril formation and plaque deposition. Accumulating evidence strongly suggests a causal role for Aβo in disease. Here, we used primary cortical neurons cultured from mice to study the effects of Aβo on neuronal signaling and plasticity. We monitored spontaneous up-states and those evoked by an indirect stimulation protocol (i.e. chemical LTP) using a GABA-A receptor antagonist (bicuculine, Bic, 50 µM), and a weak K+-channel blocker (+4-aminopyridine, 4AP, 2.5 mM).

Live confocal imaging of neurons transfected with the genetically encoded calcium indicator, GCaMP5, allowed us to determine the relative calcium levels within specific compartments, namely dendritic branches, spines or soma. Line-scans through individual spines enabled high temporal resolution of up-state events. Exposure to Aβo (100 nM) for 15 minutes did not affect basal up-state rate or intracellular calcium. However, presence of Aβo precluded the large calcium rise evoked by Bic+4AP treatment. We further assessed postsynaptic effects of Aβo under the Bic+4AP stimulation protocol by isolating the post-synaptic density-(PSD) enriched synaptic membrane compartments. Western blot analysis of these samples revealed that Aβo associate specifically with the PSD. Further, cultures exposed to Bic+4AP exhibit a robust increase in the AMPA-receptor GluR1 subunit at the PSD that is blocked by exposure to Aβo. PSD-sequestration of the proteasome is also necessary for synaptic plasticity; evaluation of the RPT1 proteasome subunit suggests that such sequestration is also sensitive to Aβo. These findings are congruent with numerous reports of Aβ effects on LTP and LTD and suggest that Aβo impair the ability of the spines to respond to changes in signaling.

P2.080

Specific deletion of *Lgi1* in forebrain excitatory neurons causes seizures in mouse models of temporal lobe epilepsy

Boillot, M. (Paris)[1], Huneau, C. (Rennes)[2], Dufresnois, B. (Paris)[1], Lehongre, K. (Paris)[1], Martin, B. (Rennes)[2], Le Guern, E. (Paris)[1], Baulac, S. (Paris)[1]
[1]Centre de Recherche de l'Institut du Cerveau et de la Moelle épinière, Paris, France, [2]Université Rennes 1, Rennes, France

Mutations of the *LGI1* (*leucine-rich, glioma-inactivated 1*) gene cause autosomal dominant epilepsy with auditory features (ADEAF), a focal inherited epilepsy syndrome of adolescence onset. Additionally, the LGI1 protein is the main antigen in patients with autoimmune encephalitis. Thus, the secreted neuronal protein LGI1, which brain function remains to be elucidated, appears to be a key protein for a spectrum of epileptic disorders. We have previously generated constitutive knockout (KO) mice for *Lgi1.* All exhibited spontaneous recurrent epileptic seizures from the second postnatal week and did not survive beyond 3 weeks of age. To dissect the role of Lgi1 in early stages of life or in adulthood in the excitatory component of neuronal networks, we generated two cortical glutamatergic neuron-specific *Lgi1* conditional KO lines, in which the loss of Lgi1 occurs during embryonic development (Emx1-cKO mice) or is delayed to 5 postnatal weeks (CaMKII-cKO mice). Here, we report the emergence of spontaneous epileptic seizures and a reduced lifespan in both conditional KO mice, with a severity of phenotype correlated to the timing of deletion. Our results demonstrate an important contribution of glutamatergic neurons in this epilepsy and highlight the critical role played by Lgi1 throughout the entire life.

P2.081

Major depressive episode and pharmacogenetic response to tianeptine associated with NR3C1, FKBP5 and CRHR1 genes

Ramoz, N. (Paris)[1], Le Strat, Y. (Paris)[1], Courtet, P. (Montpellier)[2], Gorwood, P. (Paris)[1,3]
[1]Centre de Psychiatrie et de Neurosciences, INSERM UMR 894, Paris, France, [2]CMM INSERM U888, Department of Psychological Medicine and Psychiatry, Pôle Urgences, Lapeyronie Hospital, CHU de Montpellier, Montpellier, France, [3]CMME. Hospital Sainte-Anne (University Paris-Descartes), Paris, France

Major depressive episode (MDE) is the most frequent psychiatric disorder, affecting 5-10% of the world, with a sex ratio of 2 women for 1 man. Antidepressant drugs are available for treatment of MDE

but more than 30% of patients failed to respond to this therapy. Treatment response to antidepressant drugs is modulated by genetic factors. One antidepressant treatment, the tianeptine molecule does not inhibit serotonin reuptake, as usual, but it modulates the glutamatergic pathway. A potential target of this treatment is the the hypothalamic-pituitary-adrenal axis dysregulated in MDE. Thus, tianeptine can target the glucocorticoid receptor encoded by NR3C1 gene, the co-chaperone regulating the glucocorticoid-receptor sensitivity encoded by FKBP5 gene and, the corticotrophin-releasing hormone receptor 1 (CRHR1 gene) associated with stress-related psychopathology. Thus, our goal was to study the pharmacogenetic response to this specific tianeptine treatment in MDE. In this aim, our work was the genotyping of markers in candidate genes NR3C1, FKBP5 and CRHR1 in outpatients treated with tianeptine for a MDE to search for an association to a positive treatment response.
A total of 3771 outpatients were treated with tianeptine for a MDE during 4 to 8 weeks. The criteria for a MDE were examined by the clinicians according to the DSM-IV diagnosis and the duration of each symptom was recorded during the inclusion, and at 4 to 8 weeks of treatment. The Hospital Anxiety and Depression Scale (HAD) was evaluated at the two visits. DNA was extract from saliva sample and screening of single nucleotide polymorphisms (SNPs) was carried out by Taqman assays.
A majority of patients (62%) were women. About 54% of patients were treated for a first MDE. A total of 15 SNPs were genotyped in 3 candidate genes. Only two SNPs, both within the same gene, CRHR1, were found associated with a positive treatment response. The C allele of rs878886 was over-represented in responders (p=0.001, Odd Ratio=1.2 Confident Interval 95% 1.1-1.4) and the T allele of rs16940665 was over-presented in responders compared to non-responders (p=0.001 OR=1.3 CI95%=1.1-1.5).
We found a pharmacogenetic association between variants of CRHR1 gene and the response to tianeptine in major depressive episode.

P2.082

Involvement of the nigrostriatal dopaminergic pathway in the oro-facial neuropathic pain

Dieb, W. (Clermont-Ferrand)[1], Ouachikh, O. (Clermont-Ferrand)[1], Durif, F. (Clermont-Ferrand)[1,2], Hafidi, A. (Clermont-Ferrand)[1]
[1]Faculté de médecine, EA7280, Clermont-Ferrand, France, [2]CHU Gabriel Montpied, Clermont-Ferrand, France

Positron emission tomography (PET) studies suggest that the dopaminergic system plays a role in burning mouth syndrome and atypical facial pain (Hagelberg et al. 2003). Although some animal studies suggest that this central system plays a role in pain modulation (Takeda et al. 2005), there exists no animal model study which explores the implication of the nigrostriatal dopaminergic pathway in trigeminal neuropathic pain. Therefore, we investigated two major symptoms of trigeminal neuropathic pain, static and dynamic mechanical allodynia, in the rat model of Parkinson disease (Paillé et al. 2007). In this model a bilateral 6-OHDA injection introduced into the medial forebrain bundle produces a lesion of the nigrostriatal dopaminergic pathway. Lesioned animals showed significant dynamic and static mechanical allodynia in the orofacial area that occurred from 4 days to 5 weeks post-injury. To investigate if a segmental mechanism is implicated in dopamine depletion induced neuropathic pain, the expression of the isoform gamma of protein kinase C (PKCg) has been studied in the medullary dorsal horn (MDH) in lesioned and sham animals. There was a large increase in PKCg expression in superficial laminae of the MDH. Also, intraperitoneal and intracisternal administrations of bromocriptine, a dopamine 2 receptor agonist, decreased both dynamic and static mechanical allodynia. Altogether, these data demonstrate for the first time that the nigrostriatal dopaminergic depletion produces trigeminal neuropathic pain which can be decreased by dopamine 2 receptor agonist administration.

Reference:

1- Hagelberg N, Forssell H, Aalto S, Rinne JO, Scheinin H, Taiminen T, Nagren K, Eskola O, Jaaskelainen SK., Altered dopamine D2 receptor binding in atypical facial pain. Pain 2003; 106:43-48.
2- Paillé, V., Henry, V., Lescaudron, L., Brachet, P., Damier, P., Rat model of Parkinson's disease with bilateral motor abnormalities, reversible with levodopa, and dyskinesias. 2007; Mov. Disord. 22, 533-539.

3- Takeda R, Ikeda T, Tsuda F, Abe H, Hashiguchi H, Ishida Y, Nishimori T., Unilateral lesions of mesostriatal dopaminergic pathway alters the withdrawal response of the rat hindpaw to mechanical stimulation. Neurosci Res 2005; 52:31-36.

P2.083

Loss of function of glucocerebrosidase GBA2 is responsible for motor neuron defects in hereditary spastic paraplegia

Stevanin, G. (Paris)[1,2], Martin, E. (Paris)[1,2], Rastetter, A. (Paris)[1,2], Schule, R. (Tuebingen)[3], Smets, K. (Antwerp)[4], Boukhris, A. (Sfax)[5], Loureiro, J. (Porto)[6], Mundwiller, E. (Paris)[1], Wessner, M. (Evry)[7], Lamari, F. (Paris)[8], Schols, L. (Tuebingen)[3], de Jonghe, P. (Antwerp)[4], Zuchner, S. (Miami)[9], Kabashi, E. (Paris)[1], Brice, A. (Paris)[1]
[1]Institut Du Cerveau Et de la Moelle Épinière - Cricm - Neb, Paris, France, [2]EPHE, Paris, France, [3]Hertie-Institute for Clinical Brain Research and Center for Neurology, Tuebingen, Germany, [4]University of Antwerp, Antwerp, Belgium, [5]Hôpital Universitaire Habib Bourguiba, Sfax, Tunisia, [6]UNIGENE, Porto, Portugal, [7]Genoscope, Evry, France, [8]CHU Pitié-Salpêtrière, Paris, France, [9]University of Miami, Miami, United States

Hereditary spastic paraplegias (HSPs) are heterogeneous inherited neurodegenerative disorders. Affected individuals suffer mainly from pyramidal motor neuron dysfunction caused by degeneration of upper motor neurons. *Spastic paraplegia 46* refers to a locus mapped to chromosome 9 that accounts for a complicated autosomal recessive form of HSP. Using next-generation sequencing in three independent families, we identified four different mutations in *GBA2*, three truncating and one missense variants, which were found to cosegregate with the disease and were absent in controls. *GBA2* encodes a microsomal non-lysosomal glucosylceramidase that catalyzes the conversion of glucosylceramide to free glucose and ceramide and the hydrolysis of bile acid 3-O-glucosides. The missense variant was also found at the homozygous state in a simplex case in whom no residual glucocerebrosidase activity of GBA2 could be evidenced in blood cells, opening the way to a possible measurement of this enzyme activity in clinical practice. The overall phenotype was a complex HSP with mental impairment, cataract and hypogonadism in males associated with various degrees of corpus callosum and cerebellar atrophy on brain imaging. Antisense morpholino oligonucleotides targeting the zebrafish *GBA2* orthologous gene led to abnormal motor behavior and axonal shortening/branching of motoneurons that were rescued by the human wild-type mRNA but not by applying the same mRNA containing the missense mutation. This study highlights the role of ceramide metabolism in HSP pathology.

P2.084

Metabolic scintigraphic imaging in rat models of ADHD-C and ADHD-PI phenotypes

Desfosses, E. (Tours)[1], Lepelletier, F.-X. (Tours)[1], Tauber, C. (Tours)[1], Chalon, S. (Tours)[1], Castelnau, P. (Tours)[1], Galineau, L. (Tours)[1]
[1]INSERM U930 Imagerie et cerveau, Equipe 3 : 'Imagerie moléculaire du cerveau', Tours, France

Attention-Deficit/Hyperactivity Disorder (ADHD) is a neurodevelopmental disorder characterized by hyperactivity, impulsivity and inattention. It is diagnosed in 7 to 8% of the school-aged children and frequently persists through adulthood. To date, the pathophysiology of this disorder remains unclear, even if the neuropsychological theories of ADHD support the presence of alterations in executive functions and motivation in patients. Besides, the clinical neuroimaging data showed abnormalities in the fronto-striatal networks and in the dopamine and norepinephrine neurotransmission systems in

ADHD patients. Three ADHD subtypes are commonly defined including patients with predominant inattention (ADHD-PI), hyperactivity/impulsivity (ADHD-HI) or all the symptoms (ADHD-C). However, this phenotypic heterogeneity is rarely taken into account by the clinical imaging studies.
In this study, we aimed to better define the functional cerebral alterations underlying each symptom by using rat models of ADHD and scintigraphic imaging.
It has been already demonstrated that combinations of specific strains of SHR and WKY rats can be used to model the ADHD-PI and ADHD-C phenotypes (Sagvolden et al. 2009). On the basis that brain activity is closely related to brain glucose metabolism, we performed in vivo positron emission tomography using ^{18}F-2-fluoro-2-deoxy-D-glucose (^{18}FDG) on awake SHR and WKY rats.
The results showed that rats with ADHD-C or ADHD-PI phenotypes displayed distinct brain metabolic profiles in the fronto-striatal regions. The two phenotypes exhibited significant bilateral increased metabolism in the nucleus accumbens in addition to decreased metabolism in the insular cortex. However, the ADHD-C phenotype also displayed increased metabolic activities in the dorsomedial striatum, and cingulate cortex in addition to decreased metabolic activities in the prelimbic cortex.
These preclinical neuroimaging data provide new findings, which could help to better understand the mechanisms of ADHD and strongly suggest the implication of functional alterations in limbic networks in the expression of the inattentive phenotype observed in such animal models.

P2.085

Contribution of P2X4 to transcriptional remodeling in an experimental mouse model of epilepsy

Hirbec, H. (Montpellier)[1], Peyroutou, R. (Montpellier)[2], Levavasseur, F. (Paris)[3], Croze, S. (Lyon)[4], Nazaret, N. (Lyon)[4], Lachuer, J. (Lyon)[4], Audinat, E. (Paris)[3], Rassendren, F. (Montpellier)[2]
[1]Institut de Génomique Fonction - Univ. de Montpellier 1 & 2, CNRS UMR 5203-INSERM U661, Dépt. de pharmacologie moléculaire, Montpellier, France, [2]Institut de Génomique Fonction - Univ. de Montpellier 1 & 2, Montpellier, France, [3]INSERM U603 - CNRS UMR 8154, Paris, France, [4]ProfilXpert-LCMT; CNRS, UMS 3453; INSERM, US7, Lyon, France

Extracellular ATP (eATP) is a signaling molecule involved in cell-to-cell communication. eATP is also a danger signal released upon pathogen infection or during sterile inflammation.
eATP is sensed by two main classes of purinergic receptors : the P2RY, which are a family of G-protein-coupled receptors, and the P2RX a family of ATP-gated channels. Expression of both P2RY and P2RX has been reported in neurons and glial cells. P2X4 receptors are of special interest since they are highly permeable to calcium. However, their functions within the CNS are still poorly understood. In the hippocampus, P2RX4 are expressed in pyramidal neurons where they are activated during episodes of sustained activity. Furthermore, P2RX4 mRNA is up-regulated after kainate-induced *status epilepticus (SE)*, this up-regulation being mostly restricted to activated microglia.
Because of P2XR4 high calcium permeability, we hypothesized that this receptor contributes to the hippocampus remodeling observed after *SE*. In this study, we used a genome wide approach to investigate the potential involvement of P2RX4 in a model of Kainate-induced *SE*. We thus performed a transcriptional profiling of control and kainate-treated hippocampus and compared the transcriptional remodeling of the tissue in wild-type (WT) and P2X4$^{-/-}$ mice.
Our results show that under control conditions, hippocampus from WT and KO mice express very similar repertoire of genes. However, comparison of the transcriptional remodeling induced by kainate-evoked *SE* in WT and KO reveals that the expression of genes involved in inflammatory responses are significantly reduced in P2X4-deficient mice. These results demonstrate that P2RX4 are involved in the transcriptional remodeling of inflammatory genes within the CNS in a model of sterile inflammation.

P2.086

Dclk3, a molecular marker of the striatum that protects against Nterminal domains of mutant huntingtin

Francelle, L. (Fontenay-aux-Roses)[1], Galvan, L. (Fontenay-aux-Roses)[1], Gaillard, M.-C. (Fontenay-aux-Roses)[1], Liot, G. (Orsay)[2,3,4], de Chaldée, M. (Gif sur Yvette)[5], Guillermier, M. (Fontenay-aux-Roses)[1], Houitte, D. (Fontenay-aux-Roses)[1], Chaigneau, M. (Fontenay-aux-Roses)[1], Bonvento, G. (Fontenay-aux-Roses)[1], Malgorn, C. (Fontenay-aux-Roses)[1], Petit, F. (Fontenay-aux-Roses)[1], Jan, C. (Fontenay-aux-Roses)[1], Dufour, N. (Fontenay-aux-Roses)[1], Elalouf, J.-M. (Gif sur Yvette)[5], Humbert, S. (Orsay)[2,3,4], Saudou, F. (Orsay)[3,4], Déglon, N. (Fontenay-aux-Roses)[1], Brouillet, E. (Fontenay-aux-Roses)[1]
[1]CEA - CNRS URA 2210, Mircen, Equipe 1 - Interaction cellulaire, Fontenay-aux-Roses, France, [2]Institut Curie, Orsay, France, [3]CNRS UMR 3306, Orsay, France, [4]INSERM U1005, Orsay, France, [5]CEA, Institut de Biologie et de Technologies de Saclay (iBiTec-S), Service de Biologie Intégrative et Génétique Moléculaire (SBIGeM), Gif sur Yvette, France

Huntington´s disease (HD) is an inherited neurodegenerative disorder caused by an abnormal polyglutamine expansion in the protein Huntingtin (Htt). Mutant Htt, despite its ubiquitous expression in the brain, leads to preferential neurodegeneration of the striatum through unknown mechanisms. Our working hypothesis is that gene products selectively expressed in the striatum could be involved in the high vulnerability of striatal neurons to mutant Htt. In the present study, we show that overexpression using lentiviral vectors of a newly identified “striatal” gene product, Doublecortin-like kinase 3 (DCLK3), is neuroprotective against mutant Htt in primary culture of striatal neurons and in the mouse striatum *in vivo.* Since the function of DCLK3 is totally unknown, we generated mutants and truncated fragments of DCLK3 and found that the neuroprotective effects of the protein are associated with its C-terminus part that contains a putative kinase domain. We obtained results from autophosphorylation experiments that show that DCLK3 is actually a functional kinase. We identified mechanisms through which DCLK3 could possibly modulate striatal degeneration: DCLK3 is cleaved in different cultured cells and *in vivo*. Its cleavage in transgenic BACHD mice (HD mouse model) is different from that found in WT mice. Recombinant DCLK3 is localized in the cytoplasm with a higher density in the perinuclear region, according to a web-like organization reminiscent of the cytoskeleton. Biochemical and confocal microscopy analyses indicate that DCLK3 interacts with microtubules.DCLK3 mRNA expression is significantly reduced in the striatum of knock-in mouse model of HD (KI140CAG). These novel results unravel a key role of Dclk3 in striatal neuron survival and suggest that this newly identified kinase may be an important determinant of striatal vulnerability in HD. The identification of the neuroprotective mechanisms produced by DCLK3 could lead to novel potential therapeutic strategies for HD and possibly other disorders involving the striatum.

P2.087

Sensory integrations in amyotrophic lateral sclerosis

Sangari, S. (Paris)[1], Iglesias, C. (Paris)[2], El Mendili, M.-M. (Paris)[3], Benali, H. (Paris)[2], Pradat, P.-F. (Paris)[4], Marchand-Pauvert, V. (Paris)[1]
[1]Groupe Hospitalier Pitié Salpêtrière, ER6 - UPMC,Service de Médecine Physique et Réadaptation, Paris, France, [2]UMR_S678 INSERM/UPMC, Paris, France, [3]UMR_S678 INSERM/UPMC, IHU Neurosciences Hôpital Pitié-Salpêtrière, Paris, France, [4]UMR_S678 INSERM/UPMC, Neurologie Hôpital Pitié-Salpêtrière, Paris, France

Amyotrophic lateral sclerosis (ALS) is an adult neurodegenerative disease considered as a purely motor disease. However, spinal magnetic resonance imaging (MRI) has revealed denervation and demyelination of sensory ascending tracts in ALS patients without sensory clinical signs. This unexpected result raised the question whether abnormal sensory input could participate in the pathophysiological mechanisms. MRI provides anatomical metrics but does not inform on possible functional impairment. The aim of the project was to investigate sensory integration at spinal and cortical levels, using electrophysiology, which allows testing the excitability of neural networks.

In 11 patients and 8 age- and sex-matched healthy subjects, median and ulnar nerves were stimulated electrically to activate group Ia afferents from hand muscles (clinically impaired). The resulting somesthesic evoked potential (SEP) were analyzed to evaluate the integration at cortical level. Transcranial magnetic stimulation (TMS) was applied over primary motor area in order to evoke motor evoked potential (MEP) in triceps bracchii (clinically non-affected). Median and ulnar-induced modulations of MEP size were investigated to evaluate the sensory-motor integration at spinal (motoneuron) level.
We observed no difference in SEP latency but its amplitude was smaller in patient group. The triceps MEP latency and recruitment curve were similar in both groups, supporting the clinical observation that triceps motoneurons were not affected at this stage of the disease. Median and ulnar nerve stimuli increased MEP size in healthy subjects due to the convergence of group Ia and corticospinal inputs at motoneuron level. The MEP facilitation was increased in patient group.
These preliminary results suggest central alteration of the transmission of the sensory inputs at both spinal and cortical levels in ALS patients. At spinal level, alteration of the synapse between group Ia afferents and motoneurons may contribute to the change in motoneuron homeostasis and its degradation. Our results also suggest abnormal transmission at cortical level but it seems that this did not affect the corticospinal transmission given that MEP looked normal in triceps.

P2.088

PINK1 and Parkin associate with the TOM machinery to trigger mitochondrial clearance

Corti, O. (Paris)[1], Bertolin, G. (Paris)[1], Ferrando-Miguel, R. (Paris)[1], Jacoupy, M. (Paris)[1], Dauphin, A. (Paris)[1], Waharte, F. (Paris)[2], Traver, S. (Paris)[1], Lombès, A. (Paris)[3], Bulteau, A.-L. (Paris)[1], Fon, E.A. (Montréal)[4], Brice, A. (Paris)[1]
[1]Institut Du Cerveau Et de la Moelle Épinière - Cricm - Neb, UPMC UMR_S975, Inserm U 975, CNRS UMR 7225, Paris, France, [2]Institut Curie, Cell and Tissue Imaging, Paris, France, [3]Institut Cochin, Inserm U 1016, Paris, France, [4]Neurological Institute, McGill University, Centre for Neuronal Survival, Montréal, Canada

Mutations in the genes encoding the cytosolic E3 ubiquitin-protein ligase Parkin and the mitochondrial serine/threonine kinase PINK1, cause early onset autosomal-recessive PD. Genetic studies in Drosophila demonstrated that *parkin* and *PINK1* cooperate within a pathway involved in the regulation of mitochondrial morphology and dynamics. Since 2008, a number of reports extended an initial discovery made in Richard Youle's laboratory demonstrating and involvement of the PINK1/Parkin pathway in the clearance of dysfunctional mitochondria : after dissipation of the mitochondrial membrane potential triggered by the protonophore carbonyl cyanide m-chlorophenyl hydrazone (CCCP), PINK1 accumulates on the outer mitochondrial membrane (OMM), recruits Parkin and initiates the degradation of dysfunctional mitochondria by the proteasome and autophagy pathways. We addressed the possibility that the mitochondrial recruitment of Parkin by PINK1 is caused by alterations in mitochondrial protein import efficiency in different paradigms of mitochondrial import block, caused by molecular tools or CCCP treatment. Confocal microscopy and the FRET (Förster's Resonance Energy Transfer) and FLIM (Fluorescence Lifetime Imaging Microscopy) techniques were used to analyse co-localization and investigate protein-protein interactions on the OMM in these models. Our results provide evidence that PINK1 and Parkin associate on the OMM in proximity of the Translocase of Outer Membrane (TOM), a multiprotein complex responsible for importing virtually all mitochondrial proteins encoded by the nuclear genome, including PINK1. Investigation of the role of the TOM complex in the mitochondrial clearance by RNA interference against specific TOM subunits revealed that destabilization of its core structure is sufficient to prime mitochondria for autophagy in the absence of PINK1 and Parkin. Altogether, our results indicate that the TOM machinery is a key molecular switch of the PINK1/Parkin pathway, coupling loss of mitochondrial protein import efficiency with the removal of dysfunctional mitochondria.

P2.089

PET imaging study of dopamine and serotonin compensations on asymptomatic MPTP-intoxicated monkeys during early stage and after recovery of motor symptoms

Ballanger, B. (Bron)[1], Beaudoin-Gobert, M. (Bron)[1], Neumane, S. (Bron)[1], Epinat, J. (Bron)[1], Broussolle, E. (Bron)[1,2], Costes, N. (Lyon)[3], Lavenne, F. (Lyon)[3], Lebars, D. (Lyon)[3], Zimmer, L. (Lyon)[3], Sgambato-Faure, V. (Bron)[1], Tremblay, L. (Bron)[1]

[1]Université Lyon 1 - CNRS, Centre de Neuroscience Cognitive - UMR 5229, Equipe Physiopathologie des Ganglions de la Base, Bron, France, [2]Hospices Civils de Lyon, Hôpital Neurologique Pierre Wertheimer, Service de Neurologie C, Bron, France, [3]CERMEP - Centre d'imagerie du Vivant, Lyon, France

Patients with Parkinson's disease express motor symptoms only after 60-80% depletion of striatal dopamine (DA) probably thanks to compensatory mechanisms taking place during the early phase of the disease. However the neurochemical mechanisms involved during this period are still unclear. The aim of the present study, performed on MPTP-intoxicated monkeys, was to address the role of the dopaminergic (DA) and serotonergic (5-HT) systems using a longitudinal protocol in PET with DA and 5-HT-targeted ligands ([^{18}F]Dopa; [^{11}C]Raclopride;[^{11}C]DASB and [^{18}F]MPPF). After progressive MPTP intoxication (Mounayar et al., 2007), almost all monkeys (5/7) have developed motor symptoms and then fully recovered of their symptoms in about 3-5 weeks after the last MPTP injection. During the early asymptomatic phase, we mainly observed a significant increase of [^{18}F]Dopa uptake (pre-synaptic DA activity) in the pallidum and the ventral Prefrontal Cortex (vPFC) and an increase of [^{11}C]Raclopride (D2/D3 receptors) binding potential (BP) within the posterior putamen. In addition, an increased [^{18}F]MPPF BP has been shown in the anterior cingulate cortex (ACC), the dorsolateral and medial PFC, as well as in anterior caudate and ventral striatum. After motor recovery (symptomatic vs recovered state), only a significant increased in [^{18}F]Dopa uptake has been observed within the left limbic ACC (BA 25). The results also show a decrease of D2/D3 receptors BP within the anterior striatum and posterior putamen (no different with control state) as well as the limbic ACC. No significant change has been observed for the 5-HT system. The present PET imaging study highlights the role of the DA as well as the 5-HT system in the compensatory mechanisms. Nevertheless, our data suggest that the early asymptomatic stage and the motor recovery phase do not involve the same mechanisms. Indeed, the 5-HT system is involved only before motor symptoms appearance while the DA system plays a critical role in both asymptomatic phases but involving different regions. Of interest, both striatal and extrastriatal regions, pallidum and cingulate cortex in particular, are further evidenced in those mechanisms.

P2.090

Exposures to animal models of PTSD and drug addiction increase reactivity to drug/trauma cues and lead to similar noradrenergic behavioral sensitization: towards common physiological basis?

Toledano, D. (Orsay)[1], Gisquet-Verrier, P. (Orsay)[1]

[1]Université Paris Sud, Centre de Neurosciences Paris-Sud, UMR 8195, Orsay, France

Addiction and posttraumatic stress disorder (PTSD) are two pathologies characterized by their hypersensitivity to environmental stimuli related either to drug taking or to trauma. Exposure to these stimuli induces vivid re-experiencing effects that are responsible for the high rates of relapse characterizing these pathologies. Repeated exposures to psychostimulant in mice have been shown to induce a behavioral and a pharmacological sensitization which have been attributed to an uncoupling between the noradrenergic and the serotonergic systems (Lanteri et al. 2008; Salomon et al. 2006). Based on these results, we proposed that exposure to extreme stimuli (trauma or drug) induce uncoupling, leading to an hyper-reactivity of these systems in response of trauma/drug related cues accounting for their consequences on re-experiencing and relapse in these two pathologies. Here we show that rats exposed to an animal model of PTSD (the Single Prolonged Stress (SPS); Liberzon et al. 1997) or to four repeated administration of d-amphetamine (1mg/kg) exhibited a

behavioral sensitization in response to an acute amphetamine injection, modulated by individual differences in their reactivity to novelty (high and low responders, HR/LR), known as potential vulnerability trait for drug of abuse. Our result indicated that when delivered during a single session, the amphetamine injections reversed HR/LR phenotype, replicating results obtained after PTSD (Toledano et al., submitted). We further show that delivering the α2-receptor agonist, clonidine (20 µg/kg), during SPS prevented the behavioral sensitization, indicating an involvement of the noradrenergic system, already demonstrated in the case of repeated amphetamine injections (Doucet et al. 2013). Finally, we show that both animal models have behavioral consequences, affecting anxiety (light-dark box test), arousal (acoustic startle response) and reactivity to trauma/drug related cues.
The present results provide new evidence highlighting the similarities between PTSD and the dependence on drugs of abuse and strengthening our hypothesis of common physiological basis.

P2.091
Comparison of radiolabelled antagonists for serotonin 5-HT_6 receptor PET neuroimaging

Becker, G. (Lyon)[1,2], Colomb, J. (Lyon)[3], Billard, T. (Lyon)[3], Zimmer, L. (Lyon)[1,2]
[1]Lyon Neuroscience Research Center, University of Lyon & CNRS & INSERM, Lyon, France, [2]CERMEP-Imaging Platform, Hospices Civils de Lyon, Lyon, France, [3]Institute of Chemistry and Biochemistry (ICBMS), University of Lyon & CNRS, Lyon, France

Objectives: According to their brain localization, the serotonin 5-HT_6 receptors are potent therapeutic targets for psychiatric and neurological diseases, i.e., schizophrenia and Alzheimer's disease. However, the understanding of the serotonin 5-HT_6 pharmacology is currently partial and limited to animal models, particularly because of the lack of fluorinated PET 5-HT_6 radiopharmaceuticals. In this context, we developed several 5-HT_6 radioligands and evaluated their suitability for PET imaging.
Methods: Various quinoline and pyridine-based ligands have been synthesized, inspired by the known 5-HT_6 receptor pharmacophore. These non-radioactive fluorinated ligands and their radiolabelling's precursors were obtained through a divergent synthetic strategy. Starting from *bis*-functionalized heteroaromatic core, the targeting molecules were synthesized by using coupling reactions in 3 steps. Only ligands with the higher *in vitro* affinities toward 5-HT_6 receptors and the lower affinities toward 5-HT_{2A} receptors (a close pharmacophore) were radiolabeled via ^{18}F-nucleophilic aromatic substitution. These potential radiotracers were evaluated by *in vitro* autoradiography in rat brain. The most interesting radioligands of these were later evaluated by PETscans on anaesthetized rats and cats.
Results: Eight molecules were initially synthesised and three molecules with low *in vitro* affinity for 5-HT_6 receptors were removed (Ki >10 nM). The chemical and radiochemical purities of the five remaining fluorine radiotracers were > 99%, with a radiochemical yield of 5-45% (EOB) and specific activities in the range of 40-104 GBq/µmole (EOS). Four radiolabelled molecules presented a high *in vitro* binding in the striatum area rich in 5-HT_6 receptors and a dose-dependent displacement after SB258585 addition (a 5-HT_6 receptor antagonist). The following microPET studies showed that one among these four molecules had a good brain penetration and a striatal fixation. PETscans in cats gave additional information about its pharmacokinetic.
Conclusions: Our studies allowed us to select a radiotracer-candidate with suitable characteristics for PET imaging of 5-HT_6 receptors, justifying further evaluations in post-mortem human tissues and the development of chemical analogues.

P2.092
Clinatec: a translational technology research center

Berger, F. (Grenoble)[1]
[1]*INSERM-UJF-CHU-CEA, Grenoble, France*

A major biomedical need is supporting therapeutical innovation in the field of neurology. Brain inaccessibility and functionality probably explain the major delay observed in molecular annotation and therapy for brain diseases. Deciphering the mechanisms of inaccessible brain functional areas and treating them with non-lesional strategies locally addressing neuronal network dysfunction is mandatory. Deep brain stimulation is a perfect example of implanted technology acting locally in a reversible and functional way. It paved the way for more elaborated devices benefiting from the miniaturization and multifunctionality provided by the exponential development of micro-nano-technologies and electronics.
CLINATEC is a technology translational biomedical research center implanted inside the technology MINATEC campus. Its objective is to accelerate and make safer translation at the bedside of innovative micro-nano and electronics technologies. Five connected platforms are available, including:
1) a biomedical system integration facility,
2) a biocompatibility and translational biology platform,
3) a preclinical platform,
4) an imaging platform and at the end of the process,
5) a clinical unit.
The clinical unit is devoted to clinical technology proof of concept trials. It is supported by a unique high technology surgery room and multimodal monitoring associating intra-operative imaging, electrophysiology, behavior analysis and biomolecular explorations. This unique situation should favor an early synergistic connection between physicians, technologists , biologists and physicians from the clean rooms to the bedside.
Several projects have been already implemented. The Brain Computer Interface program is developing the first integrated ECoG implant for Brain computer-interface. Explorer program develops innovative micro-nano-biomarker-interfaces technologies for the deciphering of inaccessible human brain areas. The Near Infra Red program is developing an infra-red optical stimulator for neuroprotection in Parkinson disease. Innovative pump and catheter are also developed for brain local delivery of innovative therapies.
CLINATEC also provides a project hotel open for both the academic and industrial neuroscience community.

P2.093

Biochemical characterization of the cholesterol-binding domains of Alzheimer's β-amyloid peptide and a-synuclein

Di Scala, C. (Marseille)[1], Clément, L. (Marseille)[1], Garmy, N. (Marseille)[1], Fantini, J. (Marseille)[1], Chahinian, H. (Marseille)[1], Yahi, N. (Marseille)[1]
[1]*Aix Marseille Université, PPSN EA 4674, Service 331 IMSM, Marseille, France*

Alzheimer's β-amyloid (Aβ) peptides can self-organize into amyloid pores that may induce acute neurotoxic effects in brain cells. Membrane cholesterol, which regulates Aβ production and oligomerization, plays a key role in this process. Although several data suggested that cholesterol could bind to Aβ peptides, the molecular mechanisms underlying cholesterol/Aβ interactions are mostly unknown. On the basis of docking studies, we identified the linear fragment 22-35 of Aβ as a potential cholesterol-binding domain. This domain consists of an atypical concatenation of polar/apolar amino acid residues that was not previously found in cholesterol-binding motifs. Using the Langmuir film balance technique, we showed that synthetic peptides Aβ17-40 and Aβ22-35, but not Aβ1-16, could efficiently penetrate into cholesterol monolayers. The interaction between Aβ22-35 and cholesterol was fully saturable and lipid-specific. Single-point mutations of Val-24 and Lys-28 in Aβ22-35 prevented cholesterol binding, whereas mutations at residues 29, 33, and 34 had little to no effect. These data were consistent with the in silico identification of Val-24 and Lys-28 as critical residues for cholesterol binding. We conclude that the linear fragment 22-35 of Aβ is a functional cholesterol-binding domain that could promote the insertion of β-amyloid peptides or amyloid pore formation in

cholesterol-rich membrane domains. A similar mechanism is used by Parkinson's disease-associated a-synuclein whose cholesterol-binding domain has been identified as the tilted 67-78 fragment.
Keywords: Amyloid pore; cholesterol; cholesterol-binding domain; Alzheimer; Parkinson; lipid monolayer; tilted peptide.

P2.094

Organic cation transporter 2 controls the response to stress in the brain

Courousse, T. (Paris)[1], Bacq, A. (Paris)[1], Balasse, L. (Paris)[1], Louis, F. (Paris)[1], Le Guisquet, A.-M. (Tours)[2], Schinkel, A. (Amsterdam)[3], Giros, B. (Paris)[1], Belzung, C. (Tours)[2], Gautron, S. (Paris)[1]
[1]Université Pierre et Marie Curie, Physiopathologie des maladies du Système Nerveux Central UMR-S 952/UMR 7224, Paris, France, [2]Université François Rabelais, Imaging and Brain U 930, Tours, France, [3]The Netherlands Cancer Institute, Division of Molecular Biology, Amsterdam, Netherlands

Dysfunctions in hypothalamic-pituitary-adrenal (HPA) axis have been reported for several mental disorders, the most prevalent being major depressive disorder. Low-affinity organic cation transporters (OCT) are inhibited by corticosterone *in vitro* in cultured cells, suggesting that they might be inhibited by this hormone also *in vivo* in the brain. In this study we investigated the role of organic cation transporter 2 (OCT2) in the response to stress. We showed that OCT2 is expressed in stress-related circuits such as hippocampus, paraventricular nucleus of hypothalamus, pituitary and adrenals. OCT2-deficient mice show enhanced hormonal response in an acute stress paradigm, the forced-swim stress. To determine the consequences of increased corticosterone secretion during acute stress on sensitivity to chronic stress in OCT2-/- mice, the mice were exposed to the unpredictable chronic mild stress (UCMS) protocol. UCMS is an informative model to study stress-related disorders in animals, known to reproduce a depressive-like state that develops gradually in response to mild chronic socio-environmental stress. The 8-week UCMS regimen increased in OCT2-/- mice a gradual deterioration of the coat state, and decreased score in the nest building test, two parameters known to be very sensitive to stress. These results argue in favor of a higher sensitivity to stress in OCT2-/- mice. Western blot experiments revealed an alteration of mood-related intracellular signaling pathways in OCT2-/- mice brain at basal state and after UCMS, which could be responsible for their anomalies in mood-related behavior and response to stress. Our results altogether identify OCT2 as an important mediator of the response to stress, suggesting that deletion or inhibition of this transporter could enhance vulnerability to repeated adverse events, predisposing to depression.

P2.095

Impact of early environmental enrichment on excitation/inhibition imbalance and spontaneous epileptiform activity in Tg2576 (APPswe) mice

Bezzina, C. (Toulouse)[1], Juan, C. (Toulouse)[2], Krezymon, A. (Toulouse)[2], Halley, H. (Toulouse)[2], Lassale, J.M. (Toulouse)[2], Verret, L. (San Francisco)[3], Rampon, C. (Toulouse)[2], Dahan, L. (Toulouse)[2]
[1]Université Paul Sabatier, UMR 5169 Centre de Recherches sur la Cognition Animale, Toulouse, France, [2]UMR 5169 CNRS Centre de Recherches sur la Cognition Animale, Memoire Plasticité et Vieillissement, Toulouse, France, [3]Gladstone Institute of Neurological Disease, Department of Neurology, UCSF, San Francisco, United States

Within the last decade, it has been shown that transgenic mouse models of Alzheimer's disease, which exhibit high Aβ levels and deterioration of cognitive function, display spontaneous epileptiform activity (Palop et al., 2007; Minkeviciene et al., 2009) and aberrant gamma oscillatory activity (Verret et al., 2012). These observations prompted the field to re-examine the effects of abnormal patterns of network activity on cognitive dysfunction in AD.

We recently reported that Tg2576 (carrying APPswe) mice, a model of Alzheimer disease with progressive age-related cognitive decline, display abnormal expression of the neuropeptide Y (NPY) in their hippocampus at old age (13 months) which is highly suggestive of spontaneous seizure activity (Verret et al., 2013). These compensatory anatomical mechanisms are thought to damper network hyperactivity and may also interfere with normal neuronal and synaptic functions required for learning and memory. In the same study, we showed that transient and early exposure of female Tg2576 mice to environmental enrichment (when they are 3 to 6 month old) has long-lasting beneficial behavioral and neuropathological effects (Verret et al., 2013). Our aim now is to determine whether these protective effects of environmental enrichment on cognitive functions could be attributable to a reduced hippocampal excitation/inhibition imbalance and/or spontaneous epileptiform activity.
To address this question, we first examined excitation/inhibition imbalance by measuring the severity of seizures induced by the GABAa receptor antagonist pentylenetetrazole (PTZ). We show that young (6 month old) Tg2576 mice are more sensitive to PTZ than their non-transgenic littermates. This excitation/inhibition imbalance was not corrected by environmental enrichment.
We also noticed that a subpopulation of Tg2576 mice exhibit an ectopic expression of NPY in their dentate gyrus, which is detectable as early as 3 months of age, suggesting that at least some mice might present spontaneous epileptiform activity at the age when the early exposure to enriched environment takes place. Current work uses EEG recording before, during and after early environmental enrichment (3 to 5 months) in order to assess its impact on spontaneous epileptiform activity.

P2.096

Effects of an environmental enrichment in a mouse model of mental retardation

Lunion, S. (Orsay)[1], Desvignes, N. (Orsay)[1], Hanauer, A. (Illkirch Strasbourg)[2], Laroche, S. (Orsay)[1], Poirier, R. (Orsay)[1]
[1]Centre de Neurosciences Paris Sud, Orsay, France, [2]Institut de Génétique et de Biologie Moléculaire et Cellulaire, Illkirch Strasbourg, France

The Coffin-Lowry Syndrome is a rare syndromic form of X-linked mental retardation. This syndrome is caused by mutations of the *Rsk2* gene which encode for a kinase protein, RSK2, in the MAPK/ERK signaling pathway. The characterization of behavioural phenotype of RSK2 mutant mice mainly showed that they displayed delayed acquisition and long-term deficit in a spatial reference memory task, which suggests an alteration in hippocampal function. Recently, using delayed not matching to place (DNMP) radial arm maze task, we showed that RSK2 mutant mice are also deficient in pattern separation function, a form of dentate gyrus-dependent spatial learning and memory. Here, the main goal of our study was to assess the potential beneficial effect of environmental enrichment on spatial learning and memory performances in RSK2 mutant mice. Our data show that an environmental enrichment protocol of 3h per day during 24 days can rescue the spatial learning and memory performances of RSK2 mutant mice in Morris water maze task. According to several studies, environmental enrichment in rodents is associated with an enhancement of adult hippocampal neurogenesis, a form of plasticity that plays a significant role in hippocampal-dependent learning and memory. More, some studies showed an important role for newborn neurons in the adult dentate gyrus in pattern separation function. As this function is completely altered in RSK2 mutant mice, we studied hippocampal adult neurogenesis in these mice. In basal conditions, no deficit in proliferation, survival and maturation of newborn neurons was found in dentate gyrus of mutant mice. The survival of newborn neurons is also not affected in groups of mice submitted to DNMP radial arm maze task. However, we found a deficit in survival newborn cells in RSK2 mutant mice submitted to spatial learning and memory Morris water maze task. This deficit of adult neurogenesis in dentate gyrus is completely compensated by environmental enrichment.Together, our results suggest that RSK2 mutant mice seem to display a higher susceptibility to stress and that behavioural enrichment is an effective strategy to rescue the memory impairments and the survival of newborn DG neurons in this mouse model of the Coffin-Lowry mental retardation syndrome.

P2.097

Near infrared light therapy preserves midbrain dopaminergic cells and behavior from MPTP toxicity: evidence from two mouse strains

Torres, N. (Grenoble)[1], Moro, C. (Grenoble)[1], El Massri, N. (Sydney)[2], Ratel, D. (Grenoble)[1], Johnstone, D. (New South Wales)[3], Mitrofanis, J. (Sydney)[2], Benabid, A.-L. (Grenoble)[1]
[1]CEA, LETI, CLINATEC, Grenoble, France, [2]University of Sydney, Discipline of Anatomy & Histology, Sydney, Australia, [3]University of Sydney, Discipline of Physiology, New South Wales, Australia

Background: We have shown previously that near-infrared light (NIr) treatment or photobiomodulation neuroprotects dopaminergic cells in substantia nigra pars compacta (SNc) from degeneration induced by 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP), a well-known model for Parkinson's disease, in Balb/c albino mice. The present study explores whether NIr treatment offers neuroprotection to these cells in C57BL/6 pigmented mice also. In addition, we examine whether NIr influences behavioral activity in both strains of mice after MPTP treatment. We tested for various locomotive parameters in an open-field test, namely velocity, high mobility and immobility.
Results: Balb/c (albino) and C57BL/6 (pigmented) mice received injections of MPTP (total of 50mg/kg) or saline and NIr treatments (or not) over 48 hours. After each injection and/or NIr treatment, the locomotor activity of the mice was tested. After six days survival, brains were processed for TH (tyrosine hydroxylase) immunochemistry and the number of TH^+ cells in the substantia nigra pars compacta (SNc) was estimated using stereology. Results showed higher numbers of TH^+ cells in the MPTP-NIr groups of both strains, compared to the MPTP groups, with greater protection in the Balb/c mice (30% vs 20%). The behavioral tests revealed strain differences also. For Balb/c mice, the MPTP-NIr group showed greater preservation of locomotor activity than the MPTP group. Behavioral preservation was less evident in the C57BL/6 strain however, with little effect of NIr in the MPTP-treated cases of this strain.
Conclusions: In summary, our results revealed the neuroprotective benefits of NIr treatment after neurotoxic insult at both cellular and behavioral levels and suggest that Balb/c strain might provide a more sensitive model of protection than the C57BL/6 strain.

P2.098

Microvascular plasticity after cerebral ischemia: time window for angiogenic therapies?

Remy, C. (Grenoble)[1], Moisan, A. (Grenoble)[1,2], Favre, I. (Grenoble)[1,3], Rome, C. (Grenoble)[4], Grillon, E. (Grenoble)[5], Naegele, B. (Grenoble)[3], Barbier, E. (Grenoble)[1], Detante, O. (Grenoble)[1,3]
[1]Grenoble Institut des Neurosciences, Inserm U 836 - UJF - CEA - CHU, Grenoble, France, [2]Unité mixte de thérapie cellulaire, EFS-UJF-CHU, Grenoble, France, [3]Unité Neuro-Vacsulaire - Neurologie, CHU Grenoble, Grenoble, France, [4]Institut Albert Bonniot, Inserm U 823 - UJF, Grenoble, France, [5]IRMaGe, US 017 - UMS CNRS 3552 - UJF - CHU, Grenoble, France

Stroke, leading cause of disability, lacks delayed treatment to enhance recovery. Its complex physiopathology needs to be elucidated in order to accelerate development of efficient therapies. This study describes the time-course of the microvascular plasticity after stroke and identifies an optimal time-window for angiogenic therapies.
Methods: Two groups of rats were followed during 25 days. One group underwent a transient middle cerebral artery occlusion (MCAo, n=8); the second one underwent the surgery without occlusion (Sham, n=9). Anatomic and functional characteristics of microvasculature were assessed using MRI: vascular permeability, cerebral blood volume (CBV), vessel size index (VSI, mean microvessel diameter) and vascular density. Functional recovery was assessed with modified Neurological Severity Score (mNSS) and adhesive removal test on the same animals. Angiogenic factors were quantified by RT-qPCR of brain samples of additional MCAo rats.
Results: MCAo animals revealed poor neurological scores compared to shams. VSI was higher in MCAo group with a maximal value at day 3 (D3) and a decrease afterwards. Vascular density crashed

in MCAo group at D3 and increased until D25 without reaching sham values at D25. CBV was higher in MCAo group, remaining stable from D3 to D16 with a decrease at D25. Many angiogenic factors showed a bell-shape evolution curve during 25 days with a maximum at the acute stage (eNOs and VEGFR-2), at the subacute stage (TGFβ1 and Ang1) or between both stages (Tie1, SDF-1 and CXCR-4). Ang2, VEGF and VEGFR-1 run a biphasic evolution during 25 days with a drop at D3.
Conclusions: The acute stage occurs from D1 to D3 and is characterized by high levels of Ang-2, VEGFR-2 and eNOs that altogether cause a deleterious BBB permeability and vasodilation. The transition stage takes place from D3 to D7 and involves TGFβ1, Ang1, Tie1, SDF-1 and CXCR-4. It consists in preparing a favorable microenvironment to initiate angiogenesis. The subacute stage occurs between D7 and D25 and is characterized by high levels of Ang1, Ang2, VEGF, VEGFR-1 and TGFβ1 promoting stabilization of vessels and partial restoration of vasculature. Both transition and subacute stages may represent an optimal time-window for therapies aiming to promote angiogenesis.

P2.099
State dependency and cortical excitability (TMS)

Beynel, L. (Grenoble)[1], Bécu, M. (Grenoble)[1], Chauvin, A. (Grenoble)[1], Guyader, N. (Grenoble)[2], Harquel, S. (Grenoble)[1,3], Marendaz, C. (Grenoble)[1]
[1]Laboratoire de Psychologie et Neurocognition, CNRS UMR 5105, Grenoble, France, [2]GIPSA-Lab, Grenoble, France, [3]UMS "IRMaGe" CNRS UMS 017, Grenoble, France

Introduction: Cortical excitability (CE) refers to the neural response to magnetic stimulation. CE is generally assessed on the motor cortex using transcranial magnetic stimulation (TMS). Paired-pulse TMS separated by various interstimuli-interval allow an indirect measure of neurotransmission (GABA inhibition and Glutamate facilitation). State dependency (SD) is a broad concept which integrates neuronal and physical-mental states, level of fatigue, alertness, motivation, etc. Recent studies showed that SD could dramatically modify TMS effects during perceptive tasks (Silvanto et al., 2008). Is SD also able to modify the CE? It is an important issue as CE is often used as a biomarker of various pathologies (as mood disorder). To answer this question, different CE measures were estimated according to two mental state conditions: resting state (without task) *vs.* active state (with a cognitive task).
Method: Motor cortex of 20 healthy subjects was stimulated and motor evoked potentials (MEPs) were collected on the contralateral first dorsal interosseus muscle. TMS procedure of CE included two conditions: “single pulse” condition (baseline) and “paired-pulse” condition to estimate GABA inhibition and Glutamate facilitation. Subjects' CE was estimated with and without cognitive task. The task was an easy working memory task: two different crosses, a white (25% of trials) or a grey cross (75%), could be displayed on a screen. Subjects were asked to imagine a mental calendar and add a day each time the white cross appeared. MEPs' amplitudes were analyzed offline, thanks to a toolbox (CortExTool) developed in our lab, and normalized as a function of baseline.
Results: We found a significant increase of the baseline in the task condition and an interaction between paired-pulse conditions and the task: GABA inhibition was decreased and glutamate facilitation was increased in the task condition.
Discussion: SD, here a slight cognitive load, dramatically acts on intracortical neurotransmission. CE is commonly used as a biomarker of various pathologies which imply GABA/glutamate dysfunctions. This study highlights the necessity to control subjects' mental state during CE measures.

P2.100
Analysis of the role of the PINK1/Parkin pathway in mitophagy by new quantitative methods

Jacoupy, M. (Paris)[1], Bertolin, G. (Paris)[1], Gautier, C. (Paris)[1], Ferrando-Miguel, R. (Paris)[1], Brice, A. (Paris)[1], Corti, O. (Paris)[1]
[1]*Institut du Cerveau et de la Moelle Épinière - Cricm - Neb, Hôpital Pitié-Salpêtrière, Paris, France*

Mutations in the genes encoding Parkin and PINK1 cause autosomal recessive forms of Parkinson's disease (PD). These proteins act in a pathway central for maintenance of mitochondrial quality: upon loss of the mitochondrial membrane potential (DY_{mit}) triggered by the protonophore CCCP, they cooperate to promote clearance of dysfunctional mitochondria by the ubiquitin-proteasome and autophagy pathways (mitophagy).
Our aim is to identify new actors of the PINK1/Parkin pathway involved in mitochondrial quality control (cf. abstract by Bertolin *et al*.). Mitochondrial loss promoted by PINK1 and Parkin is commonly evaluated by the quantification of the proportion of cells negative for markers of the different mitochondrial compartments. This method is subjective, time-consuming and accounts solely for the total elimination of mitochondria from cells. There is therefore great need to develop alternative methods for the sensitive, objective and quantitative estimation of the loss of mitochondrial material from individual cells, possibly amenable to large-scale analyses.
We explored the potential of the mitochondrial mass markers MitoTracker Green (MTG) and 10-Nonyl Acridine Orange (NAO) coupled with flow cytometry imaging. HEK293 cells, containing detectable amounts of PINK1 and Parkin, were chosen as a model to study mitochondrial clearance triggered by CCCP before or after siRNA-mediated downregulation of each of these proteins. The mitochondrial incorporation of MTG and NAO was sensitive to alterations in DY_{mit} in this paradigm, preventing the reliable detection of mitochondrial loss promoted by the PINK1/Parkin pathway. We therefore turned to a second method based on the quantitative analysis of individual mitochondrial proteins with different submitochondrial localization by immunofluorescent staining and confocal microscopy. This method, currently under investigation with a classical confocal microscope, will be adapted for quantitative cellular imaging using automated fluorescence microscopy.
The development of quantitative methods to monitor mitophagy will facilitate analysis of the molecular mechanisms underlying the mitochondrial quality control functions of the PINK1/Parkin pathway and their role in the pathophysiology of autosomal recessive PD.

P2.101

Immunisation against tPA/NMDA interaction provides therapeutic effects in a model of multiple sclerosis

Docagne, F. (Caen)[1], Gauberti, M. (Caen)[1], Montagne, A. (Caen)[1], Bronsard, J. (Caen)[1], Haelewyn, B. (Caen)[2], Defer, G. (Caen)[1,3], Vivien, D. (Caen)[1], Macrez, R. (Caen)[1]
[1]*Inserm U 919, Caen, France,* [2]*Centre Universitaire de Ressources Biologiques (CURB), Caen, France,* [3]*University Hospital of Caen, Caen, France*

The aim of this work was to assess the therapeutic potential of a strategy of immunisation targeting NMDA receptors in experimental autoimmune encephalomyelitis (EAE), an animal model for multiple sclerosis (MS).
Results from animal models suggest that N-Methyl-D-Asparte (NMDA) receptors are involved in the pathology of EAE (Paul & Bolton, 2002). Also the serine protease tissue plasminogen activator (tPA) participates in the pathology of EAE (Lu et al., 2002). Various studies indicate that tPA interacts with NMDA receptors (NMDAr), which results in excitotoxic neuronal death (Nicole et al., 2001), and monocyte recruitment across the blood brain barrier (Reijerkerk et al., 2010).
We thus hypothesized that blocking the interaction between tPA and NMDA receptors could result in a beneficial effect in EAE. For that, we used a strategy of immunisation that we previously reported to provide neuroprotection in animal models of stroke (Macrez et al., 2011).
We show here that the active immunization aganist tPA/NMADr interaction decreases inflammation in the brain, the cerebellum and the spinal cord and improves the neurological score by inhibiting the progression of EAE compared to control mice.
Our data show benefit of a strategy of active vaccination to prevent tPA/NMDAr interaction in an EAE model, reducing neurological deficits and inflammatory processes. So we report a proof of concept

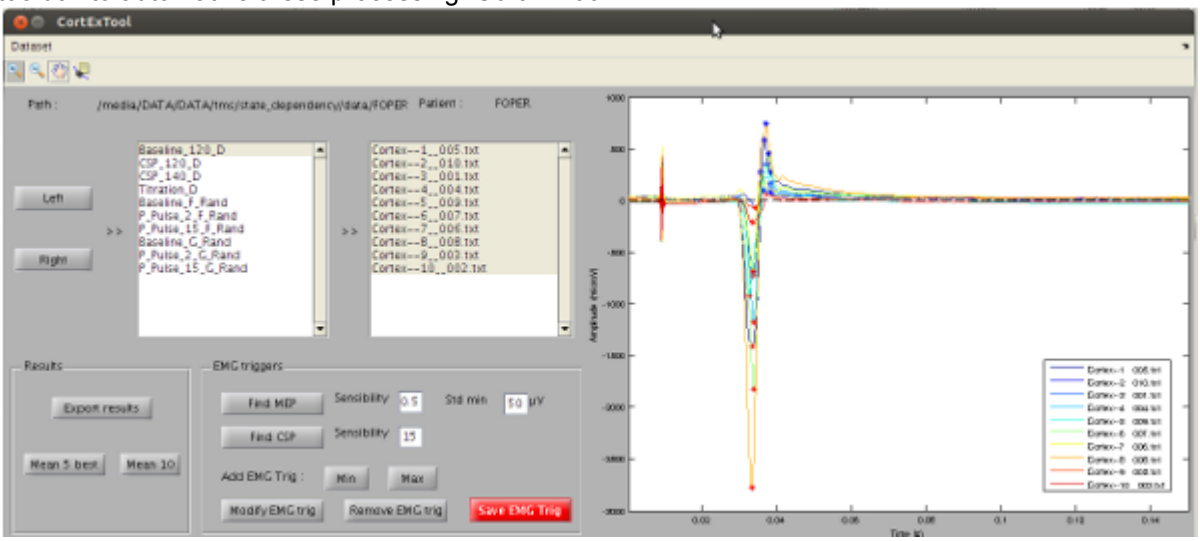
showing that the tPA/NMDAr interaction is a potential target for the development of a new therapeutic modality in MS.

P2.103

CortExTool: a signal processing toolbox for cortical excitability by transcranial magnetic stimulation

Harquel, S. (Grenoble)[1,2], Becu, M. (Grenoble)[1], Beynel, L. (Grenoble)[1], Chauvin, A. (Grenoble)[1], Guyader, N. (Grenoble)[3], Marendaz, C. (Grenoble)[1]
[1]Laboratoire de Psychologie et de NeuroCognition - UMR 5105, Grenoble, France, [2]IRMaGe - UMS 017, CHU Grenoble - CS 10217, Grenoble, France, [3]GIPSA-lab, Grenoble, France

Measuring the cortical excitability (CE) is a significant tool in clinical research. Measures are done through the electromyographic (EMG) recordings of the motor phenomena induced by transcranial magnetic stimulation (TMS). The two main indicators used in such studies are the motor evoked potential (MEP) amplitude (in V), and the cortical silent period (CSP) duration (in s). Usually, these data are manually extracted, which is time-consuming and operator dependent. We developed a toolbox to automatize these processing: CortExTool.



[CortExTool main display]

CortExTool is implemented in Matlab programming langage. It is able to manage data exported from the Dantec Keypoint system. Data are automatically re-arranged in a classified database. The toolbox provides users with an user-friendly interface in order to quickly display and classify EMG signals. A large variety of outputs can be extracted, such as amplitude, power, latency, duration, etc.
The main feature of the toolbox is its automatic detection of MEP and CSP onsets and characteristics. MEP detection is based on the correlation curve between the signal and a EMG template, corresponding to a typical gaussian-shaped MEP. First, the maximum correlation time point is used to define MEP coarse latency. MEP maximum and minimum points are then refined using local maxima detection on the 50-600Hz filtered signal curve. A standard deviation curve obtained by a 10ms moving window allows CSP detection. The minimum Min thus corresponds to the rough latency of muscular tonus cancellation. Precise time boundaries of the CSP are then fixed as to be the first samples to be superior to Sens*Min, Sens being a tunable parameter.
Future improvements of the toolbox will be : managing other data formats, defining markers of PEM quality, statistical analysis on group studies.

P2.104

Ataxin-7 plays role in differentiation of photoreceptors and cerebellar neurons

Yanicostas, C. (Paris)[1], Hibi, M. (Kobe)[2], Brice, A. (Paris)[3], Stevanin, G. (Paris)[4], Soussi-Yanicostas, N. (Paris)[5]
[1]Hôpital Robert Debré, Université Paris Diderot, Inserm U 676, Paris, France, [2]Laboratory for vertebrate axis formation, RIKEN Center for Developmental Biology, Kobe, Japan, [3]CRICM UMR_S INSERM 975, CNRS 7225, UPMC Univ, Paris, France, [4]CRICM UMR_S INSERM 975, CNRS 7225, UPMC Univ, Ecole Pratique des Hautes Etudes, Paris, France, [5]Hôpital Robert Debré, Inserm U 676, Univ Paris-Diderot, Zebrafish Models of Neurodegenerative Diseases, Paris, France

The expansion of a polyglutamine (polyQ) tract in the N-terminal region of ataxin-7 (atxn7) is the causative event in spinocerebellar ataxia type 7 (SCA7), an autosomal dominant neurodegenerative disorder mainly characterized by progressive, selective loss of rod-cone photoreceptors and cerebellar Purkinje and granule cells. The molecular and cellular processes underlying this restricted neuronal vulnerability, which contrasts with the broad expression pattern of atxn7, remains one of the most enigmatic features of SCA7, and more generally of all polyQ disorders. To gain insight into this specific neuronal vulnerability and achieve a better understanding of atxn7 function, we carried out a functional analysis of this protein in the teleost fish *Danio rerio*. We characterized the zebrafish *atxn7* gene and its transcription pattern, and by making use of morpholino-oligonucleotide-mediated gene inactivation, we analysed the phenotypes induced following mild or severe zebrafish atxn7 depletion. Severe or nearly complete zebrafish atxn7 loss-of-function markedly impaired embryonic development, leading to both early embryonic lethality and severely deformed embryos. More importantly, in relation to SCA7, moderate depletion of the protein specifically, albeit partially, prevented the differentiation of both retina photoreceptors and cerebellar Purkinje and granule cells. In addition, [1-232] human atxn7 fragment rescued these phenotypes showing strong function conservation of this protein through evolution. The specific requirement for zebrafish atxn7 in the proper differentiation of cerebellar neurons provides, to our knowledge, the first *in vivo* evidence of a direct functional relationship between atxn7 and the differentiation of Purkinje and granule cells, the most crucial neurons affected in SCA7 and most other polyQ-mediated SCAs. These findings further suggest that altered protein function may play a role in the pathophysiology of the disease, an important step toward the development of future therapeutic strategies.

P2.105

Instability and modeling of giant axonal neuropathy-associated mutations in the BTB-Kelch protein gigaxonin

Boizot, A. (Montpellier)[1], Peyressatre, M. (Montpellier)[1], Kuntz, N. (Chicago)[2], Stojkovic, T. (Paris)[3], Halbert, C. (Marseille)[4], Chabrol, B. (Marseille)[4], Houlden, H. (London)[5], Sproule, D. (New York)[6], Schulman, B. (Memphis)[7], Rautenstrauss, B. (München)[8], Bomont, P. (Montpellier)[1]
[1]INSERM U 1051 - INM, Montpellier, France, [2]Northwestern Feinberg School of Medicine, Chicago, United States, [3]Institut de Myologie, Paris, France, [4]Hôpital de la Timone, Marseille, France, [5]UCL Institue of Neurology, London, United Kingdom, [6]Spinal Muscular Atrophy Clinical Research Center, New York, United States, [7]St. Jude Children's Research Hospital, Memphis, United States, [8]Fachhumangenetiker, Neurogenetik, München, Germany

Gigaxonin plays a key role in sustaining neuron survival and cytoskeleton architecture. Indeed, recessive mutations in the gigaxonin encoding gene cause a fatale neurodegenerative disorder called Giant Axonal Neuropathy (GAN) that impacts broadly the nervous system and induces a wide disorganization of the Intermediate Filament (IF) network. Gigaxonin is a BTB-Kelch protein predominantly and equally expressed throughout the nervous system but at very low level. It has been proposed as a substrate adaptor of Cul3-mediated E3 ubiquitin ligases. To decipher the pathophysiological mechanisms caused by mutations in gigaxonin and define how this relates to E3 ligase activity, we generate the first structural model of the normal protein. The mapping of patient´s

mutations on the structure, together with the assessment of mRNA levels and the effects of mutations on protein stability reveal that both mRNA and protein instability are disease mechanisms causal for GAN. Additionally, we demonstrate that regardless of the mutations or the severity of the disease, gigaxonin´s abundance is univocally and severely reduced in all GAN patients. Moreover, determining gigaxonin´s levels prove to be of better diagnostic value than the clinical description, the nerve biopsy and the genetic screening in identifying new GAN cases among at set of individuals exhibiting neuropathies of unknown etiology.

P2.106

Expectation induced by a chronic cocaine treatment: a behavioral and neurochemical study in rats

Geoffroy, H. (Paris)[1], Puig, S. (Paris)[1], Noble, F. (Paris)[1], Benturquia, N. (Paris)[1]
[1]*Université Paris Descartes, Laboratoire de Neuropsychopharmacologie des addictions, Paris, France*

Cocaine dependence is a significant public health issue, characterized by periods of abstinence. Chronic exposure to drugs of abuse induces important modifications on neuronal systems, including the dopaminergic system. The aim of our study was to investigate and compare the spontaneous behavioural and neurochemical consequences of 14-days chronic cocaine pretreatment regimen (20 mg/kg, intraperitoneal, once daily at 10.00 AM), which are extensively used in laboratories to mimic pattern of cocaine abuse in humans. Extracellular DA levels were evaluated 1 (Withdrawal Day 1 or WD1) and 14 (WD14) days after the last cocaine injection. In parallel to this, locomotor activity was measured during 24 h on WD1 and WD14. These experiments were performed either exactly at the hour at which rats were habituated to receive a cocaine injection or at another hour of the day. We showed that the cocaine treatment led to a spontaneous increase in DA levels in the nucleus accumbens (Nac) (WD1/WD14), and in locomotor activity (WD1) when measured at the exact hour of injection. We unambiguously demonstrated that chronic cocaine treatment induced drug expectation associated with higher basal DA level in the Nac when measured at the time of usual injection. These results underline that taking into consideration the hour of the day at which experiments are performed is crucial while exploring behavioural and neurochemical alterations in rats.

P2.107

Early disruption of adult neurogenesis precedes age-dependent network remodeling in the hippocampal dentate gyrus of Tg2576 mouse model of Alzheimer's disease

Rampon, C. (Toulouse)[1], Krezymon, A. (Toulouse)[1], Richetin, K. (Toulouse)[1], Halley, H. (Toulouse)[1], Roybon, L. (Lund)[2], Lassalle, J.-M. (Toulouse)[1], Frances, B. (Toulouse)[1], Verret, L. (Toulouse)[1]
[1]*CRCA - Université Paul Sabatier, CNRS, Toulouse, France,* [2]*Lund University, Lund, Sweden*

Characterization of the timing and nature of brain dysfunction is crucial to understand the mechanisms leading to cognitive deficits in Alzheimer's disease (AD). Using the well-described Tg2576 (APPswe) mouse model of Alzheimer´s disease that develops progressive age-dependent amyloidosis and cognitive deficits, we examined whether specific stages of the disease progression were associated with the expression of anatomical markers of hippocampal dysfunction. We found that Tg2576 mice develop a complex pattern of age-dependent anatomo-functional changes in their dentate gyrus. This includes age-related aberrant expression of neuropeptide Y and reduced levels of calbindin, reflecting a profound remodeling of inhibitory and excitatory circuits in the dentate gyrus. Moreover, we identified severe alterations of adult hippocampal neurogenesis in Tg2576 mice at young age. We gathered converging data indicating that in Tg2576 mice, the maturation of new neurons is altered, suggesting that their functional integration into hippocampal circuits may be compromised. Thus, disruption of

adult hippocampal neurogenesis seems to precede profound network remodeling in Tg2576 mice and therefore, may account as an early event in the etiology of Alzheimer´s disease. Ultimately, both events may constitute key components of hippocampal network dysfunction and associated cognitive deficits in these mice. Financial support: France Alzheimer association, ANR JCJC and MALZ, the Singer-Polignac Foundation, CNRS, Toulouse University.

P2.108

Repeated amphetamine impairs α_{2A}-Adrenergic autoreceptors in the long run

Bobadilla Asensio, A.C. (Paris)[1], Doucet, E.L. (Paris)[1], Godeheu, G. (Paris)[1], Lanteri, C. (Paris)[1], Lanfumey, L. (Paris)[2], Tassin, J.-P. (Paris)[1]
[1]Univ. Pierre et Marie Curie, INSERM UMRS 952, CNRS UMR 7224-PMSNC, Neurobiologie de l'addiction, Paris, France, [2]CPN - UMRS 894, Physiopathologie des troubles de l'humeur, dépression et addiction, Paris, France

In rodents, repeated amphetamine triggers behavioural sensitization, an increase of drug-induced locomotor response that stays constant for several weeks. This phenomenon is shared by all drugs of abuse, suggesting the occurrence of long-term neuronal changes.
Previous work has shown that a neurochemical sensitization of noradrenergic and serotonergic neurons develops in parallel with the amphetamine-induced behavioural sensitization (1). We made the hypothesis that this long-lasting hyperreactivity could be sustained by a decreased inhibition of both systems. Actually, noradrenergic firing activities are in part controlled by α_{2A}-Adrenergic (α_{2A}-AR) inhibitory autoreceptors. We therefore focused on the role and possible modifications of α_{2A}-AR following repeated amphetamine injections.
First of all, an injection of efaroxan, a α_{2A}-AR antagonist, potentiates the amphetamine-induced noradrenaline (NA) release in the prefrontal cortex (PFC) of naïve mice, showing that the blockade of α_{2A}-AR autoreceptors induces the same hyperreactivity than that found in sensitized animals.
Second, we measured the capacity of clonidine, a α_{2A}-AR agonist, to induce a decrease of NA release in the PFC. After both a 4-day and a 1-month withdrawal, clonidine was no longer able to induce a decrease of NA levels in sensitized animals, suggesting a desensitization of α_{2A}-AR.
Since autoradiographic studies showed that the expression of α_{2A}-AR was unchanged in amphetamine sensitized animals, we then looked for a decrease in autoreceptors-coupled Gαi proteins and found a long-term decreased expression of Gαi1 and Gαi2 proteins in sensitized animals.
Finally, we showed that stimulation of α_{2A}-AR by clonidine completely prevented amphetamine-induced behavioural sensitization, while blocking these autoreceptors during the injections facilitated it.
Altogether, these results highlight the critical role of α_{2A}-AR in the induction and maintenance of amphetamine-induced behavioural sensitization and suggest that inhibitory noradrenergic feed-back is an indirect target of amphetamine.
(1) Salomon L, Lanteri C, Glowinski J, Tassin JP. Behavioral sensitization to amphetamine results from an uncoupling between noradrenergic and serotonergic neurons. (2006) *P.N.A.S. (USA) (Track II)* 103: 3476-3481.

P2.109

Brain state-dependent functional reconfiguration of entorhinal cortex networks in control and epileptic rats

Bernard, C. (Marseille)[1], Quilichini, P. (Marseille)[1]
[1]Institut de Neurosciences des Systèmes, INSERM UMRS_1106, Marseille, France

Purpose: In order to support the production of different cognitive functions, neuronal networks can support the emergence/expression of different oscillatory activities associated to different brain states. One possible mechanism at the core of such versatile function is that a given network could dynamically change its functional structure, e.g. the expression of connections between neurons, as a function of the brain state.

We investigated whether such a mechanism is present in the temporal lobe, which networks are known to play an important role notably in episodic memory and navigation.
Methods: We used *in vivo* high-density recordings (32-sites silicon probe) in entorhinal cortex to isolate the spike trains of numerous neurons and reconstructed their functional connectivity matrix during theta and slow oscillations in anesthetized control and epileptic (pilocarpine model of temporal lobe epilepsy) rats.
Results: We show that entorhinal cortex networks can be dynamically reconfigured in a brain state-dependent fashion in control animals. This is also of particular importance in the context of epilepsy, in which temporal lobe circuits are deeply anatomically reorganized. We show that the firing properties of principal cells and interneurons are unevenly altered through the different layers. Besides, we show a functional hyper connectivity in the entorhinal cortex in layer 2, where some neurons bearing “hub” properties seem to appear, whereas deeper layers are affected by hypo connectivity. Moreover, we investigated how such restructured networks are dynamically reconfigured and it appears that the brain state-dependent reconfiguration of the functional connectivity is lost in layer 3.
Conclusion: Such *in vivo* functional correlates to the structural reorganization of temporal lobe networks could be have a key role in neuronal synchronization and generation of seizures, but also in the emergence of the associated cognitive deficits, like an impairment of episodic memory.

P2.110
Random delay boosts musical fine motor recovery after stroke

van Vugt, F.T. (Hannover)[1,2], Kafczyk, T. (Hannover)[1], Kuhn, W. (Hannover)[1], Rollnik, J. (Hessisch Oldendorf)[3], Tillmann, B. (Lyon)[2], Altenmüller, E. (Hannover)[1]
[1]Institute of Music Physiology and Musicians' Medicine, University of Music, Drama and Media, Hannover, Germany, [2]Université Claude Bernard Lyon 1, Lyon Neuroscience Research Center, Auditory Cognition and Psychoacoustics Team CNRS-UMR 5292, INSERM U 1028, Lyon, France, [3]BDH-Klinik, Hessisch Oldendorf, Germany

Motor impairments are among the most common and most disabling results of stroke worldwide. Previous studies have revealed that learning to play the piano helps to improve motor functioning in standardised motor tasks.
One hypothesis about the effectiveness of music is that it provides the brain a time-locked auditory feedback (a musical tone) with each movement (keypress). In our study, we provide the first direct test of this rehabilitation mechanism. We hypothesise that a keyboard emitting randomly delayed tones fails to contain this temporal information about the movement and therefore the timecourse of rehabilitation should be affected.
37 patients in early stroke rehabilitation with moderate motor impairment and no previous musical background learned to play the piano using simple finger exercises using only 5 tones and familiar children´s songs. The participants were assigned randomly to one of two groups: in the *normal* group, the piano played the tone immediately at keypress, in the *delay* group, the tone occured 100-600ms after keypress. To assess recovery, we performed standard clinical tests such as the nine-hole-pegboard test and finger tapping rate and regularity. Furthermore, we used the Profile of Mood States (POMS) to establish motivational changes. Finally, we assessed whether patients were aware of the delay through a keystroke-sound sensitivity thresholds, auditory temporal thresholds and qualitative questionnaires.
Surprisingly, patients in the *delay* group improved strikingly in the nine-hole-pegboard test, whereas patients in the *normal* group did not. In finger tapping rate and regularity both groups showed similar marked improvements. The *normal* group showed reduced depression whereas the *delay* group did not. Remaining data analyses are currently in progress.
Music therapy on a randomly delayed keyboard can significantly boost motor recovery after stroke. We hypothesize that patients in the delay group may be prompted to update their internal model every time the auditory feedback did not match their temporal expectations. In this way, enhanced plasticity might be elicited. Furthermore, patients may have widened their action-perception integration window as surprisingly they did not consciously detect the sound manipulations.

P2.111

Extensive pharmacological validation of the delay discounting task in T-maze in juvenile rats as a predictive preclinical model for novel ADHD treatment

David, S. (Orleans)[1], Fery, N. (Orleans)[1], Silva, M. (Orleans)[1], Massé, F. (Orleans)[1], Trovero, F. (Orleans)[1], Bizot, J.-C. (Orleans)[1]
[1]*KEY-OBS SAS, Orleans, France*

Impulsivity is a symptom of several disorders, such as Attention Deficit/Hyperactivity Disorder (ADHD), pathological gambling or kleptomania and is frequently associated with suicide and aggressive behaviors.
Previous experiments have demonstrated that various drugs used in ADHD treatment, such as psychostimulants and NA uptake inhibitors improved impulsivity in juvenile rats submitted to a delayed discounting task (DDT) in T-maze (Bizot et al, 2007, 2011). By using this experimental paradigm the present study had three main objectives:
1- Given that stimulant drugs are anorectic the first aim of the current study was to examine whether the effects of methylphenidate result from a reduced food motivation.
2- To evaluate the effects of a chronic treatment with methylphenidate.
3- To extensively investigate the effects of other drugs used in various psychiatric indications.
All the experiments were performed in food restricted male Wistar rats, 30-35 day-old at the moment of the test. Food restricted animals were placed in individual cages with free access to food to measure food motivation. Methylphenidate (3 mg/kg, IP) did not induce anorectic effect that could interfere with the experimental procedure.
A chronic treatment with methylphenidate (3 and 6 mg/kg, PO) increased the number of choices for the large-but delayed reward, i.e. improved impulsivity.
Acute treatments with paroxetine (5, 10 mg/kg, IP) or with clonidine (0.03 and 0.05 mg/kg, IP) reduced impulsivity whilst acute treatments with haloperidol (0.01 and 0.3 mg/kg, IP) did not have any effect.
Given that paroxetine reduced impulsivity in patients, that clonidine is a potentially effective ADHD treatment and that haloperidol is an antipsychotic presumably devoid of effect on impulsivity, this study provided an extensive new validation for the use of the DDT experimental paradigm for the study of efficacy of drugs on impulsivity after acute or chronic treatments.

P2.112

Dissecting the function of the dorsal striatonigral pathway by targeted-genetic ablation

Jaouen, F. (Marseille)[1], Révy, D. (Marseille)[1], Salin, P. (Marseille)[1], Melon, C. (Marseille)[1], Tafi, E. (Rome)[2], Amalric, M. (Marseille)[3], Kerkerian, L. (Marseille)[1], Marie, H. (Rome)[2], Beurrier, C. (Marseille)[1]
[1]*Aix-Marseille Université, CNRS UMR 7288, Marseille, France,* [2]*EBRI, Rome, Italy,* [3]*Aix-Marseille Université, CNRS UMR 6155, Marseille, France*

The basal ganglia (BG) are key neural substrates that control motor and reward-associated behaviors. Their dysfunction is associated with several diseases including Parkinson's disease, schizophrenia and drug addiction. As the main BG input, the striatum is thought to be an important site for mediating many of the maladaptive processes responsible for these devastating neurological disorders. The direct (striatonigral) and indirect (striatopallidal) pathways by which the striatum controls the BG output are thought to have opposing but balancing roles on BG output and behaviors. However, to what extend does this concept apply to pathological states or more complex behaviors is still a challenging and debated issue. To decipher the specific role of the direct pathway, we developed a transgenic

mouse model enabling inducible ablation of striatonigral medium spiny neurons (MSNs) by expressing the human *diphtheria toxin receptor* (DTR) gene under the control of the *Slc35d3* promoter, a gene enriched in these neurons. Intrastriatal diphtheria toxin (DT) injection eliminated the majority of striatonigral MSNs without affecting the other striatal populations, except significantly reducing the number of cholinergic interneurons. This loss is not due to a direct-killing effect of DT as cholinergic interneurons did not express DTR; the cholinergic insult being significantly dampened by injection of a neurokinin-1 receptor agonist, it is likely the consequence of substance P depletion, a neuropeptide released by striatonigral MSNs. DT-injected transgenic mice showed dramatic reduction of L-DOPA-induced dyskinesias in parkinsonian condition. They were more sensitive to an acute injection of cocaine but showed less sensitization after repeated administration of the drug and an inability to acquire contextual cocaine-associated stimuli. Finally, DT injection reduced anxiety as evaluated in the open field and elevated plus maze tests. Our results reveal a pro-survival function of striatonigral MSNs for cholinergic interneurons and complex involvement of these populations in heterogeneous functions, ranging from motor, addictive or emotional behaviors.
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P2.113

Frataxin depletion in zebrafish embryo, toward a novel animal model for Friedreich ataxia

Soussi-Yanicostas, N. (Paris)[1], Rustin, P. (Paris)[1], Yanicostas, C. (Paris)[1]
[1]*INSERM U 676, Université Paris VII, Hôpital Robert Debré, Paris, France*

Friedreich ataxia (FRDA), the most common form of hereditary ataxia is an autosomal recessive neurodegenerative disorder affecting the central and peripheral nervous system. FRDA patients also frequently display cardiomyopathy. FRDA is caused by a trinucleotide (GAA) repeat expansion in the first intron of the nuclear *frataxin* (*FRX*) gene leading to markedly reduced synthesis of *FRX* mRNA and thus, reduced accumulation of the encoded mitochondria targeted protein, frataxin. Although several animal models of the disease have provided insights into the physiological role of frataxin as well as its requirement for the disease process, the precise role of the protein remains largely unknown.
A faithful FRDA animal model must display reduced, albeit not full, frataxin depletion to reproduce the situation observed in patients (5 to 15% compared to wild-type levels) and avoid the embryonic lethality induced by full depletion of the protein. Here we report immunocytochemical and biochemical analyses of zebrafish embryos showing severe frataxin depletion as the result of morpholino-oligonucleotide-mediated depletion of the zebrafish *FRX* gene.
We observed that while complete, or nearly complete, frataxin depletion impairs development and embryo viability, as previously described for other frataxin deficient animal models, severe, albeit not full, frataxin inactivation affects several organs or tissues, including the ear, and of particular interest, spinal motor neuron axons, cerebellum and heart. These embryos also show motility defects, as the likely result of abnormal motorneuron axon growth and guidance and decreased number of cerebellar Purkinje and granule cells. We will alo described the mitochondrial respiration and oxydative stress response in these zebrafish embryos.

References:

- Yanicostas, Barbieri, Hibi , Brice , Stevanin, Soussi-Yanicostas . Requirement for zebrafish ataxin-7 in differentiation of photoreceptors and cerebellar neurons.
PLoS One. 2012;7(11).
- Martin E, Yanicostas C, Rastetter A, Alavi Naini SM, Maouedj A, Kabashi E, Rivaud-Péchoux S, Brice A, Stevanin G, Soussi-Yanicostas N. Spatacsin and spastizin act in the same pathway required for proper spinal motor neuron axon outgrowth in zebrafish. Neurobiol Dis. 2012 48(3) 299-308.

P2.114

Small molecules decrease pathological tau hyperphosphorylation in zebrafish model of tauopathy

Alavi, M. (Paris)[1], Yanicostas, C. (Paris)[1], Schmid, B. (Munich)[2], Paquet, D. (Munich)[2], Haass, C. (Munich)[2], Jeffrey, E. (San Diego)[3], Soussi-Yanicostas, N. (Paris)[1]
[1]Inserm, U 676 - Hôpital Robert Debré, Université Denis Diderot (P7), Paris, France, [2]DZNE, Munich, Germany, [3]University of California (UCSD), San Diego, United States

Hyperphosphorylated isoforms of the microtubule-associated protein Tau (hpTau) are the major proteinaceous components of the paired helical and straight filaments, which define a major neuropathological feature of tauopathies, a family of neurodegenerative diseases comprising Alzheimer's disease (AD). Small molecules capable of mitigating the accumulation of hpTau, represent interesting candidates for drug therapy of these disorders. Here, we made use of a transgenic zebrafish line, the Tg[Huc::DsRed; hTauP301L], which recapitulates key pathological features of tauopathies, including accumulation and aggregation of hyperphosphorylated Tau and neuronal death. We took advantage of this line to screen for molecules inducing a decreased accumulation of hpTau and we identified a small molecule and a number of its derivatives, which induce a marked decrease in accumulation of hpTau in Tg[Huc::TauP301L] embryos. Our data strongly suggest that these compounds could represent an efficient therapeutical strategy for the treatment of tauopathies, including AD.
Keywords: Tauopathy, Alzheimer's disease, Zebrafish, Small molecules, Tau hyperphosphorylation
References:
- Yanicostas C, Barbieri E, Hibi M, Brice A, Stevanin G, Soussi-Yanicostas N. Requirement for zebrafish ataxin-7 in differentiation of photoreceptors and cerebellar neurons. PLoS One. 2012; 7(11).
- Martin E, Yanicostas C, Rastetter A, Alavi Naini SM, Maouedj A, Kabashi E, Rivaud-Péchoux S, Brice A, Stevanin G, Soussi-Yanicostas N. Spatacsin and spastizin act in the same pathway required for proper spinal motor neuron axon outgrowth in zebrafish. Neurobiol Dis. 2012 Dec; 48(3):299-308.
- Ayari B, Landoulsi A, Soussi-Yanicostas N. Localization and characterization of kal 1.a and kal 1.b in the brain of adult zebrafish (Danio rerio). Brain Res Bull. 2012 Jul 1; 88(4):345-53.
- Ayari B, El Hachimi KH, Yanicostas C, Landoulsi A, Soussi-Yanicostas N. Prokineticin 2 expression is associated with neural repair of injured adult zebrafish telencephalon.J Neurotrauma. 2010 May; 27(5):959-72.

P2.115

Deleterious effect of delayed hepatic tPA clearance on ischemic stroke and thrombolysis

Rubio, M. (Caen)[1], Lemarchand, E. (Caen)[1], Gauberti, M. (Caen)[1], Montagne, A. (Caen)[1], Répessé, Y. (Caen)[1], Villain, H. (Caen)[1], Vivien, D. (Caen)[1], Ali, C. (Caen)[1]
[1]Inserm U 919, Caen, France

Introduction: The consequences of chronic alcohol consumption on ischemic stroke damages and recombinant tissue Plasminogen Activator (rtPA)-induced thrombolysis (the only available acute treatment for ischemic stroke) are poorly documented.
Methods: In this study, we have first determined the progression of ischemic lesions and the benefit/risk ratio of thrombolysis after chronic alcohol consumption (10% alcohol in the drinking water during 6 weeks), using a murine model of thromboembolic stroke.
Results: Final brain lesion volumes were significantly higher in chronic alcohol-exposed mice than in control mice (drinking only water). While rtPA was equally efficient to induce thrombolysis in both groups, its beneficial effects on the extent of brain damages disappeared after chronic alcohol consumption. rtPA was found to remain longer in the bloodstream of chronic alcohol-exposed mice than in control mice. Consistently, the experimental increase in plasmatic tPA levels by hydrodynamic transfection of mice aggravated ischemic lesions. By two-photon microscopy and immunohistology of the liver, we have observed a delay in rtPA clearance by the liver. Also, near-infrared fluorescence imaging (NIRF) revealed an inversion in the uptake pattern of rtPA between the liver and the brain of

chronically alcoholised mice. These effects can be explained by a selective reduction in the levels of Low-density lipoprotein Receptor-related Protein 1 in the liver (LRP-1; the main receptor of transcytosis of tPA).
Conclusion: Together, these data suggest that after chronic alcohol consumption, the unbalanced rtPA recapture by the liver could increase the passage of rtPA into the brain and, due to the neurotoxicity of rtPA in the cerebral parenchyma, could be responsible for the loss of beneficial effect of thrombolysis. Thus, we propose here a critical role of the liver-brain axis in the consequences of ischemic stroke and thrombolysis.

P2.116

Functional cerebellar phenotypes are induced by overexpression of Pcp4/Pep19 in TgPCP4, a model of Down syndrome

Collin, T. (Paris)[1]
[1]*Université Paris Diderot, Paris, France*

The gene encoding Purkinje Cell Protein 4 (PCP4; a modulator of Ca^{2+}-CaM) is present in three copies in Down syndrome (DS). In the adult mouse cerebellum, PCP4 is expressed in Purkinje cells and molecular layer interneurons. To evaluate the consequences of 3 copies of this gene, we constructed a transgenic (TgPCP4) mouse model bearing one extra copy of human *PCP4,* whose properties were analyzed in parallel with those of an established mouse model of DS :Ts1Cje. Overall, the overexpression of pcp4 had a similar impact in both models (Mouton-Liger 2011). Transcriptome and proteome analysis of cerebellar specific markers towards development brought evidence for an increase in markers of maturation such as GADs, and a modulation of CaMKIIs at adult stage. No alteration in cerebellar volume was observed. At P14, PCP4-overexpression provokes a partial inhibition of the Depolarization-induced Suppression of Inhibition (DSI) phenomenon, which is known to be correlated to intracellular calcium dynamics. In adult TgPCP4, a pairing-procedure involving a somatic depolarization coupled to PF stimulation induced a clear-cut LTD of PF-EPSCs in cells tested (n=4), with a mean reduction in amplitude of synaptic responses of 69 +/- 8 % 30 min after the end of the second pairing. This LTD appears to be at least two orders of magnitude stronger that what is observed in WT animals (27 +/- 6 %, n=4) which is in keeping with previously reported results using the same paradigm (23 +/- 5 %; Crépel, 2009). Behavioral assessments at adult stage, showed strong deficits in the Rotarod and Treadmill paradigms indicating that PCP4-overexpression affects the motor circuitry probably through the Purkinje cells, sole output of the cerebellum. Many of the daily challenges faced by persons with DS are caused by difficulties in their perceptual-motor behavior. This murine model is likely to be of premium interest to test therapies towards the motor coordination impairment and the psychomotor developmental alterations observed in DS.

P2.117

Optogenetic control of cholinergic interneurons: implication for Parkinson's disease

Beurrier, C. (Marseille)[1], Liberge, M. (Marseille)[2], Maurice, N. (Marseille)[1], Jaouen, F. (Marseille)[1], Camon, J. (Marseille)[2], Ztaou, S. (Marseille)[2], Amalric, M. (Marseille)[2], Kerkerian-Le Goff, L. (Marseille)[1]
[1]*Aix-Marseille Université/CNRS UMR 7288, Marseille, France,* [2]*Aix-Marseille Université/CNRS UMR 6155, Marseille, France*

Striatal acetylcholine interneurons (ACh) are key players in the induction of motor learning and motor dysfunction in neurodegenerative diseases. In Parkinson's disease (PD), ACh are potent modulators

of medium spiny neurons excitability. We assessed the contribution of ACh interneurons in the basal ganglia network in normal and pathophysiological conditions, by an optogenetic approach. To specifically express the opsins in striatal Ach interneurons, we stereotaxically injected into the striatum a Cre-inducible adeno-associated virus (AAV) vector carrying the gene encoding channelrhodopsin (ChR2) or halorhodopsin (eNpHR) in transgenic mice expressing Cre-recombinase under the choline acetyltransferase promoter.
The effect of laser illumination of ACh interneurons was measured on neuronal activity and behaviour. Electrophysiological recordings of ACh interneurons in brain slice and *in vivo* showed that both opsins were functional: ChR2 drove spike activity while eNpHR silenced firing. Using a behavioural approach, we demonstrated the critical contribution of ACh interneurons in two experimental models of PD. In a pharmacological model (haloperidol-induced catalepsy), optogenetic inhibition of ACh interneurons reduced the akinesia induced by haloperidol, while their activation had no effect. This antiparkinsonian benefit was also confirmed in the lesional PD model (unilateral 6-OHDA nigrostriatal lesion). Inhibition of Ach interneurons led to a reduction of turning bias and balance deficit in the cross maze and narrow beam tests, respectively. We are currently analyzing *in vivo* in anaesthetized mice the impact of striatal ACh interneurons modulation on the basal ganglia network.
These results emphasize the critical involvement of striatal ACh interneurons in the imbalanced MSNs activity and motor deficits in PD.
This work is supported by ANR, France Parkinson, CNRS and AMU.

P2.118

Serotonin 4 receptors are involved in addiction to cocaine and anorexia: from gene to behaviors

Boussadia, B. (Montpellier)[1,2], Laurent, L. (Montpellier)[1], Marin, A. (Gignac)[3], Compan, V. (Nîmes)[1,2]
[1]Institut de Génomique Fonctionelle, Montpellier, France, [2]Université de Nîmes, Nîmes, France, [3]Contralco, Gignac, France

Addiction may result from abnormal decision-making, as this mental disease is repeatedly excessive. We postulate that transition from the abusive consumption of food or cocaine towards excessive restrictive diet, until death depends, at least for a part, on the change of activity of a serotonin sub-type (R5-HT4) from an inactive state (R) towards an active form (R*) in the nucleus accumbens (NAc) that is a crucial part of the reward system.
We previously found that stimulating R5-HT4 in the NAc increases the activity of one addiction-related pathway (cAMP/PKA/CART) to inhibit hunger. The inactive form (R) in the NAc may inhibit this pathway wherefrom food abuse while the absence of R5-HT4 may favor cocaine abuse. In this context, the objective is to identify new molecular pathway of addiction upon the R5-HT4 control. Based on our preliminary results obtained using proteom analyses (“iTRAQ”), we set out to further develop an innovative biotech tool to diagnostic dependence (confidential / industrial partnership). We have already identified at least two specific and novel biomarkers related to the consumption of drugs of abuse, additionally controlled by the R5-HT4. Changes in the levels of these both biomarkers following administration of the drug have been validated using the western blot technique.

P2.119

Elucidation of astrocytic glucocorticoid receptor actions during degeneration of midbrain dopamine neurons using mice conditionally inactivated for glucocorticoid receptors in parenchymal astrocytes

Maatouk, L. (Paris)[1], Carrillo- de Sauvage, M.A. (Paris)[1], Hunot, S. (Paris)[2], Hirsch, E. (Paris)[2], Pfrieger, F. (Strasbourg)[3], Tronche, F. (Paris)[1], Vyas, S. (Paris)[1]

[1]CNRS UMR 7224, INSERM U 952, UPMC, Physiopathologie des Maladies du Système Nerveux Central, Paris, France, [2]CNRS UMR 7225, CRICM, INSERM UMR_S 975, UPMC, Paris, France, [3]UMR 7168/LC2 CNRS/Université Louis Pasteur, INCI, Strasbourg, France

Astrocytes are implicated in physiopathology of Parkinson's disease (PD), which is characterized by loss of dopaminergic neurons (DN) of the substantia nigra (SN) and presence of aggregated α-synuclein in neurons. Transcription factors such as NRF2, nurr1 (nuclear receptor) expressed in astrocytes have been reported to regulate DN degeneration triggered by α-synuclein toxicity. In the experimental MPTP neurotoxic model of PD, we showed in mice lacking glucocorticoid receptor (GR, also a nuclear receptor) in microglia, an increase in microglial activation that augments astroglial reactivity (PNAS, 2011,108:6632-7) suggesting cross talk between astroglia and microglia during DN degeneration. To understand the actions of GR in astrocytes, we generated mice in which GR gene can be conditionally deleted in astrocytes by CreER2/loxp system. Connexin-30CreER2 mice were crossed with GRloxp/loxp mice to obtain $GR^{connexin30\text{-}CreER2}$ mutant mice. To verify for cre recombinant activity we crossed connexin30-creER2 mice with a reporter mTomato/mGFP mouse line. After tamoxifen injections, in the mutant but not control mice GFP+ cells were observed throughout brain parenchyma. The expression of cre in brain regions including SN and striatum was also verified by immunohistochemistry.
In an acute paradigm of MPTP intoxication, mice deprived of astrocytic GR showed increased loss of DNs in the SN. Analysis of glial reactivity in absence of astrocytic GR revealed enhanced microglial number and hypertrophy whereas astrocyte reactivity remained unchanged. To understand which processes are regulated by GR, expression of genes known for astrocyte functions such as inflammation, exocytosis, regulation of glutamate levels, was analyzed following acute MPTP treatment. Among the genes tested MCP-1, TNF-alpha and complexin2 mRNA levels were significantly but transiently increased in controls and mutants. ICAM-1 (inter-cellular adhesion molecule-1) transcripts were specifically increased in mutant compared to control mice, suggesting that astrocytic GR regulates the inflammatory process via ICAM-1, which acts in the recruitment of immune cells. Overall our data suggest that during DN degeneration, astrocytic GR exerts a non cell-autonomous action involving control of microglial reactivity.

P2.120

Gamma response in the ageing brain

Bompas, A. (Bron)[1,2], Tales, A. (Swansea)[3], Bayer, T. (Cardiff)[4], Singh, K.D. (Cardiff)[2]
[1]Inserm U1028, Centre de Recherche en Neurosciences de Lyon, DYCOG, Bron, France, [2]Cardiff University, CUBRIC - School of Psychology, Cardiff, United Kingdom, [3]Swansea University, Psychology Department, Swansea, United Kingdom, [4]Cardiff University, Department of Geriatric Medicine, Llandough Hospital, Cardiff, United Kingdom

Recent work has shown that the frequency of occipital sustained gamma response to a visual grating in MEG reduces between age 8 and 46 (Gaetz, Roberts, Singh & Muthukumaraswamy (2012). Functional and structural correlates of the aging brain: relating visual cortex (V1) gamma band responses to age-related structural change, HBM 33:2035-2046). The present work extends the age range to include a healthy elderly cohort aged 67 to 84, as well as six aged matched early demented patients, four diagnosed with Alzheimer's disease (AD) and two with vascular cognitive impairment (VCI). First, we report that the frequency of sustained visual gamma in the elderly cohort is only slightly lower than that observed for participants in their forties, thus suggesting that gamma frequency reaches a plateau around 40Hz rather than linearly decreasing again after 50 years old. Second, although the amplitude of the initial broadband gamma response (60-100Hz) reduces with age, we note that the broadband gamma response in 2 AD and 2 VCI patients was much higher than for aged-matched participants. We also assessed volumetric parameters of the occipital lobes using MRI and confirm that V1 cortical thickness and surface correlate negatively with age and gamma amplitude, and positively with gamma frequency. However, in agreement with Gaetz et al. 2012, the relationship between anatomical structure and gamma response is all mediated by age, suggesting that gamma response and cortical atrophy are two distinct signatures of the ageing brain.

P2.121

Role of the TREK background potassium channels in nociception

Christin, M. (Valbonne)[1,2], Pereira, V. (Clermont Ferrand)[3], Devilliers, M. (Clermont Ferrand)[3], Legha, W. (Clermont Ferrand)[3], Alloui, A. (Clermont Ferrand)[3], Lazdunski, M. (Valbonne)[1], Busserolles, J. (Clermont Ferrand)[3], Eschalier, A. (Clermont Ferrand)[3], Noël, J. (Valbonne)[1,2]
[1]Institut de Pharmacologie Moléculaire et Cellulaire, CNRS/Université de Nice-Sophia Antipolis UMR 7275, Valbonne, France, [2]LabEx Ion Channel Science and Therapeutics, Valbonne, France, [3]INSERM/Université d'Auvergne UMR 766, Pharmacologie fondamentale et clinique de la douleur, Clermont Ferrand, France

The perception of noxious stimuli is essential to an organism survival as it allows the appropriate avoidance response to potentially harmful situations. This detection occurs at the peripheral terminals of specialized sensory neurons - nociceptors. These neurons of small diameter transduce stimuli of a thermal, mechanical or chemical nature into action potentials and transmit this information to the spinal-cord. The stimulation of nociceptors at the periphery by the different stimuli relies on the expression profile of specific ion channel transducers at the plasma membrane of the axon terminals. The nociceptive system is characterized by a high degree of plasticity which is exacerbated in patho-physiological conditions.
We study the role in nociception of background K^+ channels with two-pore domains (K2p) belonging to the TREK channels subfamily. The K_{2p} channels generate background K^+ currents that play a major role in neuronal excitability and cell firing. The TREK channels subfamily is composed of TREK1, TREK2 and TRAAK. They are mechano- and thermo-activated channels that have been involved in anesthesia, depression and neuroprotection. We have previously demonstrated the role of TREK1 and TRAAK channels in polymodal pain perception (Alloui et al., 2006; Noel et al., 2009). They are involved in mechanical pain as well as in heat and cold perception. They prevent nociceptive fibers from firing at moderate temperature by opposing the depolarization resulting from the gating of excitatory channels by temperature. Whilst TREK2 is the major background K^+ current in dorsal root ganglion neurons and shares many regulations and functional properties with TREK1 and TRAAK, its role in nociception still remains unknown. The aim of our work is to investigate the role of TREK2 in nociception in physiological and patho-physiological conditions using knock-out mice. We assessed the impact of this channel on nociceptors with complementary *in vitro,* ex *vivo* (nerve-skin recordings), and pain behavior tests. We show that each member of the TREK channels subfamily contributes to thermal perception in different temperature ranges and that they are involved in neuropathic and inflammatory pain.
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P2.122

Lamina specific noradrenergic control of inhibitory synaptic transmission in the dorsal horn of the rat spinal cord

Seibt, F. (Strasbourg)[1], Schlichter, R. (Strasbourg)[1]
[1]UPR 3212 CNRS, Université de Strasbourg, Strasbourg, France

The dorsal horn of the spinal cord (DH) is a major site for the integration and the modulation of nociceptive information. Supraspinal neurons may inhibit or facilitate spinal nociceptive transmission by releasing various neuromodulators. Noradrenaline (NA) has analgesic effects and it has been shown that the activation of descending noradrenergic fibers inhibits nociceptive transmission at the spinal level. However, the detailed mechanism by which these descending noradrenergic pathways

exert such analgesic effects remains largely unclear. The analgesic effect of NA can be reproduced by intrathecal injections of alpha2 adrenergic receptor agonists, but it appears that the stimulation of inhibitory transmission in the superficial DH involves only alpha1 adrenoreceptors.
In order to clarify the respective roles of alpha1 and alpha2 adrenoreceptors, we have studied the consequences of their activation on inhibitory synaptic transmission in lamina II and laminae III-IV of the rat DH.
Application of NA (20µM) increased the frequency of sIPSCs in a reproducible and reversible manner in 95% of neurons in laminae III-IV and in 100% of neurons in lamina II of the DH. The effect of NA was reproduced by the alpha1 adrenoreceptor agonist phenylephrine (20µM) and largely inhibited by the general alpha adrenoreceptor antagonist phentolamine (10µM) in lamina II as well as in the laminae III-IV. Interestingly the alpha2 agonist clonidine (10µM) increased the frequency of sIPSCs in a subset of neurons in laminae III-IV but had no effect on inhibitory transmission in lamina II. In the presence of tetrodotoxin, NA increased the frequency of mIPSCs in 50% of the lamina II neurons but had no effect in laminae III-IV.
These results indicate that noradrenaline has differential effects on inhibitory synaptic transmission in lamina II and laminae III-IV of the DH. Interestingly, our result show that alpha2 adrenoreceptors, that are classically known to inhibit synaptic transmission and neurotransmitter release, can stimulate inhibitory synaptic transmission in the deep but not superficial DH.

P2.123

Potentiation of the anti-hyperalgesic effect of amitriptyline by co-treatment with the connexin inhibitor THN01 in sciatic nerve-ligated rats

Jeanson, T. (Fontenay-aux-Roses)[1,2], Oliveira Figueiredo, A.C. (Paris)[2], Picoli, C. (Fontenay-aux-Roses)[1], Mouthon, F. (Fontenay-aux-Roses)[1], Charvériat, M. (Fontenay-aux-Roses)[1], Bourgoin, S. (Paris)[2], Hamon, M. (Paris)[2]
[1]*Theranexus, CEA, 92260, Fontenay-aux-Roses, France,* [2]*INSERM U 894 CPN UPMC, Paris, France*

Current treatments of neuropathic pain, notably with some antidepressants, have a limited efficacy and are endowed with poorly tolerated side effects. Because recent studies pointed out the implication of glial connexins in the neuroinflammatory/sensitizing mechanisms underlying neuropathic pain, we investigated whether these gap junction proteins could be relevant targets for innovative neuropathic pain treatments. Flow cytometry parachute assays allowed identification of connexins inhibitors that we tested in combination with antidepressants to alleviate hyperalgesia in a rat model of neuropathic pain. We describe here the data obtained with the connexin inhibitor THN01 combined with amitriptyline on both mechanical hyperalgesia and associated neuroinflammatory processes.
Adult male Sprague-Dawley rats underwent a chronic unilateral constriction injury (CCI) by ligation of the sciatic nerve. Two weeks after surgery, rats were treated for 14 days with either amitriptyline (12 mg/kg/day, through a s.c. implanted osmotic minipump), THN01 (0.5 mg/kg i.p. twice daily at 9 a.m. and 6 p.m.) or the combination of both drugs. Control CCI rats received only the vehicle (0.9 % NaCl) under the very same conditions. The paw pressure Randall & Selitto test was applied to assess treatments-induced changes in mechanical hyperalgesia and qRT-PCR quantification of mRNAs encoding various markers (ATF3, Ox42, IL1β IL6, BDNF, NR2B) was carried out in dorsal root ganglia and spinal cord.
CCI-induced decrease in pressure threshold value to elicit nocifensive response (vocalization) progressively reversed during amitriptyline treatment, confirming the anti-hyperalgesic action of this antidepressant. Although the connexin inhibitor THN01 was inactive on its own, it significantly enhanced (+40%) amitriptyline-induced reduction of mechanical hyperalgesia. However, neuroinflammatory reaction markers did not differ in the various treatment groups. These data support the idea that connexin inhibition could be a promising approach toward improving current neuropathic pain therapy. The unchanged expression of neuroinflammatory/sensitization markers suggests that the anti-hyperalgesic effect of combined amitriptyline+THN01 treatment involved mechanisms downstream of these processes.

P2.124

ω3 PUFA status influences the age-related hippocampal alterations of glutamatergic neurotransmission in rats

Denis, I. (Jouy-en-Josas)[1], Latour, A. (Jouy-en-Josas)[1], Potier, B. (Paris)[2], Champeil-Potokar, G. (Jouy-en-Josas)[1], Hennebelle, M. (Jouy-en-Josas)[1], Maximin, E. (Jouy-en-Josas)[1], Langelier, B. (Jouy-en-Josas)[1], Gisquet, P. (Orsay)[3], Balasse, L. (Orsay)[3], Lavialle, M. (Jouy-en-Josas)[1], Billard, J.-M. (Paris)[2], Heberden, C. (Jouy-en-Josas)[1], Vancassel, S. (Jouy-en-Josas)[1], Neuromega3 Collaboration

[1]INRA, Nurelice, Jouy-en-Josas, France, [2]INSERM, Centre de psychiatrie et neurosciences, Paris, France, [3]CNRS, Centre de Neurosciences Paris-Sud, Orsay, France

Objectives: A poor ω3 polyunsaturated fatty acids (ω3 PUFA) status, favored by the low ω3/ω6 ratio in western diets, seems to contribute to cognitive decline in the elderly, but mechanistic evidence is lacking. We therefore explored the impact of ω3 status on the evolution of glutamatergic transmission and neurogenesis in the hippocampus during ageing in rats. These processes are involved in memory formation and their dysregulation participates to the age-related brain damage leading to cognitive decline.
Methodologies: We have compared 6 groups of rats aged 6 to 22 months fed ω3-deficient, ω3/ω6-balanced, or ω3 (fish oil) supplemented diets: Young ω3 Balanced (YB), Deficient (YD) or Supplemented (YS), and Old ω3 Balanced (OB), Deficient (OD) or Supplemented (OS) rats. We have evaluated synaptic efficacy and plasticity (electrophysiological recording), astroglial regulations (glutamate uptake and GFAP expression), neuronal markers (glutamate transporters and receptors), neurogenesis (proliferation of neuronal precursors in the sub granular zone), cognitive abilities (Barnes maze and Openfield) and analyzed brain fatty acids composition.
Results: Dietary modulation of ω3 intakes efficiently modified the incorporation of docosahexaenoic acid (DHA, the main ω3 in cell membranes) in brain (-50% deficient vs balanced, +10% supplemented vs balanced). Ageing induced a 35% reduction of synaptic efficacy due to decreased pre-synaptic glutamate release, and a 30% decrease in the astroglial glutamate uptake associated to a marked astrogliosis (+100% GFAP). ω3 deficiency further decreased these hallmarks of ageing (OD vs OB rats: -35% synaptic efficacy, -15% glutamate uptake, +30% GFAP). On the opposite, ω3 supplementation increased synaptic efficacy (+25% OS vs OD) and seems to abolish astrogliosis (OS vs YS : no change in GFAP). Neurogenesis was altered by ω3 deficiency but not by supplementation. Behavioural tests showed some increased effects of age in deficient rats and attenuated effects in supplemented ones.
Conclusion: Our results characterize some specific age-related alterations of the glutamatergic synapse in the hippocampus that are aggravated by a dietary deficit in ω3 and attenuated by ω3 supplementation.

P2.125

Explicit memory creation during sleep: a causal role of place cell on navigation

DeLavilleon, G. (Paris)[1,2], Lacroix, M. (Paris)[1,2], Rondi-Reig, L. (Paris)[2], Benchenane, K. (Paris)[1,2]

[1]Ecole Supérieure de Physique et de Chimie Industrielles, CNRS UMR 7637 - Lab. de Neurobiologie, Paris, France, [2]Laboratoire de neurobiologie des processus adaptatifs, UMR 7102, UPMC, Paris, France

It is now widely accepted that sleep is important for the consolidation of preexisting memory traces. Here we show that a place preference task can be learned during sleep without prior waking experience. We design a protocol to trigger during sleep, intracranial rewarding stimulations by the

action potentials of a unique hippocampal place cell. At awakening animals went and stayed within the associated place field. These results show that it is possible to create an artificial explicit memory during sleep that is used during subsequent waking period to drive a goal directed behavior. Moreover, it demonstrates the causal role of place cells on the mental representation of space as hippocampal cell assemblies still thus conveyed the same spatial information during sleep and wakefulness.

P2.126

Complementary and synergistic control of wakefulness by orexins and histamine, demonstrated using a double knockout mouse model

Lin, J.-S. (Lyon)[1], Anaclet, C. (Lyon)[1], Ouk, K. (Lyon)[1], Akaoka, H. (Lyon)[1], Buda, C. (Lyon)[1], Perier, M. (Lyon)[1], Ohtsu, H. (Sendai)[2], Yanagisawa, M. (Dallas)[3], Franco, P. (Lyon)[1]
[1]CRNL, INSERM U1028-CNRS UMR5292, Univ. Cl. Bernard Lyon1, Physiologie intégrée du système d'éveil, Lyon, France, [2]Tohoku University, Department of Cellular Pharmacology, Sendai, Japan, [3]University of Texas, Howard Hughes Medical Institute, Dallas, United States

We have previously shown that mice lacking histamine (HA, hdc KO) are characterized by an EEG and behavioral somnolence. Indeed, they show a deficit of wakefulness (W) when high vigilance is required such as lights-off or a new environment; whereas orexin (Ox)-KO mice are distinguished by a W deficit facing a motor challenge. We then suggested that HA and Ox exert a distinct but complementary control of W. To access the synergies of the two waking systems, we have characterized the sleep-wake phenotypes of double KO mice lacking HA and Ox, which were obtained by crossing hdc and Ox KO mice. The mouse model was validated by PCR and immunohistochemistry showing the deletion of hdc and prepro-orexin genes and absence of HA and Ox cells in the posterior hypothalamus. Cortical EEG and sleep-wake monitoring was performed in adult mice under baseline conditions and following behavioral or/and pharmacological tests. We found that double KO mice were characterized, on the one hand, by a somnolence severer than that seen with hdc or Ox single KO mice, i.e., 1) significant decrease in W (during darkness and over 24h) and in sleep latencies after behavioural tests; 2) unable to stay awake facing a new environment. This somnolence was abolished by modafinil but not by H3-receptor inverse agonists suggesting functional HA loss. On the other hand, these mice showed phenotypes characteristic of Ox KO mice, i.e., direct REM sleep onset (DREMs) and W deficit facing a motor challenge, both being rescued by central Ox-A dosing. Finally, the double KO mice displayed aggravated sleep fragmentation, obesity and phenotypes never seen with simple KO mice, e.g., EEG hypersynchronisation and cataplexy (sudden loss of muscle tone during W, characteristic of human narcolepsy). Our data suggest that HA and Ox exert a distinct but complementary and synergistic control on W, the amine being mainly responsible for cortical EEG arousal and cognitive activities during W and the neuropeptide being more involved in the behavioural activities during W. They could be co-responsible for narcolepsy: Ox deficiency is likely the direct cause of DREMs and cataplexy, whereas a decreased HA neurotransmission could account for the excessive somnolence seen in this disease and other sleep disorders.

P2.127

Long-term electrophysiological survey of cochlear implanted patients: laterality of language function and auditory maturation

Laboissière, R. (Lyon)[1], Mazzuca, M. (Lyon)[1], Arias-Rojas, V. (Lyon)[1], Thai-Van, H. (Lyon)[1], Truy, E. (Lyon)[1], Collet, L. (Lyon)[1]
[1]Centre de Recherche en Neurosciences de Lyon, Équipe DyCog, Lyon, France

We present the analysis of a large data set collected on cochlear implanted patients enrolled at the University Hospital Edouard Herriot in Lyon, France. The analysis on a reduced fraction of this data set has already been published elsewhere (Thai-Van et al., 2007). This retrospective analysis includes 232 patients (112 females and 120 males) all implanted with a Nucleus (Cochlear, Melbourne, Australia) device. Electrically evoked compound action potentials measures were collected from one month after the surgery until 14 years and a total of over 13,000 independent measures have been analysed. The latency of the the responses of the brain stem nuclei (waves II and V) were modelled using a back-fitting procedure to a mixed-effects linear model. We could find evidence for the following effects:
(1) a difference in the response of electrodes #5 and #20, related to the anatomical position in the cochlear modiolus;
(2) a latency difference between males and females, that correlates with anthropometric data;
(3) a fast maturational rate followed by a standing plateau and a later increase of the latencies along the duration of cochlear implant use;
(4) a lack of later increase in the latency time with cochlear implant duration use when the interval III-V is considered; and
(5) a difference in the behavior of right versus left implanted ears that interacts with the age at implantation.
This later effect, confirmed in a subset of the data, where patients were selected to form matched groups in age and ear side, gives strong evidence for laterality effects on the maturational aspects of the language function.

P2.128

Probing the neural basis of trimodal representation of self-body motion: an fMRI study

Kavounoudias, A. (Marseille)[1], Blanchard, C. (Marseille)[1], Nazarian, B. (Marseille)[2]
[1]Lab de Neurosciences Intégratives et Adaptatives, CNRS / Aix-Marseille université, Marseille, France, [2]Centre d'IRM fonctionnelle - INT (U1106), La Timone, Marseille, France

To perceive one's own body movements, the CNS uses multiple sensory information derived from several modalities including vision, touch, and muscle proprioception. All these sources of information have to be efficiently merged together to form a coherent percept. The aim of this study was to determine where multisensory integration processing takes place in the human brain through functional magnetic resonance imaging (fMRI) experiments.
For this purpose, illusory sensations of clockwise rotations of the right hand were induced by stimulating three sensory channels either separately or simultaneously: *Muscle Proprioception* was activated using a pneumatic vibrator applied to the *pollicis longus* muscle; *Touch* was activated by an amagnetic disk scrolling under the subject's hand, and *Vision* was stimulated using a movie of a background scene rotating under the subject's hand. Outside the scanner, the kinesthetic illusions were copied by the subjects with their left hand and the motor responses in wrist muscles were recorded.
Psychophysical and electromyographic data recorded on 19 participants show that similar perceptual and motor responses were found during the three unimodal stimulation and that bi- and tri-modal stimulation improved the resulting kinesthetic perceptions. Group analyses performed on fMRI recordings show that although differences occurred in the sensorimotor networks associated with a kinesthetic illusion depending on its sensory origin, overlapping activations can be evidenced across the 3 unimodal conditions. In addition, brain activations during bi- or tri-modal stimulation further show that multisensory networks do not result from a mere summation of the corresponding unisensory networks and that heteromodal brain regions seem specifically dedicated to multisensory integration; However, they also support the recent assumption that earlier multisensory processing might occur in primary sensory areas.

P2.129

In vivo olfactory receptor neuron responses to a binary mixture of odor molecules targeting the same OR type: citral and octanal

DuchampViret, P. (Lyon)[1], Ferry, B. (Lyon)[1], Kuczewski, N. (Lyon)[1]
[1]*INSERM U1028 - CNRS UMR5292 - CRNL, Centre de Recherche en Neuroscience de Lyon, Olfaction et mémoire, Lyon, France*

Single olfactory receptor neurons (ORN) recordings in the rat in vivo show the activity of the periphery of the olfactory system operating in conditions very close to physiology. Such experiments have definitely proved the wide molecular tuning of the ORNs and the olfactory receptors (OR) they express and the use of this technique leads us to examine the question of mixture coding. Our first study provided evidence that the molecules in a binary mixture interact simultaneously at the ORN level so as to result in suppressive or synergistic action. Our second study chose a specific woody-fruity odorant pair mixture which was reported in psychophysical studies as inducing different odor quality perception in humans when the ratio between each component was changed. Using similar stimulating protocol, we showed that ORN responses faithfully reflected these perceptual changes. Here, we chose an aldehyde pair: Citral (CIT) - Octanal (OCT), reported to target the same OR type. When used singly, OCT induced stronger responses than CIT. When used in a mixture, CIT was shown to have a suppressive action on OCT excitatory effect and the response threshold was shifted towards higher concentrations. The shifting effect suggests a binding competition between CIT and OCT at the same receptor pocket level. In addition, changes in the CIT / OCT ratio in the mixture impacted both the interaction strength and the interaction type between the two molecules and ONRs. Given these results, it makes sense to hypothesize that in natural mixtures made of several tens, hundreds or thousands of compounds, the complexity of interactions could drastically increase. Thus, the wide variety of components or notes which compose natural or synthetic scents will generate specific and complex interactions with ONRs giving birth to a specific level of activity in a unique neuron assembly. Such a peripheral coding mode would not only compensate the poor selectivity of ORNs but would confer to the olfactory system a quasi infinite plasticity. The emergent quality of an odor mixture has long been believed to arise from integration of olfactory information in the CNS. We have shown that ORN are capable of refined information processing since the entrance of the odor coding pathways.

P2.130

Bilateral control of a one-dimensional robotic actuator by operant conditioning of single units in rat motor cortex

Ego-Stengel, V. (Gif sur Yvette)[1], Arduin, P.-J. (Gif sur Yvette)[1], Frégnac, Y. (Gif sur Yvette)[1], Shulz, D. (Gif sur Yvette)[1]
[1]*CNRS UPR 3293, Unité de Neuroscience, Information et Complexité, UNIC, Gif sur Yvette, France*

Operant control of a prosthesis by neuronal cortical activity is one of the successful strategies for implementing brain-machine interfaces, by which the subject learns to exert a volitional control of goal-directed movements.
Here, several motor cortex neurons were recorded simultaneously in head-fixed awake rats and were trained, one at a time, to modulate their firing in order to control the speed and direction of a 1D actuator carrying a water bottle. In the first phase of the experiment, the bottle could only move in one direction and this was triggered by an increase in firing rate. Most neurons submitted to this conditioning successfully increased their activity during trials, and this effect was enhanced across sessions. Once trained, the neuron chosen to control the operant behavior reacted consistently more rapidly than the other recorded neurons after trial onset. We observed also that the firing rate variability increased in an anticipatory way before trial onset, specifically for the neurons that could be conditioned successfully. However, this effect was observed only in the initial sessions of the conditioning.

In the second phase of the experiment, neurons modulated their firing rate up or down in order to control the direction and speed of the water bottle. The bottle could thus move bilaterally, and the goal was to maintain the bottle in front of the rat's mouth in order to allow drinking. All conditioned neurons adapted their firing rate to the instantaneous bottle position so that the drinking time was increased relative to chance. The mean firing rate averaged over all trajectories depended on position, so that the mouth position operated as an attractor (at least for the bottle starting side). Again, the conditioned neuron reacted on average faster than the other neurons and led to a better bottle control than if trajectories were simulated using the activity of simultaneously recorded neurons.
Overall, our results demonstrate that conditioning single neurons is a suitable approach to control a prosthesis in real-time, and that these neurons occupy a lead position after learning, acting as "master" neurons in the network.
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P2.131
Double knockout mice lacking histamine and orexins: a full model of narcolepsy for physiopathological and therapeutic studies

Buda, C. (Lyon)[1], Ouk, K. (Lyon)[1], Anaclet, C. (Lyon)[1], Perier, M. (Lyon)[1], Franco, P. (Lyon)[1], Lin, J.-S. (Lyon)[1]
[1]*CRNL, INSERM U1028-CNRS UMR5292, Univ. Cl. Bernard Lyon1, Physiologie intégrée du système d'éveil, Lyon, France*

Using knockout (KO) mice lacking histamine (HA) or orexins (Ox), we have previously shown that HA and Ox exert a distinct but complementary control of wakefulness(W): the amine, mainly responsible for the qualitative cognitive aspect, its deficit leads to somnolence, whereas Ox, more involved in locomotion and behavioural activation, their defect causes narcoleptic attacks (direct onsets of paradoxical sleep from W, Anaclet et al., J.Neurosci.2009). To assess their synergy in W control under physiopathological conditions, we have generated a double KO mouse strain which is lack of both HA and Ox (HO-/-) and which shows all major narcoleptic phenotypes: cataplectic (sudden loss of muscle tone during W) and narcoleptic attacks, somnolence and hypersomnia (Anaclet et al., Sleep (suppl) 2010). This mouse strain is therefore a full model of narcolepsy. In this study we further characterized its pharmacological responses. Adult male HO-/-mice and their wild-type (WT) littermates (n=16 pairs) were simultaneously investigated using multidisciplinary approaches, e.g., PCR genotyping, sleep-wake monitoring, EEG spectral analysis and pharmacological dosing. As we have shown, HO-/-mice presented all phenotypes of the single KO mice lacking HA or Ox, notably narcoleptic attacks during darkness and an aggravated hypersomnia at lights-off, during darkness and over 24. We then found that 1) Modafinil (64 mg/kg, p.o.), a clinically used W-promoting agent, enhanced W during both lightness and darkness; 2) HA H3-receptor inverse agonists enhanced markedly W and cortical fast rhythms in WT mice, but had no effect in HO-/-mice, indicating absence of functional HA; 3) orexin-A (3 µg, i.c.v.) increased W and suppressed narcoleptic attacks in HO-/-mice; 4) scopolamine, a muscarinic antagonist (0.25-0.5 mg/kg, i.p.) induced no visible effects in WT but increased cortical slow activity and decreased W, paradoxical sleep and narcoleptic attacks in HO-/- mice. The hypersomnia of HO-/-mice indicates gravely impaired arousal mechanisms in spite of possible up-regulation of the waking systems other than HA and Ox, such as the cholinergic system. This murine model appears appropriate for pathophysiological and therapeutic studies of narcolepsy.

P2.132
Effects of olfactory experience on the glomerular maps in the mouse olfactory bulb

Martin, C. (Orsay)[1], Chery, R. (Orsay)[1], Pain, F. (Orsay)[1], Gurden, H. (Orsay)[1]

[1]*Université Paris-Sud 11, CNRS UMR 8165 Imagerie et Modélisation en Neurobio et Cancéro, Orsay, France*

Cortical plasticity is a critical feature of sensory brain areas. In the olfactory bulb, the first central relay of olfactory coding, odor stimulations evoke spatio-temporal patterns of activity. Plasticity in this structure has been shown by recording oscillatory activities of the local field potential during learning. Spatial coding is characterized by the activation of maps of glomeruli, a key functional unit that contains the synapses between the olfactory receptors and mitral cells. Although glomerular maps have been characterized for hundreds of odorants very few is known concerning their plasticity. In vivo intrinsic optical imaging is a technique based on changes of endogenous optical properties of the brain tissue during activation and is commonly used to map odor-induced spatial activity. We used this technique to compare glomerular maps obtained for the same odors before and after an operant olfactory conditioning task. Mice were implanted with a chronic cranial window centered on the olfactory bulb and activity was recorded under anesthesia for a set of odors which will be used for training. They were trained to solve an odor discrimination task (go-nogo task) where one odor is associated with a reward and one odor is not (paired condition). In another group, the odors were randomly associated with the reinforcement (unpaired condition). After mice had reached the criterion, glomerular maps were recorded in the same conditions as initially. We observed no reorganization of spatial maps or change in the intensity of odor responses in the paired group. However in the unpaired group, while the spatial maps were not modulated by the odor stimulation, we saw a decrease of activation following presentation of the odors they were exposed to. Taken together, our data demonstrate an impact of the animal's experience on the spatial dimension of odor representation.

P2.133

Thalamic function during active whisker sensing in head-restrained mice

Urbain, N. (Lyon)[1], Petersen, C. (Lausanne)[2]
[1]*Univ. Claude Bernard Lyon 1, Fac. de Médecine Lyon-Est, Site La Buire-Laennec, CNRS UMR 5292 - INSERM U1028, Lyon, France,* [2]*Ecole Polytechnique de Lausanne, LSENS, Lausanne, Switzerland*

In rodents, active somatosensory perception is particularly relevant through rhythmic and rapid sweeps of facial whiskers contacting objects in the surrounding environment. Faithful transmission of whisker-related sensory information from the periphery to the neocortex is thought to occur through the ventroposteromedial thalamic nucleus (VPM). On the other hand, neurons of the posterior group of the thalamus (Po), the other major somatosensory thalamic nucleus, respond weakly to whisker stimuli because they receive strong and rapid inhibitory inputs from the zona incerta arriving before excitatory inputs from the brainstem (1). Our current knowledge of thalamic processing in the whisker sensorimotor system is largely based on recordings from anesthetised rats, but to uncover the functional relevance of these parallel sensory pathways, there is a need to make measurements in awake animals during whisker-related behaviour.
We therefore developed a technical approach to perform intra- and extracellular single-unit recordings in the mouse somatosensory thalamus coupled with local field potential recordings in primary somatosensory barrel cortex and video-tracking of whisker movements. We found a significant increase of VPM, but not Po, single-unit activity during whisking behaviour. Intracellular recordings revealed a lack of excitatory inputs in some Po cells during whisker movements. Furthermore, inhibitory inputs immediately preceding whisker movement onset could be observed in the Po and dorsal VPM. We therefore performed single-units recordings in the ventral part of the zona incerta, which receives trigeminal inputs and sends inhibitory outputs to Po cells, and found evidence for a strong increase in the activity of these incertal cells in the 300 ms preceding whisking onset. Maximal incertal firing was recorded at whisking onset, but slowed down and returned to baseline before whisking offset. This inhibitory gating on thalamic cells by the zona incerta may therefore occur at a crucial time to shut down suboptimal sensory information due to whisker movement *per se* and hence sharpen selectivity for reliable stimulus identification.
(1) Lavallée et al., J. Neurosc., 2005.

P2.134
Terminal error vs. prediction error in prism adaptation

Gaveau, V. (Bron)[1], Priot, A.-E. (Brétigny-sur-Orge)[2], Prablanc, C. (Bron)[1]
[1]Centre de Recherche en Neurosciences de Lyon (CRNL) Inserm U 1028 - CNRS UMR 5292, Equipe IMPACT, Bron, France, [2]Institut de Recherche Biomédicale des Armées, Brétigny-sur-Orge, France

The study of short-term visuomotor adaptation to prisms shifting the visual field still raises some debate about the involved error signals. Indeed, this plasticity is based on distinct sources of error signals. The two main sources of error that can induce adaptation are: i) the terminal error given by the simultaneous vision of the target and of the hand at the movement end, and ii) the conflict between the prediction of visual feedback of the moving hand and its actual perception. These two sources of error are naturally closely linked. The objective of the present study was to determine the relative processing of the two error signals cited above. As prism adaptation may also involve a cognitive component that we wanted to mitigate, the deviation was introduced in such a way that subjects did not have any conscious experience of it. Two conditions were compared: in the first one ('static error'), the only available information was the vision of the hand and target at movement end; in the second one ('dynamic error'), the vision of the hand was limited to the duration of motion, in the absence of visual target. Only the static error condition led to the emergence and persistence of an adaptive aftereffect at the end of the prism exposure. The aftereffect consisted in a shift of the movement in the direction opposite to the prismatic deviation, whatever the direction of the targets toward which the movement was executed. This demonstrates the crucial role of the terminal error in the adaptation process.

P2.135
Functional anatomy of the perception of bilateral competitive visuotactile stimuli and of a crossmodal extinction-like effect in healthy individuals

Jacobs, S. (Bron)[1], Hadj-Bouziane, F. (Bron)[1], Salemme, R. (Bron)[1], Ibarrola, D. (Bron)[2], Sappey-Marinier, D. (Bron)[2], Meunier, M. (Bron)[1], Farnè, A. (Bron)[1]
[1]INSERM U 1028, CNRS UMR 5292, Université Lyon I, Centre de Recherche en Neurosciences de Lyon, Equipe ImpAct, Bron, France, [2]CERMEP - Centre d'imagerie du Vivant, Bron, France

Extinction patients are unable to consciously perceive a contralesional stimulus when it is presented simultaneously with an ipsilesional one, while they generally detect the very same stimulus when presented in isolation. This phenomenon may occur within and across sensory modalities. Although the involvement of the posterior parietal cortex in the perception of competitive sensory events is suggested by studies in extinction patients, the functional anatomy of competitive perception remains largely unexplored in the intact brain. As a first attempt to investigate this issue, we used functional magnetic resonance imaging to monitor neural activity while participants performed a behavioral task recently developed by our group to induce extinction-like competitive effects in healthy individuals. Participants were asked to report whether they detected a stimulus on the left or the right side, or both, while visual and/or tactile stimuli were delivered. Visual and tactile stimuli were independently titrated so that participants would detect 90% of single stimulations. Stimulation conditions involved either single stimuli (SS), or double simultaneous visuotactile stimuli (DSS). Participants responded by releasing a pedal with the foot (or feet) corresponding to the side(s) where they had perceived a stimulation.
Consistent with previous findings, participants´ performance on DSS was significantly lower than on SS, thus mimicking clinical extinction. The conscious detection of DSS as compared to that of SS (either visual or tactile) was associated with increased activity within the intra-parietal sulcus (IPS) in

the left and, to a lesser extent, right hemisphere. In addition, preliminary analyses suggest that the middle section of the left IPS responds less strongly on DSS trials where participants missed one of the two stimuli. Previous work has shown that the spatial bias exhibited by neglect/extinction patients correlates with the functional imbalance remotely induced in these areas by their typically more ventral parietal lesion. Extinction might thus be an exacerbated manifestation of a physiological limit to the sensory capacities of the normal brain, induced by a functional imbalance in the neural network normally involved in competitive perception.

P2.136

Spatial organization of neurons coding for complex multi-whisker features in S1bf

Léger, J.-F. (Paris)[1], Estebanez, L. (Paris)[1,2], Bertherat, J. (Paris)[1], Shulz, D. (Gif sur Yvette)[2], Bourdieu, L. (Paris)[1]
[1]Ecole Normale Supérieure, IBENS UMR CNRS 8197, Paris, France, [2]CNRS UPR 3293, UNIC, Gif sur Yvette, France

During exploration of the environment, rats contact objects with multiple whiskers in a complex spatio-temporal pattern. Recent electrophysiological experiments (Estebanez et al., Nat. Neurosc. 2012) have demonstrated, using multi-whisker sensory stimulations, the existence of “global” neurons that encode correlated deflections of all whiskers and “local” neurons that detect angular contrast between the principal and surrounding whiskers.
We used two-photon fluorescence microscopy (TPFM) to explore the possibility that functional maps are associated with these newly described tuning functions in the barrel cortex. A 25-whisker stimulator that allows the stimulation of each whisker in all directions (2D) and with a 1kHz bandwidth (Jacob et al., 2010) was combined with TPFM to record optically the sensory-evoked cortical activity. By combining functional imaging in the presence of correlated and uncorrelated stimulations and post-mortem histology with 50µm resolution, we demonstrate that local and global neurons are spatially segregated in layer II/III of the barrel cortex. This spatial segregation is pointing to the possibility that different neural networks in the upper layers of the barrel cortex sustain different functional roles in sensory processing and are put into play by different input statistics.

P2.137

Striatal ensembles continuously represent animals kinematics and limb movement dynamics during execution of a locomotor habit

Robbe, D. (Marseille)[1], Rueda-Orozco, P. (Marseille)[1]
[1]INMED INSERM U 901, Marseille, France

The sensorimotor striatum contributes to the normal execution of motor habits but the mechanisms underlying this function are largely unknown. Motor habits are stereotyped sequences of movements learned through a long trial-and-error process, automatically triggered by a set of sensory cues and that tend to persist despite outcome degradation (e.g. reward omission). We found that rats running on a treadmill become proficient in a fixed time interval estimation task by developing a highly stereotyped locomotor routine. Consistently with the definition of habits, the routine was acquired slowly (at least 2 months of daily practice), and once learned, it persisted for several sessions when the rewarding outcome was omitted. We took advantage of this unexpected behavior and used tetrode arrays to record the spiking activity of dorsolateral striatal ensembles while rats perform the locomotor habit. We report sequential activations of striatal neurons during the entire execution of the task. Importantly, we found that the firing rate of a large fraction of neurons was either locked to the

locomotor limb movements or correlated with the kinematics of the habit (running speed, acceleration, position and time). These results contrast with the long-standing view that striatum is mainly concerned with action initiation. Rather movements and task kinematics encoding suggest that the striatum continuously controls the spatiotemporal execution of habitual action. Additional experiments are currently being performed to further investigate this hypothesis.

P2.138

Improving BCI performance by endowing the machine with adaptive behavior

Perrin, M. (Lyon)[1], Maby, E. (Lyon)[1], Bertrand, O. (Lyon)[1], Mattout, J. (Lyon)[1]
[1]*INSERM U 1028, CNRS UMR 5292, University Lyon 1, Lyon Neuroscience Research Center, DyCog Team, Lyon, France*

A major challenge in Brain-Computer Interfaces (BCI) is the optimization of performance, despite the within and between user variability. To this aim, a promising option is to move towards adaptive BCI to accommodate differences or fluctuations and thus optimize performance, in a mental-state dependent fashion. An obvious criterion to be optimized is the speed-accuracy trade-off. Our objective was to implement and validate an adaptive approach based on the sequential updating of the machine's belief and the enabling of decision making at any time.
We designed and evaluated a BCI whose decision speed depends on the estimated reliability of accumulated evidence. We instantiated a probabilistic classifier whose outcome can be up-dated based on each new incoming information. Then an information theoretic measure enabled us to derive an optimal stopping strategy, in a user-dependent fashion.
We first evaluated the proposed approach offline, using simulated and real data in the context of P300-based spelling. We observed enhanced performance with the adaptive approach compared to the classic one, when stimulation stops after a fix number of stimulations. We then hypothesized that this improvement would prove even larger online, due to a motivation boost. Indeed, we predicted a stronger effect when the user would be instructed and would find out that the more she focuses her attention, the more accurate and the faster the spelling.
To test this hypothesis, we recorded eleven healthy and BCI-naïve subjects in three different online conditions, namely a fix (standard) mode and two (risk-prone and risk-averse) adaptive modes. This way, we could separately quantify the respective contributions of the methodological innovation and its motivational consequences. We confirmed a positive and significant effect of the adaptive methods, but we also observed an additional improvement that could only be attributed to an ensuing enhanced motivation.
To summarize, the proposed adaptive approach generates a virtuous circle such that the more the participant engages into the task, the higher the spelling accuracy and hence the higher the motivation, which in turn yields a further improved interaction.

P2.139

What does the awake monkey brain at rest tell us?

Guedj, C. (Bron)[1], Monfardini, E. (Bron)[1,2], Farnè, A. (Bron)[1], Meunier, M. (Bron)[1], Hadj-Bouziane, F. (Bron)[1]
[1]*Centre de recherche en neurosciences de Lyon-Univ Lyon 1, INSERM U 1028 - CNRS UMR 5292, Impact, Bron, France,* [2]*Institut de Médecine Environementale, Paris, France*

Intrinsic brain activity at rest revealed by functional imaging is generating a great deal of interest. Extracting low-frequency fluctuations (< 0.1Hz) from the brain activity at rest uncovers spatially-

independent correlated networks, so called resting-state networks (RSNs), that correspond to functional entities. Because, up to now, most studies have been conducted in humans, RSNs in the monkey, a major animal model in brain research, remain little known. To help fill this void, we applied independent component analysis (ICA), a statistical data-driven method that identifies spatially independent maps with different time-courses, to imaging data recorded in three awake rhesus macaques. Functional images were acquired on a 1.5 T scanner using a contrast agent (MION) while the monkeys were sitting motionless in the dark (400TRs, TR=2s, voxel size=2x2x3mm). Data were analyzed using MELODIC, a probabilistic ICA approach (Beckmann and Smith, 2004). Seven cortical RSNs were reminiscent of those consistently found in humans: the fronto-parietal, default mode, somatomotor, somatosensory/auditory, frontal executive and occipital visual RSNs. The latter was split, as in humans, into foveal and peripheral occipital visual maps. Two subcortical networks, the thalamic and the basal ganglia RSNs, were similar to those characterized in some human studies. Also, as in humans, inter-subject variability appeared higher for RSNs involving association cortices than for RSNs involving primary cortices. Only two findings were somewhat at odds with previous human data. Unlike the human “attentional” RSN, the monkey fronto-parietal RSN was bilateral with no apparent ventral vs. dorsal segregation. Conversely, we identified two temporal visual RSNs in the monkey, not typically reported in humans using ICA: a visual motion RSN, including areas MT and MST, and a visual object-recognition RSN, located around the anterior middle temporal sulcus. Beyond demonstrating strong homologies between the monkey and human brain activity at rest, our data strongly encourage cross-examination of human functional imaging data in light of monkey findings and vice versa.

P2.140

Synfire chains and gamma oscillations: two sides of the same coin?

Hahn, G. (Gif sur Yvette)[1,2], Bujan, A. (Freiburg)[3], Frégnac, Y. (Gif sur Yvette)[1], Aertsen, A. (Freiburg)[3], Kumar, A. (Freiburg)[3]
[1]CNRS, Unité de Neuroscience, Information et Complexité (U.N.I.C), UPR 3293, Gif sur Yvette, France, [2]New York University, Center for Neural Science, New York, United States, [3]University of Freiburg, Bernstein Center Freiburg, Neurobiology & Biophysics, Faculty of Biology, Freiburg, Germany

The cortex is thought to process stimuli through a hierarchy of specialized brain areas, which requires mechanisms that transmit neuronal activity faithfully from one cortical area to the next. Two different solutions have been proposed based on neuronal synchronization to establish precise communication between separated cortical areas. In the synfire chain model, synchrony spreads along specialized pathways with divergent/convergent connections and is driven by common input from presynaptic neurons. Alternatively, the coherent oscillation model exploits synchronization provided by oscillations of local neuronal populations and proposes consistent phase relationships between oscillators of different brain areas as the key for neuronal communication. Here we propose a novel framework, in which coherent oscillations are a direct consequence of synchrony emerging in diluted feed-forward networks (FFNs), whose connectivity (weight and number of connections) is insufficient to allow stable propagation of excitatory spike volleys. Using numerical simulations of diluted FFNs, we show that excitatory spike volleys can be progressively amplified by a resonant oscillation which is generated by local inhibition within each layer, and compensates for weak and sparse connections of the FFN. As a consequence, a coherent oscillation spreads across the FFN, a process which we call an “oscillation chain”. We hypothesize that the coherent oscillatory dynamics could induce synaptic potentiation and gradually transform oscillation chains into synfire chains, a prediction that was confirmed in our simulations. In summary, our results establish a link between the concepts of synfire chains, coherent oscillations and synaptic plasticity, and argue that transient oscillations are a consequence of stable propagation of spiking activity between different networks.
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P2.141

Locomotion-respiration coordinations: the remote control of pFRG neuron activity by lumbar locomotor CPG

Le Gal, J.-P. (Bordeaux)[1], Juvin, L. (Bordeaux)[1], Cardoit, L. (Bordeaux)[1], Thoby-Brisson, M. (Bordeaux)[2], Morin, D. (Bordeaux)[1]
[1]Univ. Bordeaux-CNRS UMR 5287, INCIA, Equipe CPGs, Bordeaux, France, [2]Univ. Bordeaux-CNRS UMR 5287, INCIA, Equipe OASM, Bordeaux, France

During locomotion, respiratory frequency increases to satisfy the associated increase in oxygen demands. We previously showed that one neurogenic mechanism involved in the modulation of breathing frequency during exercise relies on the recruitment of sensory afferents from limb muscles. Repetitive locomotor-related movements cause periodic activation of muscle proprioceptors, which in turn rhythmically excites the respiratory centers (1) and modulates phrenic motoneuron excitability (2). However, the role played by spinal locomotor generators (CPG) in the adaptation of respiratory frequency is poorly documented.
Using a neonatal rat brainstem-spinal cord preparation, we report that the pharmacological activation of lumbar locomotor CPG by bath-applied NMA and serotonin increases the frequency of respiratory burst activity recorded from cervical ventral roots. When present, this ascending lumbar influence facilitates the occurrence of in-phase coordinations between the locomotor discharges and the phrenic respiratory bursts. This preferential phase-relationship disappears when the ascending axonal conduction in the thoracic spinal segments is prevented by means of sucrose blockade.
Subsequent experiments have been performed to identity the potential neural target of this ascending spinal influence. During an episode of fictive locomotion, the activation of lumbar networks induces a sustained depolarization of identified respiratory neurons belonging to the parafacial respiratory group (pFRG). On the basis of the crucial role played by the pFRG in the respiratory rhythmogenesis, this tonic depolarization could be at the origin of the locomotor CPG-induced increase in respiratory rate. Indeed, we finally show that the increase in the respiratory frequency induced by the activation of lumbar locomotor CPG is no longer observed after bilateral lesion of the pFRG.
In the newborn rat, in addition to the ascending influence of proprioceptive afferents and their role in respiratory entrainment during locomotion, the spinal locomotor CPG themselves can also finely tune the respiratory frequency by an ascending modulation of the brainstem respiratory circuitry.
(1) Giraudin A et al. (2012) *J Neurosci* 32:11841.
(2) Morin D, Viala D (2002) *J Neurosci* 22:4756.

P2.142

Multi-sensory integration within the non-human primate cortex: a functional Magnetic Resonance Imaging (fMRI) study

Clery, J. (Bron)[1], Guipponi, O. (Bron)[1], Odouard, S. (Bron)[1], Wardak, C. (Bron)[1], Ben Hamed, S. (Bron)[1]
[1]Institut des Sciences Cognitives, CNRS UMR 5229, Bron, France

Multisensory integration is a neuronal process by which the response of a neuron to two sensory stimuli of different modalities presented simultaneously is different from the response to each sensory stimulus presented independently. Complementing a twin poster on multisensory convergence in the non-human primate brain (Guipponi et al., *Société française des neurosciences*, Lyon, 2013), we used functional magnetic resonance imaging (fMRI) in behaving monkeys to provide a whole brain view of visuo-tactile (VT) integration. Manipulating the spatial and temporal congruence between the visual

and the tactile stimuli, we probe the hypothesis that in the context of VT integration around the face, maximal multisensory integration takes place when the tactile stimulus follows the visual stimulus and is predicted by it (Ben Hamed et al., *Société française des neurosciences*, Lyon, 2013).
Monkeys were required to maintain fixation. In order to maximize multisensory integration, visual stimulations consisted in degraded dynamic 3D visual stimuli moving from away towards the face of the monkey and tactile stimulations were directed to the center of the face thanks to airpuffs and adjusted so as to be very faint. We used a blocked fMRI design. Visual (resp. tactile) blocks consisted in visual (resp. tactile) stimuli presented alone. VT blocks were of three types: the tactile stimulus was presented,
1) while the visual stimulus was approaching the face of the monkey (mid-course of the visual stimulus), at the location at which the visual stimulus was expected to impact the face (VT_0);
2) while the visual stimulus was approaching the face of the monkey, at the symmetric location from that at which the visual stimulus was expected to impact the face (VT_spatial;
3) at the moment when the visual stimulus was expected to impact the face, at the spatial location of the expect impact (VT_predictive).
In agreement with our working hypothesis, visuo-tactile essentially takes place within the VT convergence network. The dependence of the fMRI activations on the VT spatial and temporal congruence is complex and varies as a function of the cortical sub-network being considered. Overall, this study fosters our understanding of the neural bases of VT integration.

P2.143

Implication of neuronal excitability and electrical coupling in expression of compulsive-like feeding behavior induced by learning and dopamine in *Aplysia*

Bedecarrats, A. (Bordeaux)[1], Sieling, F. (Atlanta)[2], Simmers, J. (Bordeaux)[1], Prinz, A. (Atlanta)[3], Nargeot, R. (Bordeaux)[1]
[1]University Bordeaux, CNRS UMR 5287 INCIA, Équipe OASM, Bordeaux, France, [2]Georgia Institute of Technology and Emory University, Wallace H. Coulter Department of Biomedical Engineering, Atlanta, United States, [3]Emory University, Department of Biology, Atlanta, United States

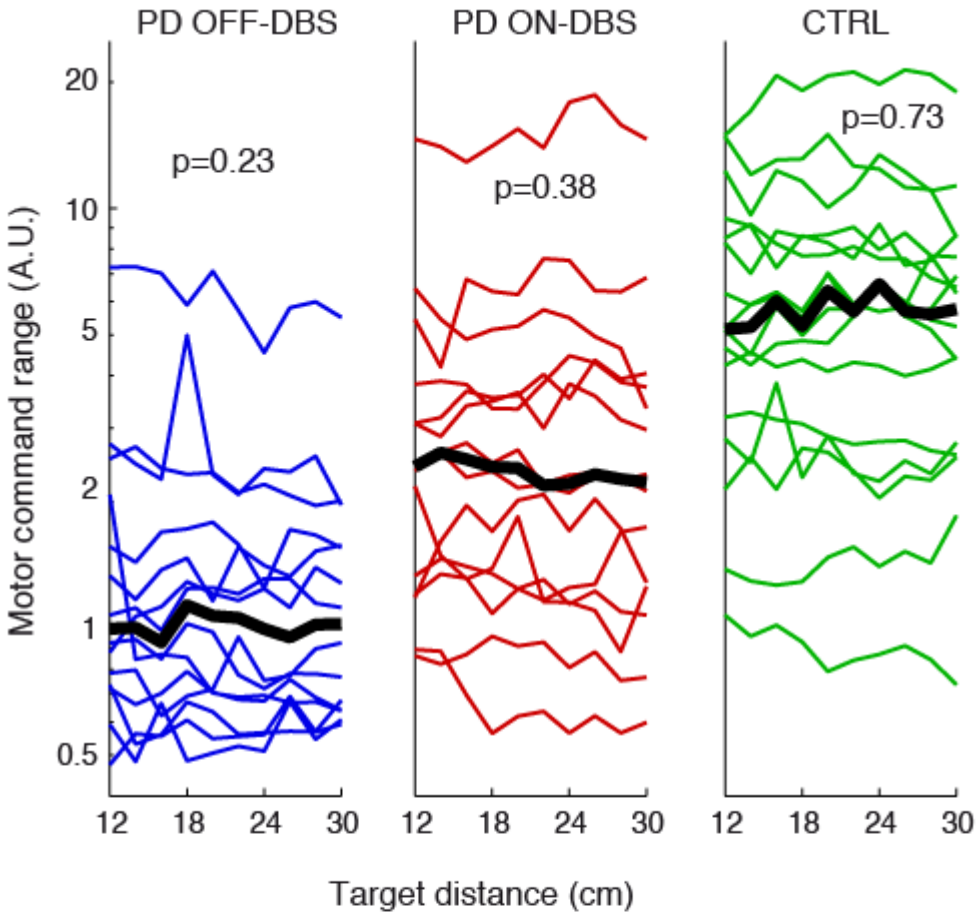
Operant reward learning of the goal-directed feeding behavior in *Aplysia* switches the infrequent and irregular tongue-like radula movements to frequent and stereotyped, compulsive-like, repetition of the action. This behavioral plasticity is associated with an increase in the excitability and electrical coupling of the neurons B63/B30/B65 which are responsible for initiating the radula motor pattern. In the present study, we explored the contribution of dopamine (DA) in the induction of the neuronal plasticity and the causal relationship between these cellular changes and the learning-induced acceleration and regularity of the motor pattern genesis.
In isolated Buccal ganglia (B.g.) which contain the radula motor pattern generating network, the learning-induced increase in the B63/B30/B65 excitability and electrical coupling was reproduced by an electrical stimulation of the esophageal nerve which convey the rewarding information. This effect was blocked by a DA receptor antagonist thereby suggesting a role for DA in the induction of this plasticity. Moreover, using a real time neuron/computer interaction (dynamic clamp) to artificially manipulate the excitability and electrical coupling in B63/B30/B65 in B.g. isolated from naïve *Aplysia,* we found that an increase in the cells excitability modified the frequency, but not the regularity, of the motor pattern genesis. An artificial increase in the electrical coupling modified the regularity but not the frequency of the motor pattern genesis. A simultaneous change in the neuronal excitability and coupling reproduced all aspects of the learning-induced plasticity including the transition from irregular/infrequent to regular/frequent motor pattern genesis. In B.g. isolated from trained animals, artificial changes in neuronal excitability and coupling switched the learning-induced cellular plasticity to a naïve state and the rhythmic/frequent motor pattern genesis to an arrhythmic/infrequent mode. These data suggest that two separate mechanisms, induced by DA, may provide a cellular substrate for the behavioral switch between impulsive to compulsive motor actions in *Aplysia*: an increase in excitability and in electrical coupling which control frequency and stereotypy of motor pattern generation, respectively.

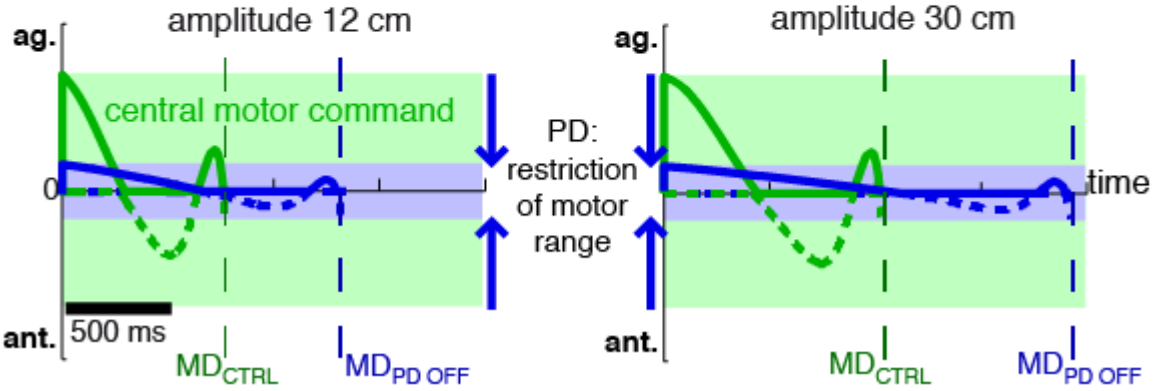
P2.144

A common optimization principle for motor execution in healthy subjects and parkinsonian patients

Baraduc, P. (Bron)[1], Thobois, S. (Bron)[2], Gan, J. (Bron)[2], Broussolle, E. (Bron)[2], Desmurget, M. (Bron)[1]
[1]*Centre de Neuroscience Cognitive, UMR 5229-CNRS, Université Lyon1, Bron, France,* [2]*Hôpital Neurologique Pierre Wertheimer, Bron, France*

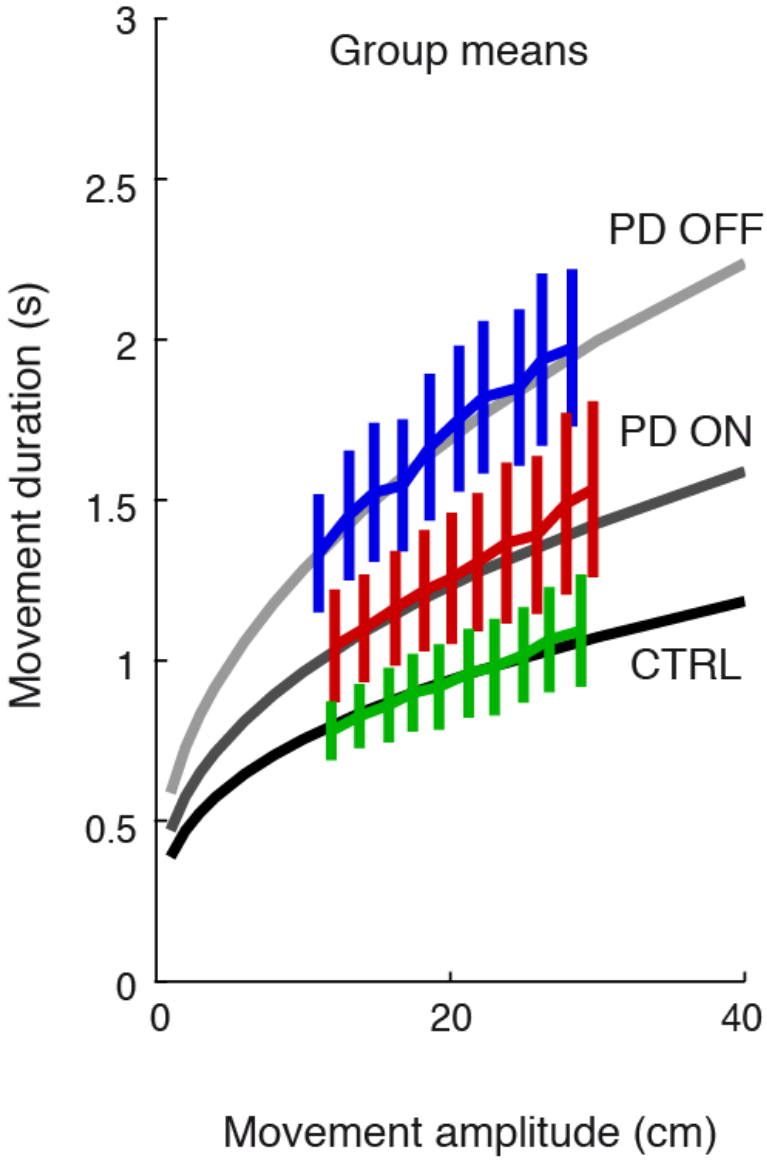
Bradykinesia in PD has recently been suggested to result from a reduction in motor motivation. This hypothesis is tested in the framework of optimal control theory, which accounts for many characteristics of healthy human movement while providing a link between the motor behavior and a cost/benefit tradeoff. This approach offers the opportunity to interpret movement deficits of PD patients in the light of a computational theory of normal motor control. We studied 14 PD patients with bilateral subthalamic nucleus (STN) stimulation and 16 age-matched healthy controls, and tested whether reaching movements were governed by similar rules in these two groups. A single optimal control model accounted for the reaching movements of healthy subjects and PD patients, whatever the condition of STN stimulation (On or Off). The choice of movement speed was explained in all subjects by the existence of a preset dynamic range for the motor signals.





[Constant motor range]

This range was idiosyncratic and applied to all movements irrespective of their amplitude. In PD patients this dynamic range was abnormally narrow and correlated with bradykinesia. STN stimulation reduced bradykinesia, widened this range in all patients but did not restore it to a normal value. These results, consistent with the motor motivation hypothesis, suggest that constrained optimization of motor effort is the main determinant of movement planning (choice of speed) and movement production, in both healthy and PD subjects



[Movement duration, data & model]

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P2.145

Grasping with allografted hands: a kinematic study

Huchon, L. (Bron)[1], Farné, A. (Bron)[1], Rode, G. (Bron)[1], Revol, P. (Bron)[1], Badet, L. (Lyon)[2], Delporte, L. (Bron)[3], Arsenault, L. (Lyon)[4], Bernardon, L. (Miribel)[5], Molia, C. (Miribel)[6], Rossetti, Y. (Bron)[7], Morelon, E. (Lyon)[8], Gazarian, A. (Lyon)[9]

[1]Centre de Recherche en Neurosciences de Lyon (CRNL), INSERM U 1028 - CNRS UMR 5292 - Université Lyon 1, Equipe ImpAct, Bron, France, [2]Hospices civils de Lyon, Service de chirurgie de la transplantation et urologie, Hôpital Edouart Herriot, Lyon, France, [3]Centre de Recherche en Neuroscience de Lyon (CRNL), Plateforme Mouvement et Handicap, Bron, France, [4]Hospices Civils de Lyon, Hôpital Henry Gabrielle, service de kinésithérapie, Lyon, France, [5]Centre Romans Ferrari, Service d'ergothérapie, Miribel, France, [6]Centre Romans Ferrari, Miribel, France, [7]Centre de Recherche en Neuroscience de Lyon (CRNL), INSERM U 1028 - CNRS UMR 5292 - Université

Claude Bernard Lyon 1, Bron, France, [8]Hospices Civils de Lyon, Hôpital Edouart Herriot - Service de transplantation, néphrologie et immunologie, Lyon, France, [9]Clinique du Parc Lyon, Service de chirurgie orthopédique, Lyon, France

Since the first human hand allograft in 1998, more than fifty patients around the world have received a hand allograft several years after an upper limb amputation, including about twenty-five cases of bilateral hand transplantation. Clinical results of these innovative surgical and immunosuppressive procedures are globally positive, with good functional outcome allowing patients to retrieve autonomy in daily life, in spite of some persistent impairments both at the level of touch sensation and range of motion. The analytic and functional features of prehension movements have been tentatively described for these subjects from a clinical perspective. The graft-induced plastic remodeling in the sensory-motor cortex has been documented by functional neuroimaging, but the molar structure and the visuomotor pattern of prehension movements have never been investigated. We have therefore conducted a kinematic study of the prehension performance in five French bilateral hand allograft receivers. Patients had been transplanted from two and half to nine years after the amputation, and the delay between the graft and our assessment ranged from twelve to five years. Grafted patients were asked to reach and grasp one of three cylinders of different sizes, located either on the left or right hemispace. Both grafted hands were separately assessed, and performance was compared with that of a group of control subjects matched for age, gender and upper limb morphometry. Preliminary results in two patients showed a significant increase of the maximum finger aperture along with the increase of object size in patients as in control subjects, which indicate a preservation of the visuomotor coding dependent on the object size. Moreover, the molar structure of the prehension movements in grafted hand patients also displayed a normal pattern, with a temporal structure of movement parameters comparable to that observed in controls, both when considering the transport component and the grasping component. Our data suggest a remarkably good recovery of the pattern of prehension after a bilateral hand allograft, especially concerning the scaling of the pinch grip according to the object size, even if the transplantation occurred several years after the amputation.

P2.146

Predicting olfactory dysfunction in young age and normal or pathological aging using linear discriminant analysis

Joussain, P. (Lyon)[1], Rouby, C. (Lyon)[1], Bessy, M. (Lyon)[1], Faure, F. (Lyon)[2], Bellil, D. (Lyon)[2], Landis, B. (Genève)[3], Hugentobler, M. (Genève)[3], Tuorila, H. (Helsinki)[4], Vento, S. (Helsinki)[4], Krolak-Salmon, P. (Lyon)[2], Delphin, F. (Lyon)[2], Bensafi, M. (Lyon)[1]
[1]Université Lyon 1, Centre de Recherche en Neurosciences de Lyon, UMR 5292, CNRS, INSERM, Lyon, France, [2]Hospices Civils de Lyon, Lyon, France, [3]Hôpitaux Universitaires de Genève, Genève, Switzerland, [4]University of Helsinki, Helsinki, Finland

The frequency of olfactory dysfunction remains poorly documented in the French population. The prime aim of this study was therefore to provide a tool to measure this prevalence by predicting the occurrence of olfactory troubles in young age and in healthy or pathological aging. To this end, 36 anosmic patients (total loss of olfaction), 36 hyposmic patients (partial loss) and 36 healthy controls were tested using the European Test of Olfactory Capacities (ETOC), a short self-administrated test, combining detection and identification of 16 odors. A discriminant analysis performed on both scores (detection and identification) classified correctly the two groups of patients and the controls with an accuracy of 86%. The model was then tested with a new sample of 212 young subjects and 125 older subjects to assess the prevalence of olfactory troubles in the population as a function of age. In the young sample (mean age 25.4+-5.4), 22 participants (10.4%) were classified as hyposmic and 1 participant (0.5%) as anosmic. In the older group (mean age 61.5+-5.5) 45 participants (36%) were classified as hyposmic and 2 (1.6%) as anosmic.
A second aim of the study was to shorten the olfactory test enabling a fast (5-10 min) clinical evaluation by ENT and neurologists. To this end, 25 patients with olfactory troubles and 25 age-matched controls were tested with a short version of the ETOC including detection and identification of 6 odors. The discriminant analysis reached 84% of correct classifications. The model was applied to 2 samples of healthy young (n=20, age) and elderly participants (n=18, mean age 83.3+-4.9) and a

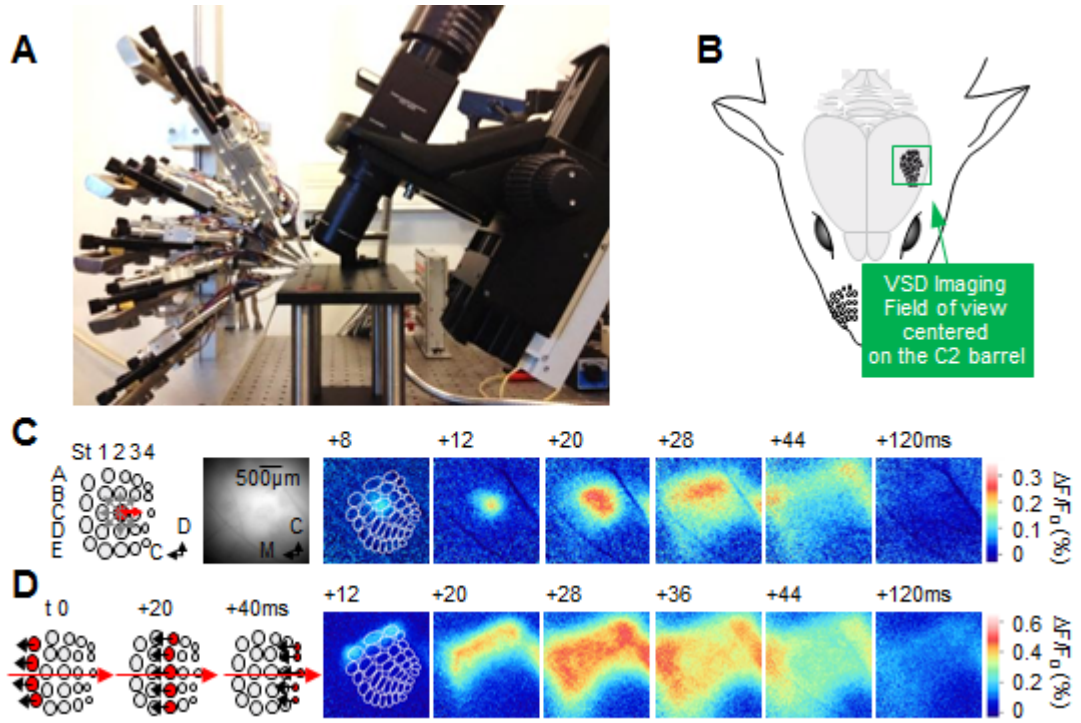
sample of 18 Alzheimer patients (mean age 79.9+-5.4). In the young sample 15% were classified as having olfactory troubles. This proportion grew to 28% in the older healthy group, and to 83% in Alzheimer patients.
This study provides a new tool to assess olfactory function in clinical settings by characterizing the prevalence of olfactory troubles in young age and during normal and pathological aging. Using linear discriminant analysis, we provide here for the first time a statistical tool that enables, for a given individual, to estimate his/her probabilities of being anosmic, hyposmic or normosmic, which is of most importance for clinicians.

P2.147

Direction selectivity in the mouse barrel cortex, a voltage sensitive dye (VSD) imaging study

Shulz, D. (Gif sur Yvette)[1], Vilarchao, E. (Gif sur Yvette)[1], Férézou, I. (Gif sur Yvette)[1]
[1]*Equipe Traitement sensoriel, Neuromodulation et Plasticité, Unité des Neurosciences, Information et Complexité (UNIC) CNRS UPR 3293, Gif sur Yvette, France*

The cortical representation of the rodent's whiskers within the primary somatosensory cortex (S1) contains structures termed “barrels” in layer 4, which are laid out in an identical pattern to the whiskers on the snout. Each barrel vertical column processes information coming primarily from its corresponding whisker and an intracolumnar direction selectivity map has been evidenced in rats. Previous experiments performed in our team with extracellular recordings showed that neurons in the rat barrel cortex and thalamus not only show a direction preference for local stimulation of the corresponding vibrissa but also for global stimulation of the whisker pad (Jacob et al., 2008; Ego-Stengel et al., 2012).
To further understand how integrative properties of the cortical network could sustain different processing modes for local and global tactile scenes, we performed VSD imaging of the mouse barrel cortex under anesthesia while applying global tactile stimulation using a 24-multidirectional whisker stimulator (Fig. A). A field of view of 2.5 x 2.5 mm allowed us to monitor the spatiotemporal dynamics of the 24 barrel-related cortical columns corresponding to the stimulated whiskers (Fig. B).
Local direction selectivity is evaluated by stimulating the C2 whisker in 8 different directions (Fig. C). This allows to assess the existence of a pinwheel-like subcolumnar, supragranular, directional map organization within S1 in mouse.
Global stimulation protocol consisted of applying multiwhisker stimuli that are locally invariant (rostro-caudal deflections), globally coherent (stimulation of several whiskers collectively), but differed in the global “apparent” direction (Fig. D). This protocol allows us to assess the possible existence of a supracolumnar, supragranular, directional topography specific for the global motion selectivity.
Supported by BrainScales, FacetsITN and ANR.



[VSD in mouse barrel cortex]

P2.148

Circadian variations of gene expression related to neuro-metabolic coupling in astrocytes of anterior and posterior hypothalamus of the mice

Petit, J.-M. (Lausanne)[1,2], Mikhaleva, A. (Lausanne)[3], Gyger, J. (Lausanne)[3], Magistretti, P. (Thuwal)[3,4]
[1]EPFL, LNDC, Brain Mind IInstitute, Lausanne, Switzerland, [2]CHUV, Centre de Neurosciences Psychiatriques, Prilly, Switzerland, [3]EPFL, LNDC, Brain Mind Institute, Lausanne, Switzerland, [4]KAUST, Division of Biological and Environmental Sciences and Engineering, Thuwal, Saudi Arabia

The posterior hypothalamus (PH) is crucial for maintaining wakefulness whereas the anterior hypothalamus (AH) constitutes a sleep-promoting region. Consequently, the energy needs of AH and PH should probably change throughout the sleep-wake cycle. The glycogen mobilization altogether with the astrocyte-neuron lactate shuttle (ANLS) likely constitutes two major mechanisms by which astrocytes ensure the neurometabolic coupling. Therefore, we hypothesized that astrocytes of AH and PH may display differences in genes expression involved in these mechanisms during the sleep-wake cycle.
To investigate this, we measured the gene expression levels in astrocytes obtained from transgenic mice expressing the fluorescent protein eGFP under the control of the astrocyte-specific gene GFAP promoter. Mice were sacrificed at four time points, ZT0, ZT6, ZT12 and ZT18 (ZT0 corresponding to light-on). After brain dissection, “punches” of AH and PH were micro-dissected from slices. Cell suspensions were obtained from a pool of 3 samples by enzymatic digestion and cell trituration. Then, GFP-positive cells, which were highly enriched in astrocytes, were sorted by FACS. Total RNA was extracted and gene expression levels were assayed by qRT-PCR and normalized by cyclophylin mRNA levels. Seven genes related to ANLS were tested: the α2 sub-unit of the Na-K ATPase (Atp1a2), the two sub-unit of the lactate dehydrogenase (LDHa and LDHb), the glutamate transporters (GLAST and GLT1) and two monocarboxylate transporters (MCT1 and MCT4). Levels of mRNA encoding genes related to glycogen metabolism such as PTG, the glycogen synthase (GS) and the glycogen phosphorylase (GPhos) were also measured Results showed that the levels of mRNA

encoding LDHa, MCT1 and MCT4 expressed significant circadian variations in AH. The GLT1 mRNA levels as well as MCT1 and MCT4 mRNA levels also displayed significant circadian variations in PH. Levels of expression of genes encoding the Gphos, GS and PTG did not display major regional circadian variation.
The difference in the pattern of expression of ANLS related genes in AH and PH, such as GLT1, suggests that neuro-metabolic coupling capacity of each structure might be different and might play a functional role in the sleep-wake regulation.

P2.149
Dynamic bilateral spatial representation in the monkey frontal cortex

Wardak, C. (Bron)[1], Astrand, E. (Bron)[1], Huguet, N. (Bron)[1], Ben Hamed, S. (Bron)[1]
[1]*Centre de Neuroscience Cognitive, CNRS UMR 5229, Bron, France*

The Frontal Eye Field (FEF) is a well-studied frontal area involved in saccadic eye movements and visual attention. In the monkey, each FEF represents the contralateral visual and motor field within a spatial eccentricity-coded map. However, behavior is rarely constrained to a unique hemifield, and both representations have to be coordinated in a certain way in order to guide an optimized behavior. Previous studies suggest that 1) one FEF could represent ipsilateral locations when the task is bilateral (Ibos et al., in revision) and that 2) both FEF could interact via callous connections, as a unilateral inactivation of FEF leads both to contralesional deficits and ipsilesional “improvements” (Wardak et al., submitted).
Here, we study how both FEF interact and represent the whole visual field by simultaneously recordings in the two areas. Our hypothesis is that we will observe signatures of the bilateral coordination both at the spike level and in the local field potential (LFP) responses. We use 24-contact linear multi-electrodes, one in each FEF, in two monkeys, while they are engaged in saccadic and attentional tasks. The first task tested is a classical memory-guided saccade task and is designed to dissociate between visual- and motor-related processing. The monkeys need to memorize the position of a visual stimulus that is flashed for 80ms and make saccade to its position after a variable delay period. The second task is a peripheral detection task. The monkeys have to fixate a central point and detect a flashed stimulus by releasing a bar. The third task is an automatic receptive field mapping. The monkeys have to fixate a central point while flashes are presented at different locations, covering a 40x40° grid.
Contrary to the classical view that FEF contains only a contralateral spatial representation, we show that FEF neurons can also represent ipsilateral locations, and that this spatial representation is dynamic and depends on task context.

P2.150
Identification of the neuronal network regulating paradoxical (also called REM) sleep in mice

Peyron, C. (Lyon)[1], Arthaud, S. (Lyon)[1], Gay, N. (Lyon)[1], Nouvian, M. (Lyon)[1], Luppi, P.-H. (Lyon)[1]
[1]*CRNL - UCBL - CNRS - UMR 5292 - INSERM U 1028, Lyon, France*

The need to study sleep in mice has drastically increased in the recent year with the development of genetically modified mice models and new methods such as optogenetics and pharmacogenetics. In addition to pharmacological microinjections and electrophysiological recordings, we have combined in rats the use of specific paradoxical sleep (PS) deprivation (PSD) and PS hypersomnia (PSR) protocols, with immunostaining of cFos, commonly used as a marker of neuronal activation. By this mean, we demonstrated in rat that the neurons generating PS are glutamatergic and localized in the sublaterodorsal tegmental nucleus (SLD). In addition, we showed that the SLD send direct projections to glycinergic neurons of the ventral gigantocellular nucleus (GiV) previously shown to be responsible

for the hyperpolarization of motoneurons during PS to induce muscle atonia. We also identified neurons inhibiting PS like the ventrolateral periaqueductal gray (PAG).
Here, using a similar paradigm of specific PSR combined with double immunostaining of cFos with several markers such as tyrosine hydroxylase (TH), acetylcholine transferase (ChAT), hypocretin-1 or melanin concentrating hormone (MCH), we evaluated the commonality and discrepancies of the network generating PS in mice compare to rats.
Neurons selectively active in PSR were found bilaterally in sections at 5.2mm caudal to Bregma, and presumably corresponding to the mouse SLD. They were just ventral to the PAG and mediodorsal to the motor trigeminal nucleus (Mo5). Some of these neurons were intermingled with the laterodorsal tegmental nucleus but none were expressing ChAT indicating that, as in rats, they are not cholinergic in nature. Compare to the rat model, the mouse SLD is slightly more caudal if we use the Mo5 as a reference. On the contrary, the SLD and the locus coeruleus (LC) can be seen at the same level on coronal sections in rats, whereas the mouse SLD is rostral to the LC as we determined using CFos/TH double-immunostaining.
As in rats, we found activated neurons during PSR in the GiV and the MCH neurons in the hypothalamus. None were hypocretin-1 labeled.
Our data indicate that the neuronal network generating PS is similar in mice and rats with some variations in the precise localization of the structures.

P2.151

Sensory hyperexcitability in the *Fmr1* knockout mouse model of fragile X syndrome

Bony, G. (Bordeaux)[1], Ginger, M. (Bordeaux)[1], Frick, A. (Bordeaux)[1]
[1]*INSERM U 862, Neurocentre Magendie, Bordeaux, France*

Fragile X Syndrome (FXS) is the most common form of inherited mental retardation syndrome and a frequent cause of autism spectrum disorders (ASD). FXS is a single gene (*Fmr1*) disorder, which can be reliably modeled by a mutant mouse model—the *Frm1* knockout (KO) mouse. Increased excitability to sensory stimuli is a prominent feature of FXS and ASD, but its underlying mechanisms are poorly understood.
To probe sensory hyperexcitability in *Fmr1*KO mice, and to investigate its cellular and network mechanisms, we performed *in vivo* patch-clamp recordings from layer (L) 2/3 pyramidal neurons in the hindlimb area of the primary somatosensory cortex in ketamine-anesthetized animals. Sensory responses were evoked using stimulation of the contralateral hindpaw (2 ms duration, 30 mA). In addition to sensory responses, we also measured changes in neuronal excitability as a cellular mechanism for sensory hyperexcitability in FXS.
We found that the average spontaneous action potential activity was significantly lower in L2/3 pyramidal neurons in *Fmr1*KO mice as compared to wild-type (WT) mice (KO, 0.057 ± 0.021 Hz; WT, 0.183 ± 0.037 Hz (mean ± SEM); $p = 0.006$). Stimulation of the contralateral hindpaw evoked postsynaptic potentials of similar amplitude in both *Fmr1*KO and WT mice (10.9 ± 1.6 mV vs 9.6 ± 1.3 mV). In contrast, our preliminary data suggest an increase in the hindpaw stimulus-evoked action potential activity (KO, 0.77 ± 0.16; WT, 0.36 ± 0.09) and a reduction in the variability of response (coefficient of variation: KO, 0.92 ± 0.29; WT, 1.61 ± 0.28) in *Fmr1*KO mice. Furthermore, the number of action potentials as a function of current injected was significantly larger in L2/3 pyramidal neurons from *Fmr1*KO mice as compared to WT mice (550 pA; KO, 9.31 ± 0.76; WT, 6.38 ± 0.73; $p < 0.01$), suggesting an increase in neuronal excitability.
Altogether, our results suggest a sensory hyperexcitability phenotype for tactile stimulation in *Fmr1*KO mice, and support the idea that both cellular and network mechanisms may underlie this neocortical hyperexcitability.

P2.152

High astrocytic energy metabolism is required in the somatosensory cortex for the occurrence of seizures in a genetic model of absence epilepsy in the rat

Jarre, G. (La Tronche)[1], Depaulis, A. (La Tronche)[1], Guillemain, I. (La Tronche)[1]
[1]*Grenoble Institut des Neurosciences, INSERM U 836 - Equipe 9,Bât. Edmond J. Safra, Isère, La Tronche, France*

Absence epilepsy is a non-convulsive epileptic syndrome characterized by spike-and-wave discharges (SWD) on the electroencephalogram (EEG), associated with a behavioral arrest. We have recently shown in the GAERS model (Genetic Absence Epilepsy Rat from Strasbourg), that spontaneous seizures are initiated in the barrel region of the somatosensory cortex (SSC) and then spread to the rest of the cortex and the thalamus. These SWD start to occur spontaneously around 25 days after birth. In the cortex of adult GAERS, an increase of glutamate amount has been reported indicating an increase of glutamatergic activities compared to non-epileptic controls. Moreover, astrocytic glutamate uptake, transport of glutamine from astrocyte to neurons and the glutamic acid dehydrogenase, an astrocytic enzyme responsible for the recycling of glutamate to neurons, were enhanced in the cortex. In addition, increases of astrocytic mitochondrial metabolism and of GFAP, a marker of reactive astrocyte, have been observed. Thus, it indicates that the astrocyte-glutamatergic neuron interactions were increased in the cortex of GAERS, in part mediated by astrocytic metabolism anomalies. We hypothesized that the alteration of the astrocytic energy metabolism could be involved in the occurrence of SWD in GAERS. To address this question, we examined the effects of bilateral intracerebral microinjections of fluorocitrate, an inhibitor of astrocytes Kreb's cycle, on the occurrence of SWD, (i) in the SSC and (ii) in the ventrolateral thalamus. We showed that injection of fluorocitrate (100-200pmol/200nl/side) in the SSC induced a dose-dependent suppression of the number and duration of the SWD, as well as a modification in their EEG pattern. On the contrary, we observed no changes in the seizure pattern nor the number and duration of SWD after injection of fluorocitrate (200-400pmol/200nl/side) in the ventrolateral thalamus. Our data suggest that the high metabolism in the astrocytes of the SSC could participate in the generation/initiation of SWD. Altogether, these results suggest that astrocytes may play a role in the physiopathology of absence epilepsy and it raises the question of the impact of this abnormal metabolism on astrocyte calcium excitability.

P2.153

Invlolvement of central amygdala in sensorial activity of the spinal cord

Roca-Lapirot, O. (Bordeaux)[1], Durieux, C. (Bordeaux)[2], Landry, M. (Bordeaux)[2]
[1]*IINS-CNRS UMR 5297, Université de Bordeaux II, Bordeaux, France,* [2]*IINS-CNRS UMR 5297, Université de Bordeaux II, 33, Bordeaux, France*

Pain induces an emotional and affective component involved in motivation and learning. Otherwise, this can be raised in chronic pain cases, making patients suffer from an intractable anxiety and depression-like state. In some clinical studies, pain catastrophizing has been related to aggravation of pain itself.
This specific component can be supported by neuroanatomical connections between emotional-responding structures and brainstem areas involved in descending control of pain. Indeed, central nucleus of the amygdala (CeA) receives nociceptive inputs from lateral parabrachial area, and projects to the ventro-lateral periaqueductal grey (vlPAG). Moreover, it has been shown that vlPAG neurons can be controlled by CeA stimulation, and that CeA can induce analgesia after acute stress or opioid micro-injection into the basolateral amygdala, depending on vlPAG activity. Although involvement of CeA in analgesia on pain-induced reflexes is well known, few is reported about whether this implies descending controls and acts directly on spinal neurons.
In this study, we aimed to show that CeA influences the integration of nociceptive and tactile activities coming into the spinal cord. For this purpose, we tested the effect of pharmacological blockade of CeA by microinjection of $GABA_A$ receptor agonist muscimol, on extracellular recording of nociceptive and

tactile activity of WDR neurons recorded in the spinal cord. We have conducted these experiments on anesthetized rats.
As a result, WDR activity was decreased by intra-CeA muscimol, suggesting that CeA normal activity has a pro-nociceptive influence on WDR neurons. This is not consistent with analgesic effects of CeA on pain reflexes. According to us, this discrepancy is related to the activation of $GABA_A$ receptors which inhibit non specifically all networks in the CeA.
To verify this hypothesis, we are currently testing the effects of cholecystokinin (CCK) microinjection into the CeA on the same protocol, because CCK has been specifically related to pronociceptive effects. These results will provide key elements about how amygdala is involved in the control of pain and how it could be related to its chronicity in patients.

P2.154

Trigeminal pain-induced anxiety is associated with decreased activation of layer V pyramidal neurons in the anterior cingulate cortex

Ducret, E. (Clermont-Ferrand)[1], Jacquot, F. (Clermont-Ferrand)[1], Descheemaeker, A. (Clermont-Ferrand)[1], Dallel, R. (Clermont-Ferrand)[1], Artola, A. (Clermont-Ferrand)[1]
[1]Neuro-Dol, Inserm, Université d'Auvergne UMR 1107, Clermont-Ferrand, France

Pain is a sensory and affective experience that warns us of imminent or actual tissue damage. When pain becomes persistent, it is associated with emotional disorders such as anxiety or depression. The anterior cingulate cortex (ACC) is known to play a key role in the affective component of pain. Does it contribute to trigeminal pain-induced emotional disorders? We behaviorally, anatomically and electrophysiologically addressed this question in a rat model of lasting but reversible inflammatory pain induced by injecting complete Freund's adjuvant (CFA) into the vibrissae pad. CFA injection caused a reversible static mechanical allodynia: facial mechanical thresholds (von Frey filaments) decreased within 3-4 days and had returned to basal values 9 days after injection. Compared with controls (saline injection into the vibrissae pad), CFA rats showed higher levels of anxiety (elevated-plus maze or light-dark box) at 10-11 but not 3 days after injection. Concomitantly, c-fos immunoreactivity in ACC was increased 3 days after CFA injection. But, conversely, it was reduced 10-11 days after it, predominantly in layer V pyramidal neurons. Recording (whole-cell patch-clamp) from these ACC layer V neurons in acute slices from rats, 10 days after CFA or saline, we found that, compared with controls, ACC layer V neurons in CFA rats showed: (1) reduced excitability due to higher rheobase, (2) rightward shift in the stimulus intensity-evoked EPSC amplitude curve and (3) decreased frequency of spontaneous EPSCs, suggesting that the depressed excitatory synaptic transmission is due to reduced presynaptic release. Interestingly, orexin A-induced increase in spontaneous EPSC frequency was also reduced in CFA compared to baseline neurons. ACC layer V output pyramidal neurons receive inputs from midline thalamic nuclei, an important relay in pain pathway. Our data indicate that these neurons undergo pain-induced plastic changes outlasting pain and suggest that such changes might be associated with trigeminal pain-induced anxiety.

P2.155

Control of voluntary movements by a cerebellar zone of cortical sensorimotor convergence

Proville, R. (Paris)[1], Spolidoro, M. (Paris)[1], Guyon, N. (Paris)[1], Selimi, F. (Paris)[2], Isope, P. (Strasbourg)[3], Popa, D. (Paris)[1], Léna, C. (Paris)[1]
[1]Institut de Biologie de l'Ecole Normale Supérieure, Equipe Cervelet - S4.9, Paris, France, [2]Center for Interdisciplinary research in biology - College de France, Paris, France, [3]Institut des Neurosciences Cellulaires et Intégratives CNRS UPR 3212, Strasbourg, France

Sensorimotor interactions are crucially involved in perception and motor control. How and where this integration takes place in the brain is still matter of intense discussion. Here we identify a new area of functional sensorimotor integration in the cerebellum where cortical sensory and motor inputs converge at the cellular level. Stimulation of this area triggers motor cortex activation and controls ongoing movements. This convergence of sensory and motor cortical information in a specific set of cerebellar neurons that are reciprocally connected with the cortex calls for a reevaluation of the role of cerebellum in the integration of cortical sensorimotor information.

P2.156
Multi-sensory integration for looming visual stimuli

Ben Hamed, S. (Bron)[1], Clery, J. (Bron)[1], Guipponi, O. (Bron)[1], Wardak, C. (Bron)[1]
[1]*Centre de Neuroscience Cognitive, UMR 5229 - CNRS Equipe de Neurophysiologie des processus cognitifs, Bron, France*

The general multisensory framework assumes that maximum multisensory integration is observed when two sensory stimuli are presented at the same location (spatial congruence) and time (temporal congruence). This makes sense when considering visuo-auditive multisensory integration. It is for example important to associate the moving mouth with the speech being produced. The ecological relevance of spatial and temporal congruence for visuo-tactile integration is less straightforward. One rarely experiences simultaneously seeing and being touched by a mosquito on the face. A more common situation is that of seeing the mosquito approach and then feeling its bite on the skin. And indeed, visual objects approaching the body have a high probability of impacting the body and are thus predictive of tactile activation - potentially harmful tactile activations. Here, we provide psychophysical evidence for maximal multisensory integration when looming visual objects are predictive of the tactile stimulation rather than when both stimuli are simultaneous.
Subjects are submitted to a simple detection task. Subjects are required to fixate a central point within a tolerance window of 1° (controlled by a video eye tracker). One to three seconds following trial start, a visual, a tactile or a visuo-tactile stimulation is presented. At the end of the trial, subjects are requested to press a 'Yes' button on tactile stimulus detection and on a 'No' button otherwise. In order to maximize multisensory integration, we use very weak tactile stimuli to the face, and degraded dynamic 3D visual stimuli moving from the background towards the face. Two types of visuo-tactile trials are used: 1) visual stimulation is predictive of tactile stimulation (V_then_T, i.e. tactile follows visual) or 2) visual stimulation is simultaneous with tactile stimulation (V_with_T). On half of the trials, no tactile stimulation is presented (catch trials). We perform a signal detection theory analysis and we extract, for each subject, the d' associated with each sensory condition. Overall, d' on T only conditions are smaller than on V_with_T condition. Maximal d' are obtained for the V_then_T condition. We propose to interpret these findings in the context of the causal inference Bayesian framework.

P2.157
Imaging Po_2 in the awake mouse brain

Lyons, D. (Paris)[1], Parpaleix, A. (Paris)[1], Goulam Houssen, Y. (Paris)[1], Charpak, S. (Paris)[1]
[1]*INSERM U 603, CNRS UMR 8154, Université Paris Descartes, Laboratory of Neurophysiology and New Microscopies, Paris, France*

Two-photon phosphorescence lifetime microscopy (2PLM) has been recently used for depth-measurements of the oxygen partial pressure (Po_2) in the rodent brain [1]. In

capillaries of olfactory bulb glomeruli, 2PLM has also allowed simultaneous measurements of Po_2 and blood flow, and revealed the presence of erythrocyte-associated transients (EATs), i.e. Po_2 gradients associated with individual erythrocytes. We recently examined EAT properties in capillaries in the olfactory bulb of anaesthetised mice [2], and found that at rest, Po_2 at EAT peaks overestimates the mean Po_2 by 26 mm Hg. In addition, we found that Po_2 between two EAT peaks is at equilibrium with and thus reports Po_2 in the neuropil. During odour stimulation, a small Po_2 decrease was detected in capillaries, prior to the onset of functional hyperaemia. This demonstrates the existence of the initial dip in Po_2 at the level of capillaries. We conclude that imaging oxygen dynamics in capillaries provides a unique, non-invasive approach to map brain activity at a fine scale. New experiments in awake, head-fixed and unstressed mice, have allowed for the measurement of vascular and tissue Po_2 values in the non-anaesthetised brain.

[1] Lecoq J, Parpaleix A, Roussakis E, Ducros M, Goulam Houssen Y, Vinogradov SA, and Charpak S. *"Simultaneous two-photon imaging of oxygen and blood flow in deep cerebral vessels."* Nature Medecine 17, 893-898 (2011)

[2] Parpaleix A, Goulam Houssen Y, and Charpak S. *"Imaging local neuronal activity by monitoring Po_2 transients in capillaries."* Nature Medicine 19, 241-246 (2013)

P2.158

Data driven cortical parcellation of the early components of the visual evoked potential

Lio, G. (Lyon)[1,2,3], Boulinguez, P. (Lyon)[1,2,3]

[1]Université de Lyon, Lyon, France, [2]Université Lyon 1, Villeurbanne, France, [3]CNRS, UMR 5229, Centre de Neuroscience Cognitive, Bron, France

Assessing the time course of early visual processing in the human brain with non-invasive methods is a huge challenge. EEG has long been used for the benefits provided by its temporal resolution, but the spatial resolution of the technique is considered as fatally low. Traditionally, VEPs extraction relies on a simple averaging technique. This method has the huge advantages of removing from the signal most part of non stimulus time-locked activities and smoothing inter-individual anatomical variations of the human striate and extrastriate cortex. However, the resulting scalp topography remains a spatio-temporal mixing of many neuronal generators, making it impossible to disentangle the different operations involved in early visual processing.

Blind source separation (BSS), a data driven technique that separates brain source activities independently of their respective power, is intended to circumvent this issue. However, the method requires sophisticated data analysis and runs into the problem of group inferences. As a consequence, this potential solution has not been exploited so far for assessing VEPs generators. We did it in the present high density EEG study. A simple visual detection task was performed by 20 subjects. Group Independent Analysis (gICA) based on the spectral signatures of the sources (UWSOBI) was used to perform separation.

This method provided highly reliable and reproducible time courses and scalp topographies of the different generators of the VEPs, leading to accurate localization of segregated visually evoked cortical activities (figure 1).

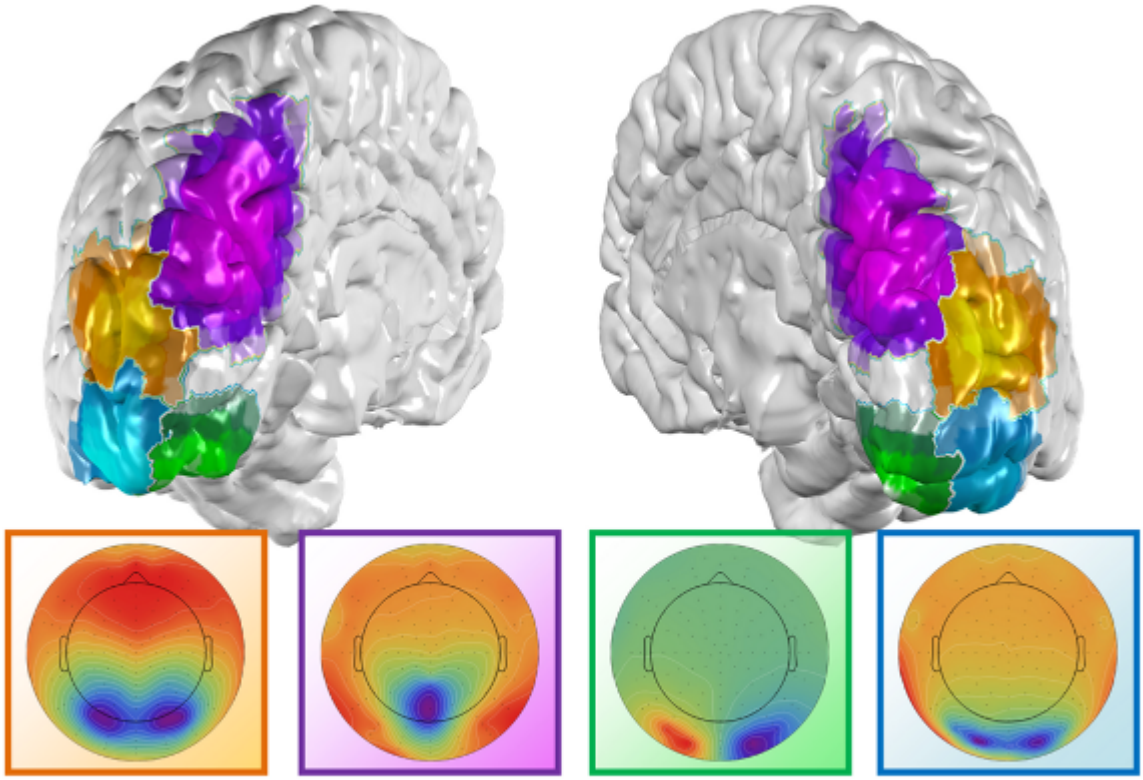


Figure 1: *Example of data driven parcellation of the visual cortex obtained from a simple visual detection task.*

[Figure 1]

P2.159

Sexual arousal, a role of histamine and orexins?

Zhao, Y. (Lyon)[1], Anaclet, C. (Lyon)[1], Perier, M. (Lyon)[1], Buda, C. (Lyon)[1], Sergeeva, O. (Düsseldorf)[2], Lin, J.-S. (Lyon)[1]

[1]CRNL, INSERM U 1028-CNRS UMR 5292, Univ. Cl. Bernard Lyon1, Physiologie intégrée du système d'éveil, Lyon, France, [2]Heinrich-Heine-University, Department of Neurophysiology, Düsseldorf, Germany

Sexual arousal ensures the necessary waking state, allowing anticipation and performance of sexual activities and so is a prerequisite for reproduction. We hypothesize that a full expression of such a behavioral state requires highly optimal convergent and divergent activities of the brain arousal systems notably histamine (HA) and orexins (Ox). To test our hypothesis, wild type (WT), histidine-decarboxylase (hdc)- and Ox- knockout (KO) mice were chronically implanted for EEG and sleep-wake monitoring under baseline conditions (12h light/dark cycle) and during the sexual arousal test, which consisted of introducing during 4h a female mouse into the habitual cage of a male mouse. The two mice were separated by a transparent plexiglass with a maximal number of holes, which allowed the physical contacts, but prevented their copulation. Placement of a male mouse was used as control. Sexual arousal is defined here as increased W in the male mouse facing the female. The test was performed both during light and dark phases. We found that the presence of a female mouse elicited in the male a significant increase in waking (+49 % vs baseline during lightness, n=32, p< 0.001). This increase is not significant if another male was introduced (+10% vs baseline, n=32,

p=0.2). This sexual arousal appears to depend on sex hormones because ovariectomized female mice or those pretreated with tamoxifen (an estrogen receptor antagonist) did not elicit significant sexual arousal in male animals and because male mice pretreated with flutamide (an androgen receptor antagonist) did not show any increased waking in the presence of a female. Secondly, acute application of α-FMH (a specific inhibitor of the HA synthesis) or SB-334867 (Ox1-receptor antagonist) both abolished sexual arousal in the male mice. Finally, whereas the sexual arousal appeared intact in hdc or Ox KO mice, it was totally absent on double KO mice lacking both HA and Ox. These data indicate that both the HA and Ox systems, most likely driven by sex hormones, participate in the promotion of sexual arousal and that chronic HA or Ox loss could be compensated by up-regulated adaptive mechanisms, which become ineffective when both HA and Ox were deficient.

P2.160

Neuroanatomical identification of the brainstem neurons suppressing the activity of hypoglossal motoneurons during paradoxical sleep in rats: consequences for obstructive sleep apnea syndrome

Brischoux, F. (Lyon)[1], El Khoueiry, C. (Lyon)[1], Valencia Garcia, S. (Lyon)[1], Libourel, P.-A. (Lyon)[1], Arthaud, S. (Lyon)[1], Fort, P. (Lyon)[1], Luppi, P.-H. (Lyon)[1]
[1]Centre de Recherche en Neuroscience de Lyon, CNRS UMR 5292 - INSERM U 1028, Equipe SLEEP, Lyon, France

Obstructive sleep apnea (OSA) is a common pathology in humans resulting in severe and deleterious consequences such as cardiovascular diseases or cognitive dysfunctions. The pathogenesis of OSA has been linked to the suppression of pharyngeal muscle activity during sleep, particularly during paradoxical sleep (PS) that leads to upper airway occlusion in individuals with narrow airways. The control of pharyngeal muscle activity by the hypoglossal motor nucleus is well described, but little is known about the neurobiological mechanisms that suppress the activity of hypoglossal motoneurons during PS.
PS is characterized by a sustained atonia of the whole skeletal musculature due to a tonic hyperpolarization of somatic motoneurons by glycine. Since the suppression of pharyngeal muscle activity occurs in parallel with the typical atonia of postural muscles during PS, it might be also due to glycinergic inhibitory mechanisms of hypoglossal motoneurons.
To localize and confirm the phenotype of the neurons responsible for the inactivation of hypoglossal motoneurons during PS, we carried out a series of experiments combining retrograde tract tracing following cholera toxin subunit b (CTb) iontophoretic injections into the hypoglossal motor nucleus, Fos expression and in situ hybridization of GlyT2 mRNA (the neuronal glycine reuptake transporter) in rats after specific PS deprivation or PS rebound. To verify if the neurons identified act specifically on the hypoglossal motoneurons or mediate inhibition of other motor systems during PS, we performed double retrograde tract tracing following CTb and Fluorogold injections, respectively into the hypoglossal and trigeminal motor nuclei of the same rats.
Our preliminary data suggest that the lateral paragigantocellular nucleus (LPGi) contains the main glycinergic premotoneuron pool in position to inhibit the hypoglossal motoneurons during PS. These neurons may form a distinct population of premotoneurons from those acting on trigeminal motor nucleus that are mostly located within the raphe magnus nucleus.
Altogether, these results suggest that the cranial and spinal motoneurons may be under the control of distinct, topographically organized populations of inhibitory premotoneurons specifically active during PS.

P2.161

Central TSH restores a summer phenotype of hypothalamic RF-amides and reproductive status in photoinhibited mammals

Simonneaux, V. (Strasbourg)[1], Sebert, M.-E. (Strasbourg)[1], Kamontip, R. (Amphur Khlongluang)[2], Laran-Chich, M.-P. (Strasbourg)[1], Klosen, P. (Strasbourg)[1,3]
[1]CNRS 3212, INCI, Strasbourg, France, [2]Thammasart University, Amphur Khlongluang, Thailand, [3]Univ. de Strasbourg, Strasbourg, France

In mammals, melatonin is the pivotal messenger synchronizing biological functions, notably reproductive activity, with annual daylength changes. Two major findings recently clarified melatonin´s impact on reproductive activity. First, melatonin was found to control the production of thyroid stimulating hormone (TSH) by the pars tuberalis. This TSH regulates the local thyroid hormone levels within the mediobasal hypothalamus through tanycyte deiodinase expression. In parallel, we found that two recently discovered hypothalamic RF-amide peptides regulating gonadotropic activity, kisspeptin and RFRP-3, are also involved in the melatonin control of reproduction. Here we demonstrate that central administration of TSH in short-day adapted Djungarian and Syrian hamsters restores the summer phenotype of both reproductive activity and kisspeptin and RFRP expression. Our data thus reveal a mechanistic link between the melatonin-driven TSH and the RF-amide control of reproductive activity. Furthermore, this TSH treatment of lean short-day adapted Djungarian hamsters increased body weight and reduced hypothalamic somatostatin expression to summer phenotype. In conclusion, our study demonstrates the pivotal role of melatonin-driven TSH for the seasonal regulation of reproduction and body weight, and uncovers the neuropeptides relaying this signal within the hypothalamus.

P2.162

Identification of microglial genes differentially expressed in aged versus young mice

Repiquet Paire, L. (Lyon)[1], Lieury, A. (Lyon)[1], Pays, L. (Lyon)[1], Nataf, S. (Lyon)[1]
[1]*Centre de Recherche en Neuroscience de Lyon (CRNL), UMR 5292 CNRS/U1028 INSERM, Centre de Recherche en Neurosciences de Lyon, Neurooncology Neuroinflammation Team, Lyon, France*

Background: Normal aging is associated with loss of cognitive functions and reduced neuronal plasticity. Among the many mechanisms possibly involved in such a process, it is now recognized that cells and molecules of the immune system are likely to play an important role. Incidence of neurodegenerative diseases increases with age and brain aging is itself accompanied by a switch of microglial functions toward a pro-inflammatory profile. It is thus considered that a low-grade chronic neuroinflammation develops with normal aging likely and contributes to the susceptibility of elderly people to neurodegenerative disorders. As human life span continue to readily increases, the need to identify mechanisms of age-related neuroinflammation is crucial in order to design new treatments that may ensure healthy aging and reduced health care costs.
Objectives and results: in this context, the present study was aimed at identifying a molecular signature of brain senescence in microglia. For that purpose, a method so-called virtual microdissection was applied to publically available transcriptomic data obtained from the brain of aged vs young mice. Briefly, this approcah consisted in the *in silico* identification of myeloid genes that are co-regulated with cannonical microglial genes, such as Iba-1, and are therefore likely to be expressed by microglia. We identified a set of myeloid genes which expression in the aged mice brain is currently assessed by *in situ* hybridization and qPCR analysis.

P2.163

Distribution of the MCH (melanin-concentrating hormone)-system in the pig (*Sus scrofa domesticus*)

Risold, P.-Y. (Besançon)[1], Chometton, S. (Besançon)[1], Houdayer, C. (Besançon)[1], Fellmann, D. (Besançon)[1], Tillet, Y. (Nouzilly)[2]
[1]*Univ. de Franche-Comté, Faculté de Médecine, EA 3922, Besançon, France,* [2]*INRA/CNRS/Université François-Rabelais Tours, UMR Physiologie de la Reproduction et des Comportements, INRA/CNRS, Nouzilly, France*

MCH neurons are described in the posterior hypothalamus of all investigated vertebrates analyzed so far. However, the fine distributions of their perikarya and projections greatly diverge even among mammals. For example neuronal soma are dorsomedial in human and cat, dorsolateral in mouse or rat, ventrolateral in sheep. Few other species have been studied until now, although divergence in the distribution of this peptide may show that corresponding neurons are involved in species-specific functions.
We report here the distribution of the MCH system in the domestic pig, which is a monogastric omnivorous, but largely vegetarian species. MCH perikarya are exclusively observed in the caudal hypothalamus, mostly in the ventral perifornical region. Projections are detected in numerous brain regions, including the lateral hypothalamus, subthalamic nucleus and ventral telencephalon, especially the medial septal region. Abundant projections are observed in the cerebral cortex, but these projections are much differentiated as they are abundant in the entorhinal cortex and hippocampus, somewhat abundant in piriform, peri-insular and cingulate cortical fields, but almost absent in most other sensory and motor fields. Caudal projections are abundant in the reticular formation, tectum and periaqueductal gray.
As a conclusion, the distribution of MCH cell bodies recalls that of the sheep, but they are slightly more medial. Divergences in their distribution between mammalian species may be related to specific behavioral repertories, for example between vegetarian preys or predatory animals. Projections are widely distributed as in most other species, but their distribution in the pallium seems to indicate that MCH may be much more involved in influencing association cortices and olfactory fields, but less in other sensory motor integrations.

P2.164

Complex c-Fos patterns in the posterior hypothalamus related to wake, exploratory or feeding behaviors

Chometton, S. (Besançon)[1], Franchi, G. (Besançon)[1], Houdayer, C. (Besançon)[1], Poncet, F. (Besançon)[1], Mariot, A. (Besançon)[1], Laroche, A. (Besançon)[1], Fellmann, D. (Besançon)[1], Risold, P.-Y. (Besançon)[1]
[1]*Université de Franche-Comté, UFR Sciences Médicales et Pharmaceutiques, EA 3922, Histologie, SFR FED 4234, Besançon, France*

The hypothalamus is a complex structure involved in multiple functions related to the expression of goal oriented behaviors such as feeding, defensive or reproductive behaviors, as well as emotion, reward or sleep/wake cycle. Regions of the hypothalamus are differentially involved in coordinating these responses. However, an integrated morpho-functional organization of the hypothalamus is not yet clear.
In this work, we analyzed the hypothalamic expression of the c-Fos protein by immunohistochemistry in nine experimental conditions related to wake, exploration, attention, appetitive responses and food consumption.
To date, c-Fos-labeled nuclei were counted in eleven structures of the periventricular, medial and lateral posterior hypothalamus. Different patterns of c-Fos expression affecting these hypothalamic regions were correlated to each experimental condition.
These results provide a first, yet incomplete, comprehensive view of the morpho-functional organization of the hypothalamus. They will be completed by analysis in anterior regions of the hypothalamus and correlated to anatomical data.

P2.165

An emerging role of miR-383 and miR-488 in the central control of energy homeostasis by the melanocortin system

Derghal, A. (Marseille)[1], Djelloul, M. (Marseille)[1], Dallaporta, M. (Marseille)[1], Airault, C. (Marseille)[1], Tardivel, C. (Marseille)[1], Bariohay, B. (Marseille)[2], Rouquet, T. (Marseille)[1,2], Bonnet, M. (Marseille)[1], Lebrun, B. (Marseille)[1], Trouslard, J. (Marseille)[1], Troadec, J.-D. (Marseille)[1], Mounien, L. (Marseille)[1]
[1]*Aix Marseille Université - Faculté des Sciences, PPSN - Unité EA4674, Marseille, France,*
[2]*Biomeostasis, Faculté des Sciences et Techniques de St Jérôme - B52, Marseille, France*

The central nervous system continuously monitors modifications in metabolic parameters (blood glucose level) or hormone levels (leptin and insulin) and elicits adaptive responses such as food intake

regulation or autonomic nervous system modulation. More particularly, within the hypothalamus and the dorsal vagal complex (DVC), pro-opiomelanocortin (POMC) neurons are critical regulators of energy balance. In mice, consistent with a pivotal role of the melanocortin system in the control of energy homeostasis, disruption of the *Pomc* gene causes hyperphagia and obesity. MicroRNAs (miRNAs) are short noncoding RNA molecules that post-transcriptionally repress the expression of genes by binding to 3´-untranslated regions (3'-UTR) of the target mRNAs. However, little is known regarding the role of miRNAs in the central control of energy homeostasis by the melanocortin pathway. The aim of our project is to demonstrate that miRNA are involved in the integration of signals generated by nutritional status modification. In the first part of this study, we used common prediction programs to search for potential miRNAs target sites on 3'-UTR of Pomc mRNA. This screening identified conserved miRNA seed sequences for miR-383, miR-384, miR-485 and miR-488. In leptin-deficient mice (Ob/Ob), we observed an up-regulation of miR-383, miR-384, miR-485 and miR-488 in the hypothalamus, whereas only the expression of miR-383 and miR-488 increased in the DVC. In C57BL/6 mice, chronic (4d) intracerebroventricular administration of leptin increased Pomc mRNA level in the hypothalamus whereas there was a decrease of miR-488 expression in this structure. In addition, in 24h-fasted mice, the decrease of Pomc mRNA level in the hypothalamus was associated with an increase of miR-383 expression. In summary, these results support the hypothesis that changes in the miRNA network may contribute to a defect in the control of energy homeostasis by the melanocortin system.

P2.166

Regulatory T cells modulate disease progression in a murine model of Alzheimer's disease

Dorothée, G. (Paris)[1], Toly-Ndour, C. (Paris)[1], Dansokho, C. (Paris)[1], Aid, S. (Paris)[2], Ait-Ahmed, D. (Paris)[1], Chaigneau, T. (Paris)[1], Holzenberger, M. (Paris)[2], Aucouturier, P. (Paris)[1]
[1]*INSERM UMRS 938, Hôpital Saint-Antoine, Laboratory 'Immune System and Conformational Diseases', Paris, France,* [2]*INSERM UMRS 938, Hôpital Saint-Antoine, Laboratory 'Pathophysiology of IGF-1 Signaling in Development, Aging and Regeneration', Paris, France*

Vaccines targeting Aβ represent promising strategies for the treatment of Alzheimer´s disease (AD). Immunization approaches provided encouraging results in mouse models and a subsequent human clinical trial (AN1792). However, whereas preclinical studies in murine models did not show evidence of T cell-related side effects, the AN1792 trial had to be interrupted due to the development of meningoencephalitis attributed to pro-inflammatory T cell responses in 6% of the patients. Several reports also suggest that Aβ-specific $CD4^+$ T cells may be implicated in the pathophysiology of AD and could have a strong therapeutic potential as well, further supporting the need for better understanding the role and regulation of T cell responses to Aβ. We previously showed that regulatory T cells (Tregs) critically control the magnitude of Aβ-specific $CD4^+$ T cell responses in both physiological and pathological settings in response to vaccination. However, the actual role of Tregs in the pathophysiology of AD remains unknown. We analyzed the impact of Tregs on disease progression in a murine model of AD. APPPS1 mice were depleted of Treg cells by injecting anti-CD25 antibodies, and the impact of Treg depletion on the neuropathology and cognitive deficits was evaluated. Depletion of Treg cells accelerated the onset of cognitive deficits in APPPS1 mice. Alteration in spatial memory was detected starting from 7 months in Treg-depleted animals, while PBS-treated APPPS1 mice were not yet cognitively impaired as compared to wt animals. Early cognitive impairment in Treg-depleted mice was correlated with alterations in the neuroinflammatory response that is associated with disease progression. Our results suggest that Treg cells play a beneficial role in the pathophysiology of AD and delay disease progression in a murine model of the disease. These data open new perspectives in the development of Treg-based innovative immunotherapy approaches for the treatment of AD.

P2.167

Transcriptomic analysis of blood-derived macrophages identifies 5-lipoxygenase activation protein as a key tumor-induced immune molecule in glioma patients

Numan, A. (Lyon)[1], Wierincks, A. (Lyon)[2], Chanal, M. (Lyon)[3], Rey, C. (Lyon)[3], Champier, J. (Lyon)[2], Fevre-Montange, M. (Lyon)[2], Honnorat, J. (Lyon)[3], Nataf, S. (Lyon)[4]
[1]Université de Lyon1, INSERM U 1028 UMR 5292, Lyon, France, [2]Centre de Recherche en Neuroscience de Lyon (CRNL), UMR 5292 CNRS/U 1028 INSERM, Lyon, France, [3]Centre de Recherche en Neuroscience de Lyon, Lyon, France, [4]Centre de Recherche en Neuroscience de Lyon (CRNL), Lyon, France

In glioblastoma (GB), as in many types of tumors, infiltrating macrophages so-called tumor-associated macrophages (TAM), engage a peculiar activation program that dampens the immune response, favors tumor outgrowth and supports neovascularization. Such a mechanism of immune escape is considered as responsible, at least in part, for the overall poor efficiency of immunotherapies in GB patients. Thus, major efforts are currently developed to decipher the mechanisms of immunosuppression and immunosubversion triggered by GB cells. In this context, our study was aimed at identifying a blood molecular signature of innate immune alterations in the monocyte/macrophage lineage of GB patients. For that purpose, we performed a transcriptomic analysis of blood-derived macrophage cultures obtained from 8 GB patients and 12 healthy control subjects. Using this approach, we identified as set of 14 immune-related genes which expression was up-regulated in blood-derived macrophages from GB patients. Interestingly, a majority of these genes was previously identified as exerting immunosuppressive functions in macrophages and, in particular, TAM. One of these genes, 5-Lipoxygenase activating protein (Alox5-AP), a major component of the leukotriene biosynthesis pathway, was further assessed for its intratumoral expression and putative role in GB immune escape. Transcriptomic analysis of GB tumors showed that Alox5-AP co-regulated with 5-lipoxygenase, a key enzyme of the leukotriene biosynthesis pathway, along with several myeloid genes known to confer immunosuppressive functions in TAM. In contrast, such a co-regulation was not observed in microarray data obtained from meningioma tissue. Finally, we demonstrated that under *in vitro* conditions of macrophages/GB cells interactions, pharmacological inhibition of Alox5-AP induced a striking up-regulation of both HLA-DR and MHC-class II transactivator gene expression in macrophages. Overall, our work unravels a new mechanism of GB immune escape involving activation of the leukotriene synthesis pathway in TAM.

P2.168

Dopamine and zebrafish reproduction

Fontaine, R. (Gif sur Yvette)[1], Affaticati, P. (Gif sur Yvette)[1], Yamamoto, K. (Gif sur Yvette)[1], Vernier, P. (Gif sur Yvette)[1], Baloche, S. (Paris)[2], Gonnet, F. (Paris)[2], Jolly, C. (Paris)[2], Dufour, S. (Paris)[2], Pasqualini, C. (Gif sur Yvette)[1]
[1]Laboratoire Neurobiologie et Développement, UPR 3294, Institut de Neurobiologie Alfred Fessard, Gif sur Yvette, France, [2]Laboratoire Biologie des Organismes marins et Ecosystèmes, UMR BOREA CNRS 7208, Muséum National d'Histoire Naturelle, IRD 207-UPMC, Paris, France

In many teleosts, the stimulatory control of the gonadotrope axis by GnRH, is opposed by an inhibitory control by dopamine (DA). The functional importance of this DA inhibition differs widely according to the teleost species. Zebrafish (Danio rerio) is a vertebrate model commonly used, but the role of DA in the neuroendocrine control of its reproduction is still unknown.
We evaluated in old, sexually regressed, females, the effects of treatments with either a DA D2 receptor antagonist (Domperidone), a GnRH agonist (GnRHa), or both in combination. Only the double treatment (GnRHa + Domperidone) was able to induce a significant increase in pituitary LHβ mRNA level, and to stimulate ovarian vitellogenesis, indicating that in old females removal of DA inhibition is required for the stimulatory action of GnRH and reactivation of ovarian function to occur. The same double treatment (GnRHa + Domperidone) was also able to increase LHβ mRNA level in young cycling females, showing that the role of DA in the control of LH is not restricted to ageing.

Using double immunofluorescent staining on pituitary, we showed in this species the innervation of LH cells by tyrosine-hydroxylase (TH) immunoreactive fibers. Then, using in situ hybridization and immunofluorescence (IF), we showed that the three subtypes of zebrafish DA D2 receptors (D2a, D2b, and D2c) were expressed in LH-producing cells, suggesting that they all may be involved in mediating this inhibition. Using retrograde tracing coupled to IF, we precisely localized the DA cell bodies projecting to the pituitary within the PreOptic Area (POA). We finally studied the ontogeny of these preoptico-hypophyseal DA neurons in embryos and observed that they appear between 60 and 72 hours, thus much later than other DA populations.
These results show that, in zebrafish, DA has a direct and potent inhibitory action capable of opposing the stimulatory effect of GnRH in the neuroendocrine control of reproduction, and provide the first developmental basis of this control.

P2.169

New molecular targets upon the control of the serotonin 4 receptors involved in anorexia

El Ouahli, M. (Béni Mellal)[1], Jean, A. (Montpellier)[2], Chigr, F. (Béni Melllal)[3], Najimi, M. (Béni Melllal)[3], Compan, V. (Montpellier)[2]
[1]Université Sultan Moulay Slimane, Faculté des sciences et techniques, Béni Mellal, Morocco, [2]Institue de génomique fonctionnelle, Montpellier, France, [3]Université Sultan Moulay Slimane, Faculté des sciences et techniques Béni Mellal, Biologie, Béni Melllal, Morocco

Even if eating is essential to survival, highly stressed adolescents affected by Anorexia nervosa can restrict their caloric intake to lethal, levels. Hunger *i.e.* the physiological need to eat is upon a cerebral autonomic regulation. Under stress, the neural control of decisions to eat appears to prevail over the autonomic control of hunger. When this response is transient, survival is not compromised and this is adaptive. In rodents, great stress *i.e.* a forced immobilization, also named the restraint stress, transiently reduces food intake (*e.g.* hypophagia). If this response persists despite mounting energy requirements, this is a valid animal model for Anorexia nervosa. Here, we show attenuated hypophagia following chronic stress in male mice lacking the serotonin 4 receptors (5-HTR_4 KO), consistently with previous studies. We further discovered that decision-making to eat following stress depends on the 5-HTR_4, specifically located in the medial prefrontal cortex. Since maladaptive decision to eat could prevail over the autonomic nervous system under stress, we set out to research how the 5-HTR_4 could influence the activity of the hypothalamus following stress. An absence of increase in pCREB/CREB ratio was found in the hypothalamus of stressed 5-HTR_4 KO mice, suggesting potential disruption of gene expression because pCREB is a transcription factor. Using transcriptom and RQ-PCR analyses, new influences of the 5-HTR_4 on hypothalamic targets involved in neurogenesis and in DNA methylation were found in stressed mice. Consistently with other of our results, present findings bring out new neural molecular pathways involved in eating disorders following stress.

P2.170

Impact of alcohol withdrawal on working memory and brain chromatin remodeling in mice

Dominguez, G. (Tours)[1], Dagnas, M. (Bordeaux)[2], Henkous, N. (Bordeaux)[2], Beracochea, D. (Bordeaux)[2], Mons, N. (Bordeaux)[2]
[1]Inserm U930 Imagerie et Cerveau, Université de Tours, Tours, France, [2]Institute de Neurosciences Cognitives et Intégratives d'Aquitaine (INCIA), Bordeaux, France

Alcohol withdrawal occurring after chronic alcohol exposure induces memory disorders, alters brain chromatin structure and regulation of CREB-mediated transcription through alterations of histone modification patterns (e.g., acetylation). This study investigated the potential contribution of deregulated histone H4 acetylation produced by short (1 week, 1W) or long (6 weeks, 6W) alcohol withdrawal periods on working memory (WM) deficits using a sequential alternation task in C57BL/6 mice.
We first showed that alcohol withdrawal was associated with a significant impairment of WM both in withdrawn 1W and 6W mice as compared to water controls. Imaging analyses further indicated that WM testing promoted histone H4 acetylation and CREB phosphorylation in the dorsal hippocampus (dHPC) and the prefrontal cortex (PFC) of water control mice -two brain areas critically involved in WM-, but such increases were not observed both in 1W and in 6W mice.
Previous evidence indicates that increasing cAMP/PKA/CREB pathway within the PFC and HPC exerts opposite effect on WM. Thus, we compared next the behavioral and molecular consequences of pharmacological enhancement of CREB function, either by acute treatment with the phosphodiesterase inhibitor Rolipram or local intra-dHPC or intra-PFC infusion of the PKA activator Sp-AMPc, before testing in water and withdrawn mice. Strikingly, in both cases, increasing CREB activity induced bidirectional cognitive effects in water control group (memory impairment) and in withdrawn groups (memory improvement). On the other hand, intra-dHPC infusions of Sp-AMPc did not affect memory in all groups.
In parallel, imaging analyses revealed that, compared to matched vehicle-treated water control groups, acute Rolipram treatment increased (in water-controls) or fully rescued (both in 1W and 6W mice) H4 acetylation specifically in the PFC, but not in dHPC.
In conclusions, these results highlight the central role played by epigenetic disturbances in the PFC in the genesis of WM impairments resulting from alcohol withdrawal and further suggest that enhancers of the cAMP/PKA signaling pathway could rescue memory dysfunction resulting from alcohol withdrawal.

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Opiate dependence induces network state shifts in the limbic system

Dejean, C. (Bordeaux)[1,2], Boraud, T. (Bordeaux)[3,4], Le Moine, C. (Bordeaux)[1,2]
[1]Univ. Bordeaux, INCIA, UMR 5287, Bordeaux, France, [2]CNRS, INCIA, UMR 5287, F-33000, Bordeaux, France, [3]Univ. Bordeaux, IMN, UMR 5293, F-33000, Bordeaux, France, [4]CNRS, IMN, UMR 5293, F-33000, Bordeaux, France

Among current theories of addiction, hedonic homeostasis dysregulation predicts that the brain rewarding systems, particularly the mesolimbic dopamine switch from a physiological state to a new “set point”. In opiate addiction, evidence show that its principal targets, prefrontal cortex (PFC), nucleus accumbens (NAC) and basolateral amygdala complex (BLA) also adapt to repeated drug stimulation. Here we investigated the impact of chronic morphine on the dynamics of the network of these three interconnected structures. We used simultaneous electrophysiological recordings in the PFC NAC and BLA in freely-moving rats implanted with continuous-release subcutaneous morphine or placebo pellets, combined with multivariate analysis methods to characterize network states under physiological conditions and after chronic morphine.
Behavioral and electrophysiological activities (LFP and single units) were recorded in 13 awake rats (morphine-treated rats, n=8, placebo-treated rats, n=5) during 20 minute sessions. Recordings took place before (baseline) and after morphine/placebo pellets implantation on days 1, 2, 3 and 4. In addition similar experimental sessions were carried on day 5 (saline injection), day 6 (naloxone) and day 7 (naloxone + 24h).
Spectral analysis during alert behavioral state showed that the PFC, NAC and BLA synchronously oscillate in three fundamental frequency bands: delta (1-4Hz), theta (5-10Hz) and gamma (40-100Hz). Chronic morphine produced a shift in the network state underpinned by changes in delta and gamma oscillations in the LFP of PFC, NAC and BLA. However despite continuous stimulation by the drug, an apparent normalization of network activity and state occurred after two days indicating large scale adaptations. Blockade of μ opioid receptors was nonetheless sufficient to disrupt this acquired new stability in morphine-dependent animals. In line with the homeostatic dysregulation theory of addiction,

our study provides original direct evidence that the PFC-NAC-BLA network of the dependent brain is characterized by a *de novo* balance for which the drug of abuse becomes a main contributor.

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Modulation of the motor system during singing-voice perception

Lévêque, Y. (Bron)[1,2], Schön, D. (Marseille)[3]
[1]INSERM/CNRS, Centre de Recherche en Neurosciences de Lyon, Bron, France, [2]Laboratoire Parole et Langage, Aix-en-Provence, France, [3]Institut de Neurosciences des Systèmes, Marseille, France

Although there has been great interest in how motor representations are involved in speech perception and comprehension, several questions remain unsolved concerning the functional role and the conditions of motor resonance in auditory perception. In the present work, we investigated the perception of a singing voice, a stimulus that is not primarily linked to articulatory processes. Does listening to a singing voice induce an activity of the motor system? Is this motor activity stronger for voice than for a non-biological musical sound?
Participants performed two perceptual tasks in an fMRI scanner. In the first task, they passively listened to short 5-note melodies, and were sporadically asked to judge the familiarity of a target-melody. In the second task, the same melodies were presented with the instruction to be prepared to sing as soon as a go signal would appear. In both tasks, melodies were played with a natural vocal timbre or with a computer-generated complex timbre.
A functional connectivity analysis (Psychophysiological Interactions, PPI) revealed that voice stimuli presentation in task 1 was linked to an increased connectivity between the right vocal motor cortex and the primary auditory cortex and the area Spt at the parietal-temporal boundary in the left hemisphere.
Furthermore, vocal melodies elicited significantly more activation within the lateral premotor cortex than computer-generated melodies, possibly due to a better matching between the perceived sound and internal motor representations required to sing the melodies back or memorize them.
Overall, our results suggest that perception of a singing voice without linguistic information entails a subtle motor activity, modulated by the biological dimension of the sound.

P2.173

Environmental enrichment duration differentially affects behaviour and neuroplasticity in adult mice

Freret, T. (Caen)[1], Léger, M. (Caen)[1], Paizanis, E. (Caen)[1], Dzahini, K. (Strasbourg)[2], Quiédeville, A. (Caen)[1], Bouet, V. (Caen)[1], Schumann-Bard, P. (Caen)[1], Boulouard, M. (Caen)[1], GMPc - EA4259
[1]Université de Caen Basse-Normandie, GMPc Groupe Mémoire et Plasticité comportementale, Caen, France, [2]Laboratoire d'Imagerie et de Neurosciences Cognitives - Université de Strasbourg, Strasbourg, France

Environmental enrichment constitutes an interesting model to elicit cerebral and behavioural plasticity in rodent. Learning and memory performances are known to be enhanced by this housing condition. However, the onset and the time course of such beneficial effects are not well described. We thus aimed to assess whether different durations of enriched condition (EC) may differentially influence behavioural plasticity, especially regarding long-term memory, as well as neurobiological plasticity such as neurogenesis and neurochemical changes. To this end, adult male mice were housed in standard condition (SC) or EC for 1 day, 1, 3 or 5 weeks before behavioural and neurobiological

assessments. Memory performances were tested in the passive avoidance task and in a novel object recognition paradigm. The hippocampal neurogenesis was investigated through immunohistochemistry methods whereas cerebral monoamines levels were examined by high performance liquid chromatography. Behavioural assessment shows a transient improvement of aversive memory performances after a 3-week exposure to EC. By contrast, early and stable improvement of object recognition memory was observed in EC mice as soon as after 1 day of EC. While EC did not modify hippocampal cell proliferation, the 5-week EC exposure enhanced neuronal survival. Finally, a transient higher serotonin level was found in the frontal cortex of 3-week EC mice compared to respective SC ones. Taken together, these findings suggest that different mechanisms underlie the various behavioural effects of EC. The onset of behavioural changes in EC animals did not correspond to the onset of neurogenesis changes, suggesting the involvement of other neuroplasticity mechanisms. By contrast, the transient aversive memory improvement after 3-week EC exposure could be underlie by a transient modulation of the serotonergic system in the frontal cortex, a brain region previously reported to be involved in the beneficial effects of EC on aversive memory. Finally the critical influence of EC exposure duration on behaviour and neurobiological changes could explain the heterogeneity of literature's data.

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Sleep architecture and homeostasis of MCH-ataxin-3 transgenic mice

Varin, C.G. (Lyon)[1], Arthaud, S. (Lyon)[1], Gay, N. (Lyon)[1], Luppi, P.-H. (Lyon)[1], Léger, L. (Lyon)[1], Fort, P. (Lyon)[1]
[1]*Centre de Recherche en Neuroscience de Lyon (CRNL), UMR 5292 CNRS/U1028 INSERM, Équipe Physiopathologie des réseaux neuronaux du cycle veille-sommeil, Lyon, France*

Evidence supports a role of hypothalamic neurons secreting melanin-concentrating hormone (MCH) in the regulation of vigilance states. They indeed densely innervate brain areas involved in promotion of Waking (W), Slow Wave Sleep (SWS) and Paradoxical Sleep (PS). They strongly express Fos during sleep rebound (mainly composed of SP) that follows selective PS deprivation in rats. Juxtacellular unit recordings across the natural sleep-waking cycle in rats showed that neurochemically-identified MCH neurons increase firing during SWS and discharge maximally during PS. Intracerebroventricular MCH infusion induces a strong increase of SWS (+70%) and PS (+200%) amounts whereas systemic administration of MCH-R_1 receptor antagonists suppresses SWS and PS. To decipher the precise contribution of MCH neurons to SWS and/or PS regulations, we studied spontaneous sleep and its homeostatic regulation in transgenic mice with a selective loss of MCH neurons using ataxin-3-mediated ablation strategy.
We first determined that hypothalamic MCH mRNA expression was markedly reduced (-70%) in MCH-ataxin-3 compared to WT mice (male, C57BL/6, 16-20 weeks). Further, ≈30% of MCH-immunostained cell bodies were ablated (*vs* WT), whereas remaining ones displayed an altered morphology. An obvious decrease in brain density of MCH-immunoreactive axons was also noticed. The baseline polysomnographic recordings revealed that MCH-ataxin-3 mice exhibited a pronounced SWS fragmentation (reduced bout duration) compared to WT. Regarding PS, amounts were increased during sleep period (light on) but decreased during active period (light off). Transgenic mice were then submitted to either a total sleep (4h) or PS-selective deprivation (24h). In both cases, sleep homeostasis was impaired during recovery period (*vs* WT) with a delayed and partial refund of sleep debt induced by the deprivation. Modifications in dynamic processes of SWS maintenance and PS promotion were also observed (*vs* sleep-deprived WT mice).
Although preliminary, the present results indicate that MCH-expressing neurons may play a key role in the fine tuning of the sleep-waking cycle and in sleep homeostatic processes, by consolidating SWS and/or facilitating transitions from SWS to PS when sleep needs increase.

P2.175

Juvenile high-fat diet consumption enhances amygdala plasticity and emotional memory: involvement of HPA axis dysregulation

Boitard, C. (Bordeaux)[1], Maroun, M. (Haifa)[2], Tantot, F. (Bordeaux)[1], Cavaroc, A. (Bordeaux)[1], Sauvant, J. (Bordeaux)[1], Castanon, N. (Bordeaux)[1], Darnaudery, M. (Bordeaux)[1], Laye, S. (Bordeaux)[1], Vouimba, R.M. (Bordeaux)[3], Ferreira, G. (Bordeaux)[1]
[1]NutriNeuro, INRA - Bordeaux University, UMR 1286, Bordeaux, France, [2]Department of Neurobiology, University of Haifa, Haifa, Israel, [3]INCIA, CNRS- Bordeaux University, UMR 5287, Bordeaux, France

The current obesity pandemic is directly linked to overconsumption of high-fat diet (HFD). In addition to metabolic and cardiovascular disorders, obesity is associated with adverse cognitive and emotional outcomes as well as dysregulation of the stress hypothalamic-pituitary-adrenal (HPA) axis. The growing prevalence of obesity during adolescence is a major concern since this period is crucial for the maturation of both HPA axis and brain structures, like amygdala, involved in shaping adult cognitive and emotional processes. In this study, we investigate the impact of juvenile HFD consumption (jHFD, from weaning to adulthood, i.e. covering adolescence) on amygdala-dependent plasticity/memory and stress reactivity as well as the causal link between the two. Our results show that jHFD intake enhanced amygdala-dependent emotional memories, assessed through conditioned odor aversion and cued fear conditioning. Synaptic plasticity in the basolateral amygdala (BLA), evaluated by long-term potentiation in the pathway from entorhinal cortex to BLA was also enhanced in jHFD. Since high levels of glucocorticoids are known to enhance amygdala-dependent plasticity and memory, we measured plasma corticosterone levels. After acute psychogenic or systemic stressors, jHFD consumption induced a prolonged corticosterone release and enhanced BLA activation (using c-Fos and Egr-1 immunohistochemistry). Yet, jHFD intake did not modify anxiety-like behaviours, adrenal weight or amygdala GR expression. Interestingly, similar duration of HFD consumption restricted to adulthood was without consequences on either aversive memory or corticosterone release and amygdala activation after acute stressor. Finally, injection of a GR antagonist reversed the enhancement of both emotional memory and BLA plasticity induced by jHFD consumption. These results demonstrate that adolescence represents a vulnerable period to the effect of HFD consumption and suggest jHFD intake dysregulates the HPA axis which subsequently enhances the amygdala functions at both behavioral and synaptic levels.

P2.176

Effect of juvenile high-fat diet consumption on dopamine reward system: behavioral evidence and potential role of glucocorticoids

Tantot, F. (Bordeaux)[1], Boitard, C. (Bordeaux)[1], Naneix, F. (Bordeaux)[2], De Smedt-Peyrusse, V. (Bordeaux)[1], Pape, J.-R. (Bordeaux)[2], Marchand, A. (Bordeaux)[2], Layé, S. (Bordeaux)[1], Coutureau, E. (Bordeaux)[2], Ferreira, G. (Bordeaux)[1]
[1]NutriNeuro, INRA - Bordeaux University, UMR 1286, Bordeaux, France, [2]INCIA, CNRS - Bordeaux University, UMR 5287, Bordeaux, France

Increased consumption of palatable food, especially high-fat diet (HFD), participates in the current obesity pandemic. Recent evidence from human and animal models indicates that HFD-induced obesity is associated with alterations of the brain reward circuitry, in which dopamine (DA) system plays a crucial role. Since major changes in this system occur during the adolescence period and that a growing population of adolescents are obese, it might be hypothesized that juvenile HFD consumption altered the normal DA-related neurocognitive maturation. In the present study, we therefore investigated in rats the effects of HFD consumption from weaning to adulthood on behaviors supported by the DA system, namely: 1- goal-directed behaviours as assessed in specific instrumental and Pavlovian conditioning tasks and 2- locomotor activity induced by the DA-stimulating drug amphetamine.

Using instrumental conditioning, we first showed that juvenile HFD consumption did not affect goal-directed behaviours as assessed using outcome devaluation and contingency degradation procedures. In addition, we are currently complementing these study by evaluating the effects of juvenile HFD on the ability of Pavlovian conditioned stimulus paired with food to modulate the performance of instrumental responses for food (Pavlovian-to-instrumental transfer) and consummatory responses to food itself (conditioned stimulus-potentiated feeding). Juvenile HFD consumption had no effect on spontaneous locomotor activity, but it greatly enhanced locomotor response to amphetamine (1 mg/kg), a behavior mediated by the mesolimbic DA pathway. Nevertheless, at the cellular level, DA levels, DA turnover and DA receptors expression in striatum and medial prefrontal cortex were not affected. Further analysis of the data revealed a direct relationship between plasma corticosterone levels and locomotor response to amphetamine, suggesting HFD exposure induces some subtle changes between DA mesolimbic system and the stress hypothalamic-pituitary-adrenal axis, which is currently under scrutiny.

P2.177

Characterization of the brain regions and molecular mechanisms associated with the storage of an episodic-like memory in rodents

Veyrac, A. (Orsay)[1], Kenney, J. (Lyon)[2], Allerborn, M. (Lyon)[2], Gros, A. (Orsay)[1], Michon, F. (Lyon)[2], Raguet, L. (Lyon)[2], Godinot, F. (Lyon)[2], Messaoudi, B. (Lyon)[2], Thevenet, M. (Lyon)[2], Garcia, S. (Lyon)[2], Laroche, S. (Orsay)[1], Ravel, N. (Lyon)[2]
[1]*Centre des Neurosciences Paris Sud (CNPS), CNRS UMR8195, Université Paris Sud 11, Orsay, France,* [2]*Centre de Recherche en Neuroscience de Lyon, Equipe Olfaction : du codage à la mémoire, CNRS UMR5292, INSERM U1028, Université Lyon 1, Lyon, France*

One of the major questions in Neuroscience is how the activity of specific neurons in the brain gives rise to the long lasting traces of experience that underlies memory. Although formation and stabilization of long-lasting associative memories require time-dependant coordinated hippocampal-cortical interactions, the mechanisms underlying formation and storage of episodic memory remain unknown. Studies in birds and rodents have shown that animals can have episodic-like memory. We developed an original episodic memory task for rats based on the ability to recall « What-Where-In Which context » (see Poster by Allerborn et al; “A new paradigm to test episodic-like memory in rats based on odor-place-context recollection”). By combining pharmacological inactivation and *ex vivo* functional brain imaging, we identified brain regions that are recruited by recall of an episodic-like memory.
First, pharmacological inactivation of the hippocampus by Muscimol injection before testing memory retention 1 day after the episode, impaired the animals'ability to remember an integrated combination of what and where information. This result confirms a key contribution of the hippocampus to the recollection of episodic memory components. Second, imaging of activity-dependant genes expression (*c-Fos* and *zif268*) revealed a specific recruitment of ventral and dorsal hippocampus (CA1-3, Dentate Gyrus) and of many regions of the prefrontal cortex (anterior cingulate, orbitofrontal and limbic cortex) that positively correlates with individual performance of rats in recollecting episodes. In contrast, other regions such as piriform, lateral entorhinal, perirhinal and posterior part of cingulate cortex were not specifically activated during recall of the episodes. Interestingly, we observed a dissociation between *c-Fos* and *zif268* induced-expression only in the orbitofrontal cortex, in which *c-Fos* was induced while *zif268* expression was similar for experienced and control rats. These results suggest a contrasted contribution of genes-dependent transcriptional regulation and open new insights on the role of synaptic plasticity during episodic memory formation.
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P2.178

A new paradigm to test episodic-like memory in rats based on odor-place-context recollection

Allerborn, M. (Lyon)[1], Courtiol, E. (Lyon)[1], Gros, A. (Orsay)[2], Kenney, J. (Lyon)[1], Messaoudi, B. (Lyon)[1], Thevenet, M. (Lyon)[1], Garcia, S. (Lyon)[1], Laroche, S. (Orsay)[2], Veyrac, A. (Orsay)[2], Ravel, N. (Lyon)[1]
[1]*CRNL, CNRS UMR5292, INSERM U1028, Team: Olfaction: from Coding to Memory, Lyon, France,* [2]*Center of Neuroscience Paris-Sud, CNRS-UMR8195, University Paris-Sud, Orsay, France*

Episodic memory is our capacity to recollect precisely some of our life episodes. It has been initially defined in terms of the different types of information it contains: **what** kind of event are we remembering, **where** and **in which context or when** did it take place. First considered as the privilege of humans, evidence for the existence of episodic memory has also been found in non-human species, notably in birds and rodents, thereafter referred to as episodic-like memory.
The aim of the present study was to design and validate an adequate behavioral paradigm for investigating the formation and recall of episodic-like memory in rats. Given the importance of olfactory experience in rat's everyday life, odors were central to this new paradigm. We also tried to minimize any training effect by restricting the task to short episodes and examined the evolution of memory across longer periods of time compared to previous studies.
In the first phase (**encoding**), rats experienced distinct episodes each characterized by a unique combination of what (odors), where (places) and rich contextual information (sounds, visual stimuli, floor texture) leading either to positive or negative rewards. In the second phase, either 1 or 30 days later, rats were tested for **the recall** of these episodes. Behavioral analysis allowed us to extract which type of particular information (**what, where, in which context**) the animals were able to recollect either at short or long term.
We confirmed that some rats were indeed able to recall a combination of what, where, in which context information, while others could only remember parts of the episode, similar to what has been observed in humans. Performance at long term test also indicated that some elements of this episodic-like representation could still be recollected one month after the episode.
The different animal profiles provide an interesting model to gain an insight into neuronal correlates underlying episodic-like memory. Therefore, a new group of four rats implanted for multisite local field potential recordings were exposed to the same experimental situation and tested for their ability to recollect. Preliminary results of this pilot study will also be presented.
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Distance coding by entorhinal grid cells

Jacob, P.-Y. (Marseille)[1], Horne, M. (Marseille)[1], Poucet, B. (Marseille)[1], Save, E. (Marseille)[1], Sargolini, F. (Marseille)[1]
[1]*UMR 7291, Laboratoire de Neurosciences Cognitives, Marseille, France*

Grid cells are spatially selective neurons in the medial entorhinal cortex (MEC) whose firing fields form a regular triangular pattern across an environment. This activity has been suggested to form a euclidian map-like representation of the rat's location and orientation based on self-motion information. To examine whether changes in the exploratory pattern would affect grid cell firing properties, we recorded grid cell activity while the animals explored different environments: an open arena (150 cm in diameter) and a circular track (CT, same diameter). The results show that the firing spots of grid cells adapted to CT geometry where they tended to be organized in a linear way. The interfield distance in the CT increased by a factor that reflects the expansion of the shortest functional distance between two diametrically-opposed places in the CT compared to the arena. These results suggest a role of grid cells in estimating the distance actually travelled by the animals between two locations. To test this hypothesis behaviorally, we designed a protocol in which rats with NMDA lesions of the MEC were trained to estimate three distances on a linear track (30, 60, and 90 cm) both in light and darkness. Without reference to external cues correct estimation of the distance involved the use of self-motion

information only. We found that MEC-lesioned rats were impaired in estimating the long distances (60, 90 cm) but not the short distance (30 cm), indicating a role of MEC in the self-motion estimation of distance. In contrast, rats with ibotenic lesions of the dorsal hippocampal, like sham-lesioned rats were not impaired for any distances. Overall, these results show that grid cells play a crucial role in the estimation of a functional distance during self-motion-based navigation.

P2.180

Dynamic scenes as an experimental tool to investigate Coarse-to-Fine categorization of scenes within scene-selective cortical areas

Kauffmann, L. (Grenoble)[1], Musel, B. (Grenoble)[1], Chauvin, A. (Grenoble)[1], Guyader, N. (Grenoble)[2], Pichat, C. (Grenoble)[1], Ramanoël, S. (Grenoble)[1,3], Le Bas, J.-F. (Grenoble)[3], Peyrin, C. (Grenoble)[1]
[1]Laboratoire de Psychologie et Neurocognition, CNRS UMR5105, Grenoble, France, [2]Grenoble-Image-Parole-Signal-Automatique, CNRS UMR 5216, Grenoble, France, [3]Grenoble Institute of Neuroscience Inserm U 836, Grenoble, France

Many studies on visual perception suggest that scene recognition follows a predominant coarse-to-fine processing sequence: Low spatial frequencies (LSF) may rapidly reach the orbitofrontal cortex, allowing an initial coarse parsing of the scene, which could then be “retroinjected” through feedback connections into occipito-temporal areas to guide the slower fine analysis of high spatial frequencies (HSF). However, coarse-to-fine processing of scenes was never investigated in areas selectively involved in scene processing: the parahippocampal place area (PPA) and the restrosplenial cortex (RSC). The present fMRI study aimed to test whether a coarse-to-fine strategy occurs in scene-selective cortical regions. In previous works, coarse-to-fine processing was studied using static LSF and HSF scenes displayed at different duration time. We used dynamic scenes as stimuli in order to simulate and impose a coarse-to-fine (CtF) or a reverse fine-to-coarse (FtC) processing of spatial frequency information. Dynamic scenes were composed of six band pass spatial frequency-filtered versions of the same natural scene, from LSF to HSF (CtF sequence) or from HSF to LSF (FtC sequence). Fourteen participants performed first a categorization task (indoors vs. outdoors) on dynamic scenes in block-designed fMRI recording session. Then, to delineate the scene-selective regions, they were submitted to a localizer scan during which they performed a 'one-back' task on scenes and faces. We first assessed whether dynamic scenes elicited the neural network usually involved in CtF analysis. Consistently with previous studies, CtF sequences activated more strongly the orbitofrontal cortex and occipito-temporal areas, relative to the FtC sequences. Our dynamic scenes seemed therefore well suited to investigate the CtF categorization within scene-selective regions. Data from the localizer showed that the contrast between scenes and faces elicited activation within the PPA and the RSC. Therefore, these regions were defined as regions of interest (ROI) and ROIs' activity was compared between CtF and FtC sequences. Results show that only the PPA was more activated by the CtF than the FtC sequences. The present study suggests the CtF strategy as a plausible modus operandi in the PPA.

P2.181

The visual observation of a complex movement improves its execution by an observer

Khaled, F. (Villeurbanne)[1], Rifai Sarraj, A. (Beirut)[2], Rabahi, T. (Villeurbanne)[1], Fargier, P. (Villeurbanne)[1], Khalil, M. (Tripolis)[3], Massarelli, R. (Villeurbanne)[1]
[1]Universite Claude Bernard, Centre de Recherche et d'Innovation sur le Sport, Villeurbanne, France, [2]Université Libanaise, Campus Rafic Hariri, Hadath cedex, Département de Physiothérapie, Beirut, Lebanon, [3]Centre AZM de Biotechnologie, Tripolis, Lebanon

The visual observation (VO) of a motor action has been shown to activate areas of the motor cortex similar to thosethat are functional during the actual execution of the action. Following the hypothesis that such an effect might result in the improvement of a motor action, the presentexperiments were performed to show whether the VO of a complex motor action (a *squat vertical jump*, SVJ) might result in an improvement of a similar movement in an observer.Three groups of 7 male subjects each were asked to just execute SVJ (references, group G1) or to execute the same action after VO of 1 SVJ performed on a video screen (G2) or of 2 videos (executing the same action asynchronously and on the same screen, G3). The test was repeated twice/week (wk) during 6 wks. The results showed slight, not significant changes in G1. When the height values were normalized to a control SVJ (executed in all groups before VO, except references who did no VO) the performance increased, in G3, from 1.8% to a maximum of 4.4% after 5 weeks (not significantly). In G2a significant improvement reached 12.9% after 4 wks from an initial 8.6%. It was further observed that all groups increased the actual values of the SVJs heights as function of time. Thus G1 went from 30.5cm to 31cm (not significant), G2 from 29.2cm to 31.7cm and G3 from 29.8cm to 33.9cm probably indicating a motor learning effect as also show by the flattening of the curves after 6-7 wks. The results show for the first time: a. the possibility that VO of a SVJ may improve the execution of the same jump of an observer, b. the possibility that an increase in stimuli (2 videos) may further increase the motor performance.

P2.182

A new unsupervised automated method for selective sleep deprivation to study sleep in mice

Arthaud, S. (Lyon)[1], Libourel, P.-A. (Lyon)[1], Gervasoni, D. (Lyon)[1], Fort, P. (Lyon)[1], Luppi, P.-H. (Lyon)[1], Peyron, C. (Lyon)[1]
[1]*Centre de Recherche en Neuroscience de Lyon, CNRS UMR5292, Inserm U1028, Equipe SLEEP, Lyon, France*

Paradoxical sleep (PS) is homeostatically regulated: an increase in PS quantities always follows its selective deprivation (PSD). We have extensively used this paradigm in rats to identify the neuronal networks and mechanisms responsible for PS regulation. However, the need to study sleep in mice has increased in the recent years with the development of genetically modified mice models and new methods such as optogenetics. It is therefore of particular interest to develop a PSD method for mice. Here, we report the results after 6, 24 and 48h of PSD with a new unsupervised automated method, an alternative of the “flower pot”.Mice were implanted with EEG/EMG electrodes for sleep recordings and placed in their deprivation barrels containing bedding. PSD is based on a real-time detection of Wake, Slow Wave Sleep (SWS) and PS via an adaptive algorithm analyzing EEG/EMG signals. When PS is detected, a TTL pulse is applied to a mechanical device leading to a vertical movement of the barrel's floor, waking up the mice. After baseline (BL) recording, PSD was performed during 6h, 24h or 48h always starting at 10 AM. Mice were allowed to have a PS Rebound (PSR) after PSD.

PSD Duration	PS % during PSD	PS % in BL during corresponding PSD duration	PS % during first 90 min PSR	PS % in BL during corresponding 90 min PSR
6 H (n = 15)	1.22 ***	9.53	18.8 ***	10.6
24 H (n = 17)	1.23 ***	6.27	20.5 ***	9.70
48 H (n = 17)	2.31 ***	6.27	24.2 ***	9.70

*[PS % during PSD and PSR vs. BL (*** p<0.001)]*

PS latency after 6, 24 and 48h PSD was of 2.0, 2.2 and 4.2 min. No difference in SWS amount was found during PSD and PSR compare to BL. These data indicate that PSD is highly specific to PS and is not stressful as mice fall asleep instantly after PSD.

Double-staining for Fos and melanin concentrating hormone (MCH) or Hypocretin (Hcrt) were performed after 48h PSD with or without a 2.5h PSR. In agreement with rat data, we found that a majority of MCH neurons were Fos+ after PSR but none after PSD. Conversely, a majority of Hcrt neurons were Fos+ after PSD but none after PSR.
Altogether, these data clearly show that the automated system we developed is a suitable method for identifying the neuronal network of PS.

P2.183

Hippocampal dynamics during encoding of a contextually ambiguous environment

Hok, V. (Marseille)[1], Poucet, B. (Marseille)[1], Save, É. (Marseille)[1]
[1]*LNC-UMR 7291, Aix-Marseille University, Marseille, France*

Place cells are pyramidal neurons in the hippocampus selectively activated while the animal's head is in a particular location in a particular environment. Place cells are therefore characterized by location-specific firing (“place fields”), and their activity is subject to, among other variables, environmental manipulations. In normal conditions, environments are reliably encoded by groups of neurons displaying place fields that can be stable for up to several months. Contrasting with this strong spatial reliability, it has been previously shown that a striking discharge variability exists from one pass to another as the rat goes through a firing field. The aim of this study is to clarify the relationship between this neuronal firing variability, termed “overdispersion”, and the pattern separation process that takes place in the CA3 hippocampal subfield. As we exposed alternately the animals to two environments differentiated only by their color (black *vs.* white), we found that CA3 hippocampal representations were progressively separated in a similar vein to what has been found in CA1 hippocampal place cells (Lever et al., 2002). Once a clear separation of the two hippocampal representations was obtained, we then exposed the animals to an ambiguous environment (grey color) after the animals have been exposed to either the white or the black arena. Preliminary results show that the representation used to encode the grey arena is dependent of the pre-exposure to one of the familiar environment, a mechanism presumably relying on the rate remapping process (Leutgeb et al., 2005). This phenomenon is still present even after a week of daily exposure to the grey arena. For cells that have been recorded over several days, overdispersion analysis might strengthen the idea that spike firing variability is a key component in signal processing, supporting various features such as cognitive inter-individual variability and encoding of the environment's identity.

P2.184

Goal-related firing and the representation of goal value in hippocampal place cells

Duvelle, E. (Marseille)[1,2], Hok, V. (Marseille)[1], Poucet, B. (Marseille)[1], Arleo, A. (Paris)[2], Save, E. (Marseille)[1]
[1]*Laboratory of Cognitive Neuroscience, UMR 7291, Aix-Marseille University, CNRS, Marseille, France,* [2]*CNRS, University Pierre & Marie Curie P6, Unit of Neurobiology of Adaptative Processes, UMR 7102, Paris, France*

Place cells are hippocampal pyramidal neurons that discharge when an animal is in a specific region of its environment. Theories of hippocampal function suggest that these place cells implement multiple spatial and non-spatial components in their representations, thereby providing substantial support to episodic memory formation. Recently, it has been shown that place cells, in addition to their main place field, display a secondary, weaker field at a goal location when rats perform a continuous place navigation task. In this task, the animals have to reach an unmarked goal location in an arena with a

single polarizing cue, and stay immobile for 2 seconds to release a food pellet from an overhead dispenser. The observed goal-related firing is thought to provide a signal indicating that the animals have reached the goal. Whether this out-of-field activity incorporates non-spatial components of the task is not known. In the present study, we manipulated the amount of reward in a place navigation task that involved two competing goal locations. Rats implanted with four tetrodes in the dorsal hippocampus were trained to freely visit the two goal locations. In the standard condition, a correct visit to each goal location was rewarded with 1 pellet (1:1 distribution ratio). In subsequent manipulations, the distribution ratio for correct visits was either 1:0 or 1:3. The results show that all rats were able to learn the task, visiting almost equally the two goal locations in the 1:1 condition. Additionally, the animals flexibly adapted their behavior within the sessions as they progressively increased the number of visits to the most rewarded goal. Finally, goal-related firing was observed at both goal locations in most place cells. Whether changes in goal value modulate the goal-related firing is currently being analyzed. This should provide some insight into deciphering the exact significance of the hippocampal goal-related firing.

P2.185

Impairment of fast synaptic actin dynamics causes autistic-like behaviour in a profilin 2 knock-out mouse model

Pilo Boyl, P. (Bonn)[1], Di Nardo, A. (Boston)[2], Liemersdorf, C. (Bonn)[1], Witke, W. (Bonn)[1]
[1]University of Bonn, Institute of Genetics, Bonn, Germany, [2]Children's Hospital Boston, Boston, United States

Autism spectrum disorders (ASD) are a collection of neurological syndromes of genetic origin but with a strong dependence from exposition to a variety of yet unidentified pre-, peri-, and post-natal environmental modifiers. Incidence of ASD is rapidly growing in the Western world for several reasons, reaching nowadays a prevalence of 4-10:1000 children. Although it has been shown a male to female ratio of about 4:1, there is no absolute genetic evidence of X-linkage of these neuropathologies. Many different genetic mutations have been identified in humans, involving more than 100 genes; nevertheless the most recent view in the field is that any alteration of synaptic function which increases excitability or reduces inhibition can prime an individual to the outset of the syndrome. Removal of the neuronal actin binding protein profilin 2 (pfn2) in a conventional knockout (ko) mouse model alters synaptic physiology by increasing the pre-synaptic excitability of glutamatergic neurons, thus unbalancing the system excitatory/inhibitory ratio in the brain (Pilo Boyl et al., 2007). In the last years specific behavioural paradigms have been developed in order to assess autistic-like behaviour in mice. Taking advantage of this knowledge, here we show that the pfn2 ko mouse displays behavioural endophenotypes that correspond to the diagnostic symptoms of human ASD (repetitive behaviour/resistance to changes, decreased or altered communication ability, deficit in social interactions) and therefore represents a potentially good animal model to study the physiology of the syndrome.
With the use of an additional mouse model expressing a pfn2-EGFP fusion protein from the pfn2 locus we also show that pfn2 is present in several relevant neuronal systems, namely the glutamatergic, the dopaminergic, and the serotoninergic systems, which therefore are all possible candidate circuits, alone or in combination, in the aetiology of the autistic phenotype.

P2.186

Short modulation of REM sleep quantity bidirectionally modulates hippocampal synaptic plasticity and memory

Hamieh, A.M. (Lyon)[1,2], Ravassard, P. (Lyon)[1], Youssef, M. (Lyon)[1], Fraize, N. (Lyon)[1], Le Barillier, L. (Lyon)[1], Libourel, P.A. (Lyon)[1], Arthaud, S. (Lyon)[1], Touret, M. (Lyon)[1], Malleret, G. (Lyon)[1], Salin, P. (Lyon)[1]
[1]Université Lyon 1, Lyon, France, [2]CRNL, CNRS UMR5292, INSERM U1028, Lyon, France

Study objectives: Growing evidence supports that rapid eye movement sleep (REMS) plays a crucial role in memory. Previous studies have shown that long-term REM sleep deprivation (RSD) impairs synaptic plasticity, encoding and memory consolidation in rats. However, long-term RSD induces stress which is known to alter memory. Thus, we performed a mild-non stressful- RSD in order to ascertain a potential role of REM sleep in synaptic plasticity and memory. Then, we examined whether this RSD or REMS rebound (RSR) obtained 4 hours after RSD affects CA1 hippocampal synaptic plasticity, encoding and memory consolidation of contextual fear conditioning (CFC).
Methods: RSD was induced using a dedicated protocol to induce short (4h) and selective deprivation in order to avoid any stress. RSD was carried out before and after CFC encoding. Animals were examined 1 hour later to test the effect on encoding or 24 hours later to test the effect on consolidation.
Results: The results demonstrated that RSD decreases synaptic LTP selectively in dorsal CA1 and RSR rescues these deficits. RSD performed immediately following encoding impaired consolidation of CFC at test. In contrast, animals subjected to RSR before encoding showed an increase in the amount of freezing response during training. Moreover, increase in REMS quantity (RSR) after encoding facilitated consolidation of CFC at test. Our results suggest that an increase in the amount of REMS facilitates LTP, memory consolidation and encoding, while a decrease in REMS impairs LTP and memory consolidation.
Conclusion: These results suggest that REMS quantity may regulate synaptic plasticity, encoding and consolidation of memory in a bidirectional way.
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P2.187

Face the hierarchy: processing the social rank of faces is associated with specific modulations of event-related potentials and alpha power

Breton, A. (Bron)[1], Jerbi, K. (Bron)[2], Hénaff, M.-A. (Bron)[2], Cheylus, A. (Bron)[1], Baudouin, J.-Y. (Bron)[1], Schmitz, C. (Bron)[2], Krolak-Salmon, P. (Bron)[2], Van der Henst, J.-B. (Bron)[1]
[1]Institut des Sciences Cognititves, L2C2 - UMR 5304 CNRS, Bron, France, [2]Centre de Recherche en Neuroscience de Lyon INSERM 1028 CNRS UMR5292 Univ Lyon 1, Bron, France

A growing body of neuroscience studies argues that a major evolutionary force that shapes the brain of primates is the complexity of the social environment. Within this framework, *hierarchy* stands out as a key feature that contributes to such complexity. In order to navigate their social world, individuals need to recognize hierarchical asymmetries. Neuroimaging studies have identified brain structures involved in the processing of hierarchical stimuli but the temporal dynamics of brain activity associated with such processing remains largely unknown. Here, we used electroencephalography to examine the effect of social hierarchy on neural responses elicited by faces. In contrast to previous studies, the key manipulation was that a hierarchical context was constructed, not by varying facial expressions, but by presenting neutral-expression faces in a game setting. Participants were presented with high-rank, middle-rank and low rank faces and had to evaluate the social position of each face with respect to their own position. Both event-related potentials and task-related oscillatory activity were investigated. Three main findings emerge from the study. First, the experimental manipulation had no effect on the early face-related N170 component, which suggests that hierarchy did not modulate the structural encoding of neutral-expression faces. Second, hierarchy significantly modulated the amplitude of the late positive potentials (LPPs) within a 400-700 ms time-window, with high-rank faces elicited the largest LPP amplitude. Third, high-rank faces were associated with the highest reduction of alpha power. Taken together these findings provide novel electrophysiological evidence for enhanced allocation of attentional resource in the presence of high-rank faces. At a broader level, this study brings new insights into the neural processing underlying social categorization.

P2.188

Beta band activity in the subthalamic nucleus of Parkinsonian patients is modulated during proactive and reactive inhibition but not during attentional capture

Benis, D. (La Tronche)[1], David, O. (La Tronche)[1], Lachaux, J.P. (Lyon)[2], Fraix, V. (La Tronche)[1], Seigneuret, E. (Grenoble)[3], Chabardès, S. (La Tronche)[1], Bastin, J. (La Tronche)[1]
[1]Institut des Neurosciences de Grenoble, Université de Grenoble, La Tronche, France, [2]INSERM, U821, Brain Dynamics and Cognition, Lyon, France, [3]University Hospital, Department of Neurosurgery, Grenoble, France

Several studies demonstrated that the subthalamic nucleus (STN) is a critical node of the cortico-basal ganglia loop underlying motor inhibition (Alegre et al., 2012; Aron & Poldrack, 2006; Ray et al., 2011). Precise timing of STN response during stopping is however lacking to unequivocally show its involvement in the motor inhibition process *per se* by comparing electrophysiological and behavioral measurements. Behaviorally, inhibitory performance and the duration of the cognitive processes that enable response suppression can be estimated using the stop signal task (SST), a simple paradigm that dissociates motor and non-motor processes efficiently. Here, we recorded STN activity during three (non-motor) executive control functions in twelve Parkinsonian patients treated deep brain stimulation. We used a modified SST that was designed to disentangle proactive (i.e. preparation to inhibit a move) from reactive (i.e. suppression of a prepotent move) inhibition and attentional capture. Our results demonstrate that, as expected, beta band activity (BBA, 13-35 Hz) decreased in the STN during motor execution whereas this BBA decrease was significantly of smaller amplitude and of shorter duration during reactive response inhibition. Importantly, differential responses linked to response inhibition took place around 150ms before the stop signal reaction time (SSRT), which demonstrate that STN is directly linked to the inhibition process. We also observed a decrease of BBA during proactive inhibition . Taken together these results provide the first electrophysiological evidence that the time course of STN response is compatible with the hypothesis of a direct participation into proactive and reactive inhibition processes.

P2.189

Functional representation of learned communication signals in the songbird brain

Cousillas, H. (Rennes)[1], Alcaix, S. (Rennes)[1], Henry, L. (Rennes)[1], Richard, J.-P. (Rennes)[1], Hausberger, M. (Rennes)[1], George, I. (Rennes)[1]
[1]Univ. de Rennes 1 - Campus de Beaulieu - Bât. 25, CNRS UMR 6552 - Ethologie animale et humaine, Rennes, France

Birdsong, like speech, is a learned vocal behaviour whose development critically depends on social interactions. In highly social songbirds such as starlings, the lack of direct social contacts with adult conspecifics severely impacts the development of song behaviour. This raises the question of what functional representation of their vocalizations such deprived animals develop and what are the neural consequences of such abnormal development. We have recently observed, in the caudo-medial nidopallium (NCM) of adult male starlings, differential neuronal responses to distinct classes of sounds that have different functions and social values, and that are produced separately and differentially according to context. This suggests that this non-primary, associative auditory area that is analogous to the mammalian secondary auditory cortex could be the place for sorting sounds into functional categories in the songbird brain. Here we show that the development of these response properties is experience-dependent: young starlings that we raised without any direct contact with adults not only failed to differentiate starlings' typical song classes in their vocalizations but also failed to develop differential neural responses to these songs. The fact that song classes showing species-typical acoustic morphology were not differentiated in the experimental birds' vocalizations suggests that the observed deficit in neuronal responses to these song classes is likely to be linked to a failure to acquire songs' functions and may provide a model for abnormal development of communicative skills, including speech.

P2.190

Prism adaptation induces a long-lasting bisection bias in healthy subjects

Schintu, S. (Bron)[1], Pisella, L. (Bron)[1], Jacobs, S. (Bron)[1], Salemme, R. (Bron)[1], Reilly, K.T. (Bron)[1], Farnè, A. (Bron)[1]
[1]*Lyon Neuroscience Research Center, INSERM U1028, CNRS UMR5292, ImpAct Team, Bron, France*

Rightward prism adaptation (PA) has been shown to ameliorate visuospatial biases in right brain-damaged patients with neglect, and a single PA session can lead to improvements that last up to several days. Leftward PA in neurologically healthy subjects induces visuospatial neglect-like biases. The duration of these biases, typically assumed to be ephemeral, is actually not known. Here we assessed the time-course of a PA-induced neglect-like bias by repeatedly administering a line bisection task over a period of ~40 minutes after a single session of leftward PA. The typical pattern of pseudo-neglect observed at baseline was counteracted by a PA-induced rightward bias, which was maximal between 5 and 10 minutes. This bias vanished and reappeared several times in the 40 minutes following PA and was present as late as 35 minutes. Besides confirming that PA induces a rightward directional bias in healthy participants, these results demonstrate that the PA-induced bias i) takes time to fully develop, ii) lasts at least 35 minutes, and iii) is not stable, but oscillates. By showing that PA in the undamaged brain is not an ephemeral phenomenon, but rather induces long-lasting cognitive effects, these findings reveal the presence of another, so-far neglected dimension in the domain of prismatic adaptation, namely, time. The prolonged duration of PA-induced spatial biases, previously considered to be a feature of PA unique to brain-damaged subjects, might also apply to the normal brain. These results have implications for models which attempt to explain the mechanisms underlying prismatic adaptation's effects on neglect symptoms in brain damaged patients. We are now investigating the physiology of this visuospatial neglect-like bias using dual site paired-pulse TMS.

P2.191

Dynamic learning of a spatio-temporal sequence as a model of memory interaction

Babayan, B.M. (Paris)[1], Watilliaux, A. (Paris)[1], Girard, B. (Paris)[2], Rondi-Reig, L. (Paris)[1]
[1]*Neurobiologie des Processus Adaptatifs, UMR 7102, Université Pierre et Marie Curie, Paris, France,* [2]*Institut des Systèmes Intelligents et de Robotique, UMR 7222, Université Pierre et Marie Curie, Paris, France*

Spatio-temporal sequence learning requires distinguishing between different locations visited at different time points and associating to each location a particular action. It particularly relies on the ability to maintain a representation of the order in which locations have been experienced over time. Such complex behavior involves interaction between different memory systems to encode spatial as well as temporal information and motor behavior. We have shown that such learning requires the hippocampus, in mice as well as in humans. Our aim is to model spatio-temporal sequence learning in mice at two levels: biological, to identify the structures underlying such learning, and computational, to understand the learning processes.
Our task consists in learning a 2-turn route in a multiple Y-maze, with no environmental cues. Here, the spatial component does not refer to an allocentric representation of space but to the representation of the order in which the intersections have been visited.
We first identify the structures underlying spatio-temporal sequence learning by Fos imaging following the behavioural study. We are investigating the different learning stages: initial exploration, acquisition of the sequence, and over-training with an automation of the sequence. Conjointly, we propose a computational model of the observed learning dynamics which identifies the learning algorithm that

best describes the behavior as well as the learning characteristics such as the learning rate, i.e. how often must an action be reproduced to be remembered, and the trade-off between exploration of the environment and exploitation of what has been learned. The learning model thus gives an insight into the putative computational roles of the networks of structures identified by correlation analyses of Fos imaging.

P2.192

A role for anterior thalamic nuclei in contextual fear memory

Wolff, M. (Talence)[1], Marchand, A. (Talence)[1], Faugère, A. (Talence)[1], Coutureau, E. (Talence)[1]
[1]*Institut de Neurosciences Cognitives, & Intégratives d'Aquitaine (INCIA),CNRS UMR 5287 - Université Bordeaux 1, Talence, France*

Understanding the neural processes that govern the attribution of a predictive value to environmental stimuli is a major issue in behavioural neuroscience. The main strategy to explore this question has been the use of pavlovian fear conditioning paradigms. While a majority of studies have focussed on the specific role of the hippocampus and amygdala in contextual versus cued fear, very few studies examined the potential role of subcortical limbic areas. Among those, the anterior thalamic nuclei (ATN) connect to both the hippocampus and the amygdala but also to the cingular region which is known to support fear-related activity. Here we show that rats sustaining ATN lesions exhibit a specific impairment following context but not tone conditioning. ATN lesions slowed down acquisition without preventing normal freezing behaviour when rats were reexposed to the conditioning context 24h later. However, ATN rats exhibited poor retrieval of contextual but not cued fear when assessed three weeks after conditioning. In addition, extinction was faster in ATN rats and spontaneous recovery of contextual fear was impaired by the lesions. These deficits indicate that contextual fear memories established in the absence of the ATN are not robust when faced with conflicting information. Collectively, these findings support an involvement of the anterior thalamic nuclei in the circuits underlying contextual fear memory.

P2.193

Spatially and Temporally distinct correlates of acoustic and phonemic components in speech perception

Bouton, S. (Genève)[1], Giraud, A.-L. (Geneve)[2]
[1]*Département des Neurosciences, Centre Médical Universitaire, UNIGE, Genève, Switzerland,*
[2]*Centre Médical universitaire, Département de Neurosciences, Geneve, Switzerland*

Phonological processing during speech perception extracts phonetic information from a highly variable acoustic signal. A powerful theory assumed that this phenomenon, categorical speech perception, corresponds to the perception of oppositions between phoneme categories. According to this theory, it should exist a synergy between an acoustic and a phonological determination of the boundary between two phonemic categories. We used a continuum of acoustically varying sounds for testing the identification of distinct phoneme categories (/ba/-/da/). The continuum /ba/-/da/ varied on second formant values which is a frequency variation. Categorical choices are preceded by the perception of sensory evidence in favor of one action to another. Using MEG recording, we found that listening to speech stimuli varying in small and acoustically equal steps caused a fundamental and qualitative change in both the frequency and spatial distribution of cortical activity. The present study showed distinct correlates of acoustic and phonemic components. The encoding of acoustic component peaked at 100 ms following the onset of element at A1, whereas the encoding of phonemic

component showed a negative component at 200 ms followed by a positive one at 300 ms at STS. These findings draw a clear distinction between acoustic and phonemic processing of perceptual decision making, and provide evidence for acoustic-to-phonemic level encoding of speech sounds in human language perception cortex.

P2.194

The hand brake and the foot brake of motor control

Criaud, M. (Lyon)[1,2,3], Longcamp, M. (Marseille)[4], Anton, J.-L. (Marseille)[5], Nazarian, B. (Marseille)[5], Roth, M. (Marseille)[5], Boulinguez, P. (Lyon)[1,3,6]
[1]Université de Lyon , F-69622, Lyon, France, [2]Université Claude Bernard Lyon 1, Villeurbanne, France, [3]CNRS, UMR5229, Centre de Neuroscience Cognitive, Bron, France, [4]Laboratoire de Neurosciences cognitive, CNRS UMR 7291 Aix-Marseille Université, Marseille, France, [5]Centre IRMf de Marseille, Institut de Neurosciences de la Timone, UMR7289 Aix-Marseille Université & CNRS, Marseille, France, [6]Univeriste Claude Bernard Lyon 1, Villeurbanne, France

Most studies of response inhibition have focused on reactive mechanisms triggered by the stimulus one must refrain to react to, either by using experimental tasks probing specifically these processes (Stop Signal Task), or by analyzing only brain activity evoked by the stimulus one must refrain to react to. Yet, recent behavioral investigations have highlighted the possible role of proactive mechanisms in inhibitory control. Proactive inhibitory control (PIC) would operate as a gating mechanism acting on movement initiation processes in anticipation of stimulation.
Two main points still need to be clarified: 1-Identifying the functional anatomy of PIC, and 2-Understanding the interplay between proactive and reactive mechanisms. We adapted a simple Go/Nogo task in two fMRI experiments in order to allow proper recording of the brain activity preceding stimulation. We differently emphasized the need to refrain from reacting in the two experiments.
In the first experiment, subjects were asked to comply with a maximum error rate of 10%. Proactive activity related to PIC was found in the dorsomedial prefrontal cortex, the precuneus, the SMA, the temporal lobes, the hippocampus and the inferior parietal lobule. No activation was found in the acknowledged reactive network (inferior frontal gyrus, inferior parietal lobule, dorsolateral prefrontal cortex). In the second experiment, no maximum error rate was imposed. Results were more variable, either across subjects or across trials, but consistently reported reactive activation in the absence of proactive activity, and vice versa. We conclude that proactive and reactive controls are two complementary inhibitory mechanisms, acting respectively to prevent movement initiation or to catch up ongoing action. They are the parking brake and the brake pedal of motor control.

P2.195

Labial EMG activity during wilful inner speech and during auditory verbal hallucinations in schizophrenia

Loevenbruck, H. (Grenoble)[1], Rapin, L. (Montréal)[2], Duval, C. (Grenoble)[3], Dohen, M. (Grenoble)[3], Polosan, M. (Grenoble)[4], Perrier, P. (Grenoble)[3]
[1]LPNC & GIPSA-lab, CNRS UMR 5105 & 5216, Grenoble, France, [2]Université du Québec à Montréal, Department of Education and Pedagogy, Montréal, Canada, [3]GIPSA-lab, Département Parole et Cognition, Grenoble, France, [4]CHU de Grenoble, Pôle Psychiatrie et Neurologie, Grenoble, France

Background: It has been suggested that inner speech shares properties with motor actions (Feinberg, 1978; Frith, 1992). An influential motor control model (the 'predictive model') claims that when motor commands are sent to the motor system to achieve a particular intended end-state, an

efference copy is issued in parallel (Miall & Wolpert, 1996). This efference copy is used to calculate a prediction of the sensory outcome of the motor plan. It is suggested that if the actual sensory feedback matches the predicted outcome then self-authorship is experienced. But if predicted and actual sensory outcomes do not match, then some external influence must have been at work, resulting in delusion of control symptoms. This model has been extended to inner speech (Frith, 1996). It has been suggested that a failure in the efference copy system could underlie auditory verbal hallucinations (AVH) in schizophrenia, with self-initiated covert verbal actions experienced as originating from an external cause.
Aim: The present study examines two hypotheses: 1. Inner speech is a kind of action; 2. AVHs are disrupted inner speech. If these hypotheses are correct, then inner speech and AVHs should involve motor commands. Electromyographic (EMG) activity should be detectable in speech muscles.
Methods: We examined lip and arm surface EMG activity in 23 healthy participants in tasks involving wilful inner speech (silent reading and silent generation of word definition) compared to relaxation and in 11 schizophrenia patients during AVH and rest periods.
Results: Results in healthy participants show that wilful inner speech corresponds to a significant increase in lip EMG activity compared with relaxation, while the activity in the arm muscle remains constant. Results in schizophrenia patients show a significant increase in lip EMG activity during AVHs relative to rest, without any increase of EMG activity in the arm muscle.
Conclusion: The increase in lip EMG activity during wilful inner speech compared to relaxation in healthy participants is in favour of the “inner speech is a kind of action” hypothesis. The similar EMG increase in schizophrenia patients during AVHs relative to rest highlights the motor nature of AVHs and suggests that they are associated with inner speech.

P2.196

Altered encoding and retrieval of melodies in congenital amusia: dynamic causal modeling of MEG data

Albouy, P. (Lyon)[1], Mattout, J. (Lyon)[1], Sanchez, G. (Lyon)[1], Tillmann, B. (Lyon)[1], Caclin, A. (Lyon)[1]
[1]*CRNL - UCBL - CNRS - UMR5292- INSERM U1028, Lyon, France*

Congenital amusia is a lifelong disorder that prevents individuals from acquiring basic musical skills (Ayotte et al., 2002). The core deficit seems to be related to pitch perception and memory, and to functional and anatomical abnormalities in a fronto-temporal pathway, including auditory regions and inferior frontal cortices (Hyde et al., 2011; Albouy et al., 2013). Our study investigated the cerebral correlates of the short-term memory deficit in congenital amusia using MEG. Amusics and matched controls performed a short-term memory task with melodies. Participants had to indicate whether sequences of six tones that were presented in pairs were the same or different (in melodic contour). We analyzed brain responses during encoding and retrieval of melodic information using distributed source modeling of MEG data to investigate whether altered functioning of the auditory and/or frontal cortices might underlie the disorder. We explored the potential differences between groups in effective connectivity between these sources using Dynamic Causal Modeling (David et al., 2006).
Behavioral results indicated that performance was impaired in amusic participants in comparison to controls. Source modeling of MEG data revealed that evoked responses during melody processing were generated in bilateral fronto-temporal regions (in bilateral STG and IFG pars opercularis), for the encoding and retrieval parts of the experimental trials in both participant groups. Amusics showed altered brain responses in terms of amplitude and latency for encoding and retrieval. Dynamic Causal Modeling revealed in particular that during melody encoding, compared to controls, amusic participants showed an abnormally increased lateral connectivity between the two auditory cortices, decreased intrinsic modulations in both auditory cortices, and decreased backward connectivity between the right inferior frontal gyrus and the right auditory cortex.
Our data present a major contribution to the understanding of the neural networks impaired in congenital amusia, in particular as previous data have suggested that the functional neural anomaly mainly lies outside the auditory cortex (Peretz et al., 2005, 2009; Hyde et al., 2011).

P2.197

Human intracranial recordings reveal a functional link between expectation suppression and mechanisms of conscious perception

Vidal, J.R. (Lyon)[1], Perrone-Bertolotti, M. (Lyon)[1], Jerbi, K. (Lyon)[1], De Palma, L. (Grenoble)[2], Minotti, L. (Grenoble)[2], Kahane, P. (Grenoble)[2], Lachaux, J.-P. (Lyon)[1]
[1]INSERM U1028 - CNRS UMR5292 - CRNL, Lyon, France, [2]CHU Grenoble and Department of Neurology, INSERM U704, F-38043, Grenoble, France

To efficiently guide behaviour our brain uses prior knowledge of the environments statistical regularities to facilitate the interpretation of sensory information. For instance, frequent and highly expected stimuli facilitate information processing in the brain which can boost behaviour. At the neural level this facilitation is often accompanied by activity suppression in sensory cortices (i.e. expectation suppression) which is interpreted as a simultaneous underlying response sharpening of stimulus selective neurons and a response silencing of all non-selective ones. Sensory adaptation by perceptual expectations is usually tested with consciously perceived stimuli, though some suggest that this phenomenon should be automatic and unconscious. Perceptual expectations and consciousness have both respectively been associated to top-down modulation of sensory processing at advanced latencies (>200 ms) but have never been disentangled at the neural level. Here we address this issue specifically for the case of word recognition by recording intracranial broadband gamma-band responses (50-150 Hz) from the occipital, temporal and frontal cortices of epileptic patients. We manipulated sensory evidence and conscious perception with a visual masking procedure. To control perceptual expectations we compared repeated versus non-repeated words. We report that cortical networks in these regions elicit expectation suppression at various latencies reflecting the temporal incidence of top-down modulation of word recognition on these local networks. We also show the temporal spread of cortical signal amplification across distant networks related to conscious word perception. Finally, we report that expectation suppression appears to be linked to sensory signal amplification caused by feedback influences. Our data suggests an underlying link between the mechanisms of perceptual expectations and conscious perception.

P2.198

Action selection in preSMA in conflicting situations: a tDCS study

Spieser, L. (Marseille)[1], van den Wildenberg, W.P.M. (Amsterdam)[2], Hasbroucq, T. (Marseille)[1], Ridderinkhof, R. (Amsterdam)[2], Burle, B. (Marseille)[1]
[1]CNRS - Aix-Marseille University, UMR 7291 Laboratoire de Neurosciences Cognitives, Marseille, France, [2]University of Amsterdam, Department of Psychology, Amsterdam, Netherlands

In everyday life, we have to select and perform actions adapted to the context, sometimes despite the presence of conflicting information in our environment. When a rapid choice between two actions is required, electrophysiological data suggest that an action selection process takes place in the dorso-medial frontal cortex, and more specifically in the pre-Supplementary Motor Area - preSMA (Carbonnell et al., 2004; Vidal et al., 2011). Moreover, preSMA has also been shown to play an essential role in conflict situations, where subjects have to select a response based on the relevant feature of a stimulus and ignore the irrelevant feature that can be incompatible (Taylor et al., 2007; Forstmann et al., 2008; van Gaal et al., 2011). Here, the role of preSMA in action selection and conflict resolution is assessed using transcranial Direct Current Stimulation (tDCS), a non-invasive stimulation technique that uses weak electrical current either to increase or decrease the activity of the cortical targeted area. Anodal tDCS (causing increased activation) or cathodal tDCS (causing inhibition) were applied over preSMA while subjects performed a stimulus-response compatibility task. The analysis of

both subjects' performance and electromyographic (EMG) activity of the muscles involved in the response (allowing to reveal covert incorrect response activations) allows to characterize more precisely which processes are underlying by preSMA in choice situations, either in case of conflicting or non-conflicting information. Preliminary results indicate that performance is improved under anodal tDCS compared to cathodal tDCS, and this effect seems to be specific to fast error trials occurring in incompatible situation, which is coherent with previous fMRI results from the literature (Forstmann et al., 2008).

P2.199

H3 antagonist ciproxifan prevents working memory impairments induced by sleep-restriction by boosting prefrontal cortex activity

Chauveau, F. (Brétigny-sur-Orge)[1], Laudereau, K. (Brétigny-sur-Orge)[1], Pierard, C. (Brétigny-sur-Orge)[1], Thomasson, J. (Brétigny-sur-Orge)[1], Beracochea, D. (Talence)[2]
[1]IRBA Institut de Recherche Biomédiale des Armées, Brétigny-sur-Orge, France, [2]INCIA, Talence, France

Background: Histamine receptor type 3 (H3) antagonists are promising awakening drugs for treatment of sleep disorders such as narcolepsy. Many studies provide strong evidence for awakening and promnesic properties in physiological or pathological states. However, few works have tried to identify their cognitive effects after sleep restriction and the associated neural network.
Methods: Bl/6J male mice were submitted to acute sleep restriction in a shaker apparatus that prevent sleep by transient (20-40 ms) up and down movements. Number of stimulations (2-4), and delay between 2 stimulations (100-200 ms) were randomized. Each sequence of stimulation were also randomly administered (10-30 s interval) for 20 consecutive hours during light (8 h) and dark (12h) phases. Immediately after 20h-sleep restriction, mice were injected with H3 antagonist (ciproxifan 3mg/kg ip) and submitted 30-minutes later to a working memory (WM) assessment using spontaneous spatial alternation task in a T-maze. After behavioral testing, brains were perfused for Fos immunohistochemistry to assess neuronal brain activation.
Results: Sleep restriction paradigm significantly decreased slow wave sleep (from 38.23 ± 3.96 % to 10.15 ± 4.01 % of time, $p< 0.05$) and was followed by significant sleep rebound (55.91 ± 4.73 %, $p< 0.05$). Sleep restriction significantly decreased WM at long (30 s) but not short (5s) inter-trial intervals. WM impairments were mainly observed at the end of the test session, i.e. when proactive interference effect is high. Interestingly, ciproxifan administration prevented WM deficit induced by sleep restriction and also increased specific prefrontal subarea activation, specifically in the prelimbic and cingular corteces. Sleep restriction also induced an increase of immunopositive neurons in dorsal dentate gyrus (dDG), which was not observed in ciproxifan-injected animals.
Conclusion: Ciproxifan 3mg/kg has an enhancing effect on WM after an acute sleep restriction, likely via the decrease of proactive interference sensitivity. The cognitive beneficial effect of ciproxifan is probably due to specific prefrontal cortex area activation and/or to the blockade of dDG over activation associated with WM processes.

P2.200

Study of the anxiolytic effect of Ormenis multicolis extracts

Wakrim, E. (Marrakech)[1], Khalki, H. (Marrakech)[2], Farahan, H. (Marrakech)[2], Chait, A. (Marrakech)[2]
[1]Faculté des Sciences Semlalia de Marrakech, Université Cadi Ayyad, Marrakech, Morocco, [2]Faculté des Sciences Semlalia de Marrakech, Marrakech, Morocco

Chamomile flower heads and its extracts are widely used in pharmaceutical and cosmetic industry. Anti-spasmodic, anti-inflammatory and antimicrobial properties of chamomile are mainly due to the presence of nonvolatile and volatile components.Chamomile tea is used to relieve spasms and inflammatory conditions of the gastrointestinal tract, spasms, ulcers, menstrual disorders and is also used as a gentle sleep aid, particularly for children.
The recent studies tested a Chamomile extract and suggested as an anxiolytic, sedative, myorelaxant and anticonvulsive activity potential.
The aim of our study is to evaluate the effect of this plant on anxiety and locomotor activity. We used for that two tests to evaluate anxiolytic effect (the dark/light box and Marble burying test) for locomotor activity we use the open field test.
Our results show that in the light/dark transition test, the extract of chamomile increased the time spent in the light area and the number of transitions between the two compartments by treated rats compared to controls rats. The extract of chamomile has an anxiolytic effect in the Digging and marble buring test objectified by increasing number of marbles buried. In the open field test the extract significantly affect both the number of squares traveled and the number of adjustments made by the animal.
In conclusion, methanolic extract of chamomile reduces the anxiety in the different tests are investigated.

P2.201

Small-conductance Ca^{+2}-activated K^+ channels distribution in the basal ganglia network and their role in Parkinson's disease

Camon, J. (Marseille)[1], Detheil, T. (Marseille)[1], Manrique, C. (Marseille)[2], Guiraudie-Capraz, G. (Marseille)[3], Turle-Lorenzo, N. (Marseille)[1], Chen, L. (Marseille)[1], Amalric, M. (Marseille)[1], Mourre, C. (Marseille)[1]
[1]CNRS - Aix-Marseille University, LNC, UMR 7291, Pôle 3 C, Marseille, France, [2]CNRS - Aix-Marseille University, FR 3512, Pôle 3 C, Marseille, France, [3]CNRS - Aix-Marseille University, NICN, UMR 7259, Marseille, France

The progressive neurodegeneration of dopaminergic (DA) neurons in the basal ganglia (BG) network causes several motor symptoms observed in Parkinson's disease (PD). This disrupts the firing pattern in specific structures and results in imbalance between inhibitory and excitatory BG pathways. Small-conductance Ca^{+2}-activated K^+ channels (SK channels) are known to adjust the firing pattern of BG structures. Moreover apamin, a selective blocker of SK2 and SK3 channels, promotes burst and irregular firing in DA neurons and increases DA release in the ventral striatum. We therefore questioned whether in parkinsonian rats, the SK channel expression in the BG is modified and how this could impact the behavioural response in a reaction time task.
As a model of early PD, rats received progressive and partial nigrostriatal DA denervation by local infusions of 6-hydroxydopamine (6-OHDA) bilaterally into the striatum. *In situ* hybridization of SK2, SK3 mRNA and iodinated apamin binding were performed at 21 post-lesion days (PLD) in control and lesion groups. A significant decrease of SK3 channel expression level was found in the SNc with no change in SK2 channel expression. Interestingly, an upregulation of SK2, SK3 mRNA and apamin binding was found in the subthalamic nucleus (STN). Functionally, apamin infusion into the STN of parkinsonian rats enhanced the deficits produced by nigrostriatal DA lesions in the reaction time task while apamin infusion into the substantia nigra pars compacta (SNc) had an opposite effect.
In conclusion, SK3 channels variation in the SNc might be due to the loss of DA neurons suggesting that SK2 channels are not localized on DA neurons. However, the behavioural data showed that the blockade of SK channels into the SNc partially reverse DA lesion-induced effects. Moreover in PD conditions, the abnormal firing pattern of STN neurons is highly correlated with motor deficits. Here we showed that an upregulation of SK channels in the STN may underlie the physiological adjustment of subthalamic excitability following partial DA denervation. Regulating SK channels in the BG with appropriate tools may therefore lead to new therapeutic targets in addition to L-DOPA treatment.

P2.202

Selective involvement of the lateral entorhinal cortex and the dorsal hippocampus in acquisition and flexibility of cross modal associations in the rat

Ferry, B. (Lyon)[1], Boisselier, L. (Lyon)[2], Sauvageon, M. (Lyon)[3], Humblot, P. (Lyon)[3], Gervais, R. (Lyon)[1]
[1]CNRS UMR 5292 - INSERM U 1028 - UCBL1, Centre de Recherches en Neurosciences de Lyon, Lyon, France, [2]CNRS UMR 5292 INSERM U 1028, Université C. Bernard Lyon 1, Centre de Recherches en Neurosciences de Lyon, Lyon, France, [3]Université Claude Bernard Lyon 1, Lyon, France

Most rodent models devoted to the study of neural basis of learning and memory are based on tasks in which the conditioned stimulus is processed by one sensory system (a tone, an odor…). The present work is based on a new behavioral paradigm that allows us to investigate the neural basis of cross-modal olfacto-tactile associations. In the rat, these two modalities are of prime behavioral importance for object exploration. The task consisted in finding one baited cup (+) among three, each of the cups presenting a different and specific odor / texture (O/T) combination. During the acquisition of a first task (task 1), the three cups were associated with the following O/T combination: O1T1 for baited cup; O2T1 and O1T2 for non-baited ones. Most rats learn this task within 3 to 6 training sessions (20 trials / session). In a successive task (Task 2) animals had to pair another O/T combination with the reward using a new set of stimuli (O3T3 for cup +, O4T3 and O3T4 for non-baited cups). Results showed that rats manage to learn Task 2 within 1 to 3 training sessions only. In a third task (Task 3), animals had to learn another O/T combination based on previously learned items (ex: O2T3+, O2T4, O1T3). This task is called the “flexibility task” since animals are expected to neglect previously reinforced items and learn the new specific reinforced combination to solve this task. Surprisingly, most rats solve the flexibility task within 1 or 2 training sessions. The main purpose of our work consisted of testing the role of lateral entorhinal cortex (LEC) and the dorsal hippocampus (DH) in each of the 3 tasks. To this aim, animals bilaterally implanted with cannulas in the LEC or the DH were microinfused with lidocaine (4%; 0,4 µL) just before the test session of each task. The results showed that inactivation of either structure did not impair recall of task 1. Interestingly, LEC inactivation selectively impaired acquisition of Task 2 without affecting unimodal learning tasks. In contrast, DH inactivation impaired the acquisition of Task 3 without affecting acquisition of Task 2. Taken together, these results suggest that LEC is involved in the formation of new olfacto-tactile associations, while DH is important for flexibility of this cross-modal associations.

P2.203

Dissociating brain choice signals in group *versus* individual decision making

Girard, R. (Bron)[1], Ligneul, R. (Bron)[2], Wydoodt, P. (Bron)[3], Dreher, J.-C. (Bron)[2]
[1]CNRS, Bron, France, [2]CNRS UMR 5229, Bron, France, [3]CNRS 5229, Bron, France

A fundamental question in social cognition is to know whether an individual behaves differently when making decisions on behalf of a group than for himself. Another question is to know whether individuals behave differently when facing a single individual than when facing a group. To know whether the brain processes differently economic decisions related to social norm violations in these different situations, we used event-related fMRI in a new 2*2 factorial design with a single-shot repeated fairness game (ultimatum game) in which a subject in the scanner responded to monetary offers, either for himself or on behalf of his group (A), facing either a single individual or another group (B). The two groups (A and B) were each composed of 3 individuals. 23 right-handed males were recruited (mean age 22 ± 2.4). Regression analysis based on choice behavior showed that more offers were rejected when playing on behalf of one's own group than for oneself and when facing a single individual than the adversary group. Using an extended computational inequity aversion model, we estimated the probability of accepting the monetary offer as a function of relative inequity (i.e., a

contextual metric of fairness), agent type (for oneself/on behalf of own group A) and opponent type (single individual/adversary group B). We then searched for brain regions showing BOLD response correlating with these parameters using model-based fMRI. Neuroimaging data were preprocessed and analyzed using SPM8. When facing a single individual, as compared to when facing the adversary group, a steeper correlation between BOLD response and inequity aversion was observed in the inferior frontal gyrus, anterior insula and caudate nucleus. Moreover, when individuals made decision on behalf of their group, responses in the posterior insula and amygdala showed a more robust correlation with inequity aversion than when subjects made decisions for themselves. Together, these results indicate that distinct brain regions are engaged in signaling social norm violations according to the type of opponents they are facing (single individual/group) and according to their sense of responsability (on behalf of their group/for themselves).

P2.204

Identifying the proactive inhibitory control network in humans by assessing prestimulus EEG oscillatory activity at the source level

Albares, M. (Lyon)[1,2,3], Lio, G. (Bron)[3], Boulinguez, P. (Lyon)[1,2,3]
[1]Université de Lyon, Lyon, France, [2]Université Claude Bernard Lyon 1, Villeurbanne, France, [3]CNRS, UMR 5229, Centre de Neuroscience Cognitive, Bron, France

Response inhibition plays a major role in cognitive control. It is classically modeled as a set of phasic processes that are intended to detect conflict and suppress the inappropriate motor activations specifically evoked by irrelevant stimuli. However, it was recently suggested that response inhibition might rather rely on proactive processes consisting in locking anticipatorily movement triggering mechanisms.
Proactive inhibition has been studied with event-related fMRI, but the issue is still highly controversial. The main reason is that blood flow analyses provide indirect measures of neural activity and have poor temporal resolution. Critically, the BOLD signal confuses excitation and inhibition. EEG might be complementary, not only because of its temporal resolution, but essentially because it can highlight inhibition-related activity by means of frequency-domain analyses. However, classical scalp EEG measurements are based on the mixing of signals of different types that seriously obscures the meaning of the variables under scrutiny.
Here, we used a slightly adapted go/nogo protocol, and performed advanced EEG blind source separation. We highlighted a set of sources located in the medial parieto-frontal axis, whose activity increases in unpredictable environments (i.e., when subjects must prevent automatic reaction to upcoming events), and desynchronizes just before movement initiation. Such proactive control would activate anticipatorily/deactivate temporarily the self-inhibitory circuitry within the supplementary motor complex (as indexed by alpha power).
These findings reveal the complex and paradoxical mechanisms by which voluntary control of action may be achieved. While controlled actions would rely on the implementation of inhibition of automatic response, automatic behavior would require top-down control to deactivate anticipatorily and temporarily the automatic self-inhibitory circuitry when the environment becomes predictable.

P2.205

Brain processing of emotional scenes with age: effect of arousal context

Mathieu, N. (Grenoble)[1], Harquel, S. (Grenoble)[2], Vercueil, L. (Grenoble)[3,4], Bonnet, S. (Grenoble)[5], Guillemaud, R. (Grenoble)[5], Ida, M. (Grenoble)[6], Campagne, A. (Grenoble)[7]
[1]CNRS - UPMF, LPNC UMR 5105, Grenoble, France, [2]CNRS - LPNC - UMR 5105, Grenoble, France, [3]Centre Hospitalier Universitaire Nord, Service Exploration fonctionnelle du système nerveux, UJF,

Grenoble, France, [4]Grenoble Neurosciences Institut, Inserm U 836, La Tronche, France, [5]CEA, Laboratoire d'électronique et de technologie de l'information, Grenoble, France, [6]CEA, MINATEC IDEAs Laboratory, Grenoble, France, [7]CNRS - UPMF, LPNC - UMR 5105, Grenoble, France

It is well known that negative stimuli induce a higher attention and a greater brain activity than positive stimuli in younger adults. Aging reduces this negativity bias and is sometimes associated with a positivity bias. The reasons of this age-related effect and conditions under which it appears remain in debate. Previous evidences suggest that aging particularly affect the processing and the experience of negative stimuli. But, the occurrence of this effect seems to depend on the arousal level of stimuli. Additionally, the affective context would possibly be an additional moderator in age-related effects. From the cerebral analysis of the Late Positive Potential which is sensitive to attention, our study investigated to what extent the arousal level of negative stimuli modulates these age-related effects and to what extent it might also contribute to the elimination of the negativity bias with age by contextually affecting the processing of positive stimuli. Fourteen young adults and fourteen older adults were instructed to specify their emotional experience (fear/well-being/ no emotion) for 180 natural scene pictures (while their brain activity was recorded via the electroencephalography (EEG) method. Pictures were divided into two blocks. Each of them consisted of a random sequence of 30 negative pictures, 30 positive pictures and 30 neutral pictures. The two blocks differed by the nature of their negative pictures: one block included high-arousal pictures and the second block included low-arousal pictures, so as to constitute two different affective contexts. Results showed that the two arousal levels of negative scenes are processed on an equivalent scale between the two age groups. In the context of high-arousal negative stimuli, a negativity bias was preserved with age. Nevertheless, in the context of low-arousal negative stimuli, young adults maintained a preference for negative scenes by reducing their brain activity to positive whereas older adults favored an emotional bias by reducing their brain activity to neutral scenes. Overall, the present study confirms the interest in considering both the context and the intrinsic properties of stimuli during visual processing of emotional stimuli with age.

P2.206

A potential neural substrate for individual recognition in the songbird auditory telencephalon

Del Negro, C. (Orsay)[1], Menardy, F. (Orsay)[1]
[7]*Université Paris Sud, CNPS UMR 8195, Orsay, France*

Songbirds provide a useful model for studying the neural code underlying the perception of acoustic communication signals. They rely on auditory processing of vocalizations for a number of social behaviors, such as pair-bonding. Vocalizations, songs as well as calls, are multiple cue signals that may convey information about the identity of the bird. An avian brain nucleus that is analogous to the mammalian secondary auditory cortex (the caudo-medial nidopallium or NCM) has recently emerged as part of the neural substrate for sensory representation of conspecific vocalizations. This led us to investigate whether, in the zebra finch, NCM neurons could contribute to the discrimination among vocalizations that convey information about the individual identity of the bird. We focussed on the long distance call. Males and females can indeed identify their mates on the basis of this call alone. We examined whether NCM neurons in paired birds showed a preference for the mate's call over calls of familiar or unfamiliar individuals. To this end, adult male and female zebra finches were paired for two months in the aviary while other males or females remained unpaired (control birds). Three days prior the electrophysiological investigation, each pair was placed in another cage that allowed visual and acoustic contact with another pair. As a first step, single-unit recordings were performed in both paired and control anesthetized birds. Then, by using a telemetric device, we collected multi-unit responses in freely behaving paired birds. Results indicated that, in both anesthetized and awake paired females, neurons exhibited auditory responses of greater magnitude to either the mate's call or the call of the familiar male than to the unfamiliar call, with no difference between the mate's call and the familiar one. In contrast, no such differential responsiveness was observed in both anesthetized and awake paired males and also, in control birds. Results therefore suggest that, in females, social conditions that lead to call-based recognition of individuals affect auditory properties of neurons in NCM. They

also provide evidence that experience-dependent plasticity of NCM properties differs between the sexes.

P2.207

Behavioral assessment of emotional and motivational appraisal during visual processing of emotional scenes according to the type of spatial frequency

Fradcourt, B. (Grenoble)[1], Baciu, M. (Grenoble)[1], Campagne, A. (Grenoble)[1]
[1]*Université Pierre Mendes France - LPNC UMR 5105, Laboratoire de Psychologie et Neurocognition, Grenoble, France*

The present behavioral study investigates the effect of cognitive appraisal, either emotional or motivational, during visual processing of emotional scenes. Previous studies performed on visual processing of emotional stimuli have revealed preference for a specific type of visual spatial frequencies (high spatial frequency, HFS; low spatial frequency, LSF) according to task demands. The majority of the studies focused on the appraisal of the emotional state of others. In this study, we have specifically focused on detecting the preferred type of spatial frequency in stimuli presented under supraliminal perception duration and by focusing on self-perceived emotions and on action tendency. Our results suggest that HSF information was the most relevant to rapidly identify the self-emotional experience (emotional appraisal) while LSF was required to rapidly identify the tendency to action (motivational appraisal). These results were obtained independently from the emotional content of scenes. However, the tendency to action based on the rapid LSF analysis seemed to be temporarily prioritized with respect to emotional experience and more specifically for negative stimuli, whereas the identification of emotional experience based on HSF analysis, showed priority for pleasant stimuli. The present study confirms the interest in considering emotional and motivational characteristics during visual processing of emotional stimuli.

P2.208

Perinatal exposure to high fat diet: sensitization to hippocampal dysfunctions induced by high fat diet in adulthood

Lépinay, A. (Bordeaux)[1,2], Joffre, C. (Bordeaux)[1,2], Larrieu, T. (Bordeaux)[1,2], Aubert, A. (Bordeaux)[1,2], Acar, N. (Dijon)[3,4], Bretillon, L. (Dijon)[3,4], Parnet, P. (Nantes)[5,6], Guesnet, P. (Jouy-en-Josas)[7], Garate, I. (Bordeaux)[1,2], Castanon, N. (Bordeaux)[1,2], Ferreira, G. (Bordeaux)[1,2], Darnaudéry, M. (Bordeaux)[1,2], Layé, S. (Bordeaux)[1,2]
[1]*INRA, Nutrition et neurobiologie intégrée, UMR 1286, Bordeaux, France,* [2]*Univ. Bordeaux, Nutrition et neurobiologie intégrée, UMR 1286, Bordeaux, France,* [3]*INRA, UMR 1324 CSGA, Eye and Nutrition Research Group, Dijon, France,* [4]*Université de Bourgogne, Dijon, France,* [5]*INRA, UMR 1280, Physiologie des Adaptations Nutritionnelles, Nantes, France,* [6]*Université de Nantes, Nantes, France,* [7]*INRA UR 0909, Nutrition et Régulation Lipidique des Fonctions Cérébrales, Jouy-en-Josas, France*

Maternal obesity is well known to affect offspring´s health, increasing the risk of metabolic disorders in adulthood. More recently, maternal obesity has also been reported to contribute to hippocampal inflammation and memory disturbances in offspring. However, the impact of maternal diet independently of maternal obesity remains poorly explored. Our study aims to investigate the impact of high fat diet from conception until adulthood on spatial memory and hippocampal response to inflammation in adult rats. Dams were fed either on a control (5% of lipids) or a high fat diet (20% of lipids) during gestation and lactation. After weaning, male offspring were placed either on control (C-C, HF-C) or high fat (HF-HF) diets. Results showed that dams' body weight was not affected by high fat diet. In the offspring, only HF-HF rats displayed an increased body weight, but both HF-C and HF-HF groups exhibited elevated cholesterol and leptin plasma levels. Spatial memory assessed in the water

maze was impaired in HF-HF rats, while HF-C rats displayed intact performances. In an additional study, we reported that rats fed a high fat diet starting at weaning (C-HF) did not show any memory deficit despite a weight gain similar to the HF-HF group. These results suggest that overweight alone cannot explain memory impairments observed in HF-HF rats. Using TaqMan Low Density Array, we reported a downregulation of genes such as synaptotagmin, synaptophysin or DNA methyltransferase in HF-HF hippocampus. Finally, while plasma cytokines were drastically increased after a peripheral immune challenge in all groups; in the hippocampus, the inflammatory response, assessed by cytokines mRNA expression was blunted in HF-HF rats and exacerbated in HF-C animals. In conclusion, high fat diet restricted to the perinatal period programs long-term metabolic alterations and exacerbates LPS-induced cytokines expression in the hippocampus. In contrast, perinatal high fat diet has no effect on spatial memory suggesting that early high fat exposure is necessary, but not sufficient to induce memory deficit. All together, our results demonstrate that perinatal high fat diet sensitizes to adverse effects of high fat diet on memory in adulthood which is consistent with the concept of early programming.

P2.209

Enhanced vulnerability for cocaine reinforcing effects in female H/Rouen mice selectively bred for depressive-like behavior

Rappeneau, V. (Lyon)[1], Morel, A.-L. (Lyon)[1], Luppi, P.-H. (Lyon)[1], Vaugeois, J.-M. (Rouen)[2], El Yacoubi, M. (Lyon)[1], Bérod, A. (Lyon)[1]
[1]CRNL Lyon, Lyon, France, [2]Université de Rouen, EA 4651-ABTE /ToxEMAC, U.F.R. Médecine & Pharmacie, Rouen, France

Cocaine addiction is strongly associated with depression, however the mechanisms underlying this co-occurrence are not well understood. To shed light on the neurobiological mechanisms by which depression-like states enhance addictive behaviors, we employed a mouse model of depression based on immobility time in the Tail Suspension Test: the Helpless mice (H/Rouen) versus the Non Helpless mice (NH/Rouen). We first demonstrated that female H/Rouen mice were less sensitive to acute cocaine psychomotor stimulant effects compared to female NH/Rouen mice; interestingly, female H/Rouen and NH/Rouen mice displayed similar sensitivity to chronic cocaine psychomotor stimulant effects assessed in the behavioral sensitization paradigm. We next demonstrated that female H/Rouen mice exhibited a stronger cocaine-induced conditioned place preference compared to NH/Rouen mice, indicating a higher sensitivity to cocaine reinforcing properties. Neuroanatomical studies exploring the neural substrates that mediate the increased sensitivity to cocaine reinforcing effects observed in female H/Rouen mice indicated that cocaine-induced conditioned place preference was associated with an increase in Fos in subregions of the prefrontal cortex and limbic regions such as the nucleus accumbens (core and shell), the medial and basolateral amygdala and the dorsal hippocampus (dentate gyrus, CA1 and CA3) in both lines. However, preliminary results highlight a lower activation of subregions of the prefrontal cortex in female H/Rouen mice compared to female NH/Rouen mice, suggesting a reduced executive control over motivation triggered by the cocaine-paired environment. Moreover, they underline a higher activation of the nucleus accumbens core and the amygdala in female H/Rouen mice compared to female NH/Rouen mice, that may reflect their higher propensity to conditioned drug-seeking. Further analyses are currently performed to fully characterize the neurobiological mechanisms underlying the enhanced vulnerability of H/Rouen females for rewarding effects of cocaine.

P2.210

Measuring fast calcium currents using low-affinity calcium indicators

Jaafari, N. (Grenoble)[1], Canepari, M. (Grenoble)[2]
[1]*Inserm U 836, Grenoble Institute of Neuroscience, Canaux calciques fonctions et pathologies, Grenoble, France,* [2]*Inserm U836, Grenoble Institute of Neuroscience, Grenoble, France*

The study of fast Ca^{2+} currents is usually performed using whole-cell patch-clamp recordings. In cells with complex morphology and non-uniform membrane potential, this method presents three problems. First, there is no information on the site of origin of the current. Second, the membrane potential at the site of the patch can be different from that at the site of the current. Third, the measured current can be distorted by electrical filtration and it can be the summation of several currents originating from different sites. In contrast to electrode recordings, local Ca^{2+} influx measured by fluorescence imaging techniques can approach the integral of the Ca^{2+} current at each site of origin and at the temporal limit of the dye-Ca^{2+} binding reaction. It is well known that the kinetics of this reaction depends on the affinity of the indicator and it equilibrates in less than a millisecond with indicators having equilibrium constant (Kd) > 10 µM. Here, using a low-noise high temporal resolution imaging system, we studied in detail how fast fluorescence transients can be correlated to Ca^{2+} currents using different Ca^{2+} indicators. We use simultaneous voltage imaging to correlate local Ca^{2+} currents with changes of membrane potentials. By combining experimental results with computer simulations, we analyse at which extent a Ca^{2+} current can be extracted by fluorescence transients and the possible distortions due to the concomitant reactions with the endogenous Ca^{2+} buffers. Here, we show that the low-affinity indicator Oregon Green-5N can follow the fast rise time of Ca^{2+} currents, associated with an action potential and mainly mediated by L-type Ca^{2+} channels, in hippocampal neurons. Thus, using this approach, we show a first preliminary study of these currents at different dendritic sites in this neuronal type.

P2.211

A mean-field model of the whole basal ganglia addressing mysteries of the circuit´s connectivity

Girard, B. (Paris)[1], Liénard, J. (Paris)[1]
[1]*Centre National de la Recherche Scientifique, Université Pierre & Marie Curie, Paris, France*

We propose a mean field model of the whole basal ganglia (BG) circuitry of primates, designed to address some questions related to its structure and putative function of selection: How can we reconcile the existence of an inhibitory projection from the external part of the globus pallidus (GPe) to the internal (GPi) part with electrophysiological evidences of similar activities in both nuclei? Is the direct/indirect pathway segregation really crucial for the understanding of selection? Is symmetry the rule concerning the projections from the striatum and subthalamic nucleus to the GPe and the GPi? The proposed model includes simulations of the dynamics of the post-synaptic potentials, depending on the type of receptors, and of the signal attenuation along the dendrites. Its numerous parameters are constrained to plausible ranges, based on anatomical (counts of neurons in the BG nuclei, counts of axonal varicosities, approximative position of these on the dendrites) and electrophysiological (average firing rates at rest and rates recorded with the injections of several neurotransmitter blockers in GPe and GPi) data. These parameters are then optimized by a multi-objective evolutionary algorithms, to identify a set of solutions in accordance all with all these constraints.
The existence of a set of solution shows that all the gathered biological data are not contradictory. The analysis of these solutions reveals
1) that the GPe-GPi projection is weak but existent, and that it would be more compatible with the selection function hypothesis, if it was diffused rather than focused;
2) that the explicit segregation of two pathways stemming from the striatum is not necessary to obtain selection, allowing a reconciliation with monkey anatomical data showing a strong overlap between them;
3) that the inputs to the GPe and the GPi are unbalanced, in favor of the former.
Finally, the obtained parameter values for varicosity counts and localization of the receptors along the dendrites are testable predictions.

P2.212

Recurrent neural networks for online language processing: relation between predictive representation and P600 ERP

Hinaut, X. (Bron)[1,2], Petit, M. (Bron)[1,2], Dominey, P.F. (Bron)[1,2]
[1]Inserm U846, Stem Cell and Brain Research Institute, Bron, France, [2]Université de Lyon, Université Lyon 1, Lyon, France

Primates can learn complex sequences that can be represented in the form of categories, and even more complex hierarchical structures such as language. The prefrontal cortex is involved in the representation of the context of such sequential structures, and the striatum permits the association of this context with appropriate behaviours.
In this experiment, the Reservoir Computing paradigm is used: a recurrent neural network with fixed random connections models the prefrontal cortex, and the read-out layer (i.e. output layer) models the striatum.
The model was applied to language syntactic processing, especially thematic role assignment: given a sentence this corresponds to answer the question "Who did what to whom?". The model processes categories (i.e. abstractions) of sentences which are called "grammatical constructions".
The model is able to
(1) process correctly the majority of the grammatical constructions that were not learned, demonstrating generalization capabilities, and
(2) to make online predictions while processing a grammatical construction.
Moreover, we observed that when the model processes less frequent constructions an important shift in output predictions occurs. It is proposed that a significant modification of predictions in a short period of time is responsible for generating Evoked-Related Potentials (ERP) like the P600: this typically occurs when unusual sentences are processed.
The use of the model for complex abstract sequence processing shows that the faculty of representation and learning of sequences in the brain may be based on highly recurrent connections. This experiment suggests that artificial recurrent neural networks can provide insight into the underlying mechanisms of human cortico-striatal function in sentence processing.
Finally, to show the ability of the model to deal with a real-world application, the model was successfully applied in the framework of human-robot interaction for both sentence comprehension and production. The sentence production model was obtained by "reversing" the sentence comprehension model: it processes meanings as input and produces sequence of words in output.

P2.213

Neural coding in the prefrontal cortex in a decision making task: insights from the reservoir computing

Enel, P. (Bron)[1], Procyk, E. (Bron)[1], Dominey, P.F. (Bron)[1]
[1]INSERM U846 / Université Lyon 1, Bron, France

It is now accepted that the prefrontal cortex (PFC) plays a major role in higher cognitive processes that include sequence processing. How is this mechanism implemented in the PFC still remains a open question. One possible explanation lies in the recurrent nature of local cortical connections which is especially true for the PFC.
A decision making task involving sequential behavior was developed at our laboratory to explore single-neurons prefrontal activity in macaque monkeys. Monkeys had to sequentially explore targets to find that which was rewarded by fruit juice, and then repeat the rewarded choice. A significant number of neurons recorded in the PFC showed a clear dynamical shift between the exploration and the repeat phase. Certain neurons specifically encoded the first reward, an essential information in the

task. However, equally important, the previously chosen target was not explicitly represented in neuronal activity.
We approached the PFC sequential processing property through modeling with Reservoir Computing. In this framework, the reservoir is a neural network with fixed recurrent connections that are generated through a controlled stochastic process in order to elicit particular dynamics suitable for sequence processing. In addition, a readout layer is fully connected to the reservoir neurons with connections modified through linear regression learning.
Training the reservoir with different strategies including monkey behavior showed that random behavior cannot be generated by a simple recurrent network. Furthermore, it allowed to characterize monkey´s behavior as structured.
Reservoir activity displayed similarities with unit recordings from monkeys: several neurons in the reservoir showed a different activity between the two phases of the task. While we did not find neurons encoding the first reward, this information was sparsely encoded in the network and could be retrieved through learning. Similarly, while as in the monkey, no single units encoded the previously chosen target, neurons could be successfully trained to respond to it, demonstrating that this information was robustly present in the reservoir, an important results emphasizing that unexpressed information may be sparsely encoded in the activity of neural populations.

P2.214

Which Temporal Difference learning algorithm best reproduces dopamine activity in a multi-choice task?

Khamassi, M. (Paris)[1,2], Bellot, J. (Paris)[1,2], Sigaud, O. (Paris)[1,2], Girard, B. (Paris)[1,2]
[1]*Université Pierre et Marie Curie, Institut des Syst□emes Intelligents et de Robotique, Paris, France,*
[2]*Centre National de la Recherche Scientifique, UMR 7222, Paris, France*

The activity of dopaminergic (DA) neurons has been hypothesized to encode a reward prediction error (RPE) which corresponds to the error signal in Temporal Difference (TD) learning algorithms. This hypothesis has been reinforced by numerous studies showing the relevance of TD learning algorithms to describe the role of basal ganglia in classical conditioning. However, recent recordings of DA neurons during multi-choice tasks raised contradictory interpretations on whether DA's RPE signal is action dependent or not. Thus the precise TD algorithm (i.e. Actor-Critic, Q-learning or SARSA) that best describes DA signals remains unknown. Here we simulated and precisely analyzed these TD algorithms in relation with previous electrophysiological recordings in a multi-choice task performed by rats. We found an apparent dissociation between the signal encoded by dopamine neurons and behavioral adaption of the animals. Moreover, this neural activity seemed to be only partly consistent with an RPE. Further analyses of the evolution of dopamine neurons activity across learning indicated that, complementarily to the RPE, the value function fits well with the activity. Thus in this task, information about both RPE and value may be conveyed by dopamine activity.

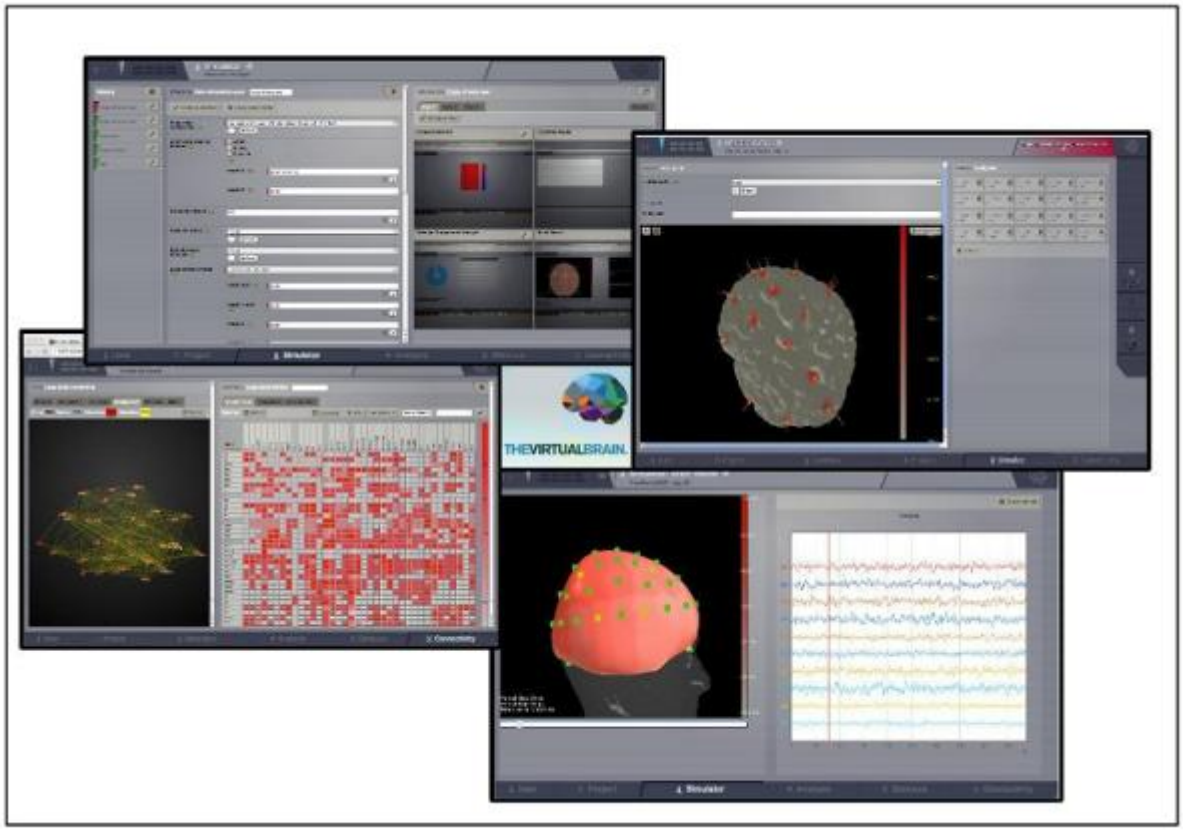
P2.215

The Virtual Brain: a simulator of the human brain

Sanz Leon, P. (Marseille)[1], Woodman, M.M. (Marseille)[1], Jirsa, V. (Marseille)[1], Theoretical Neuroscience Group
[1]*Institut de Neurosciences des Systèmes, Marseille, France*

TheVirtualBrain (TVB) is a simulation tool for large-scale brain modeling. We have reduced the complexity of large-scale brain simulations while still keeping it sufficiently realistic by integrating concepts from theoretical, computational and cognitive neuroscience, together with clinical data. In TVB the large space-time structure of the couplings is given by anatomically realistic connectivity matrices obtained from diffusion tensor imaging (DTI) and their associated fiber lengths, from which we derive the signal transmission delays. Neural mass models define the physiological dynamics of the individual regions serving as network nodes. In this scheme, no particular scale dominates the resulting full brain network model. Instead, the multiple scales interact through mutual

interdependence, which has also the beneficial effect in terms of computational load. Simulations using TVB deliver unique insights into the functioning of the human brain's resting state network. Since the diseased brain shows different resting state behaviors such as in epilepsy and stroke, our connectivity based approach bears the promise to gain novel insights into the origins of network disorders. Each simulation can be completely customized to an individual patient's brain. TVB provides multimodal simulated data under the form of neuroimaging methods commonly used in clinical research such as EEG, MEG and fMRI; and thus allowing a straightforward comparison between simulated data and a patient's real recordings. Our vision is to offer a scientific tool to devise, benchmark and test therapies before pharmaceutical or surgical application. TVB is an open source software written in Python and is not only available as a standalone application but is remotely accessible through a web browser.



[tvb_web_ui]

Figure1. TVB is accesible to everyone through its public web interface.

P2.216

Modeling alternation between gamma and beta oscillations in mammal olfactory bulb

Llorens, V. (Lyon)[1], Voegtlin, T. (Nancy)[2], Fourcaud-Trocmé, N. (Lyon)[1]
[1]*Université Claude Bernard Lyon 1, Centre de Recherche en Neuroscience de Lyon, CNRS UMR5292 - INSERM U1028, Lyon, France,* [2]*INRIA, LORIA, Nancy, France*

Mammalian main olfactory bulb (MOB) dynamics are characterized by oscillatory rhythms both at slow frequency (2-10Hz, linked to animal respiration) and high frequency (beta range, 15-30Hz, and

gamma range, 40-90Hz). The fast rhythm dynamics appear to be determined by intrinsic MOB network properties but their occurrences have been shown to be linked both to odor stimulus and animal state or experience. Thus they are thought to participate and modulate odor coding in behaving animal. To have a better understanding of these fast rhythms, we started a modeling study aiming at explaining how beta and gamma rhythms can be generated in the MOB. Which network parameters determine their intrinsic dynamics (in particular their frequency) and how external inputs, both sensory or cortical, can switch the network from one rhythm to the other.
In a first study, we have shown that a simplified olfactory bulb model including a single glomerulus, (homogeneous external activation of the model) can generate two distinct oscillatory modes. First, in a *low noise* mode, all mitral cells (MOB main excitatory neurons) are well synchronized and saturate the network recurrent inhibition provided by granule cells (MOB interneurons). In this mode, the mitral cell population alternates between an active and a silent state in a beta frequency range. The network frequency is mainly independent on network excitatory input strength thanks to the saturation of recurrent inhibition. Second, in a *high noise* mode, mitral cell discharge is more random and network oscillations, in a gamma frequency range, can be seen only by averaging activity across the whole network. Inhibition is not saturated and a linear analysis of network dynamics shows that, in this mode, the network frequency increases linearly with the excitatory input. This seems contradictory with recordings in anesthetized animals.
In a recent work we increased the network complexity to include multiple glomeruli in order to reproduce the alternation between gamma and beta regime as observed in anesthetized animals. A detailed analysis of this model should help to understand how external or centrifugal input can modulate network parameters, in particular the excitatory-inhibitory balance, to favor either beta or gamma regime.

P2.217
Natural firing patterns reduce sensitivity of synaptic plasticity to spike-timing

Ostojic, S. (Paris)[1], Graupner, M. (New York)[2]
[1]*Ecole Normale Superieure, LNC, Inserm U960,Group for Neural Theory, Paris, France,* [2]*New York University, Center for Neural Science, New York, United States*

Synaptic plasticity is sensitive to both the rate and the timing of pre- and postsynaptic spikes. In experimental protocols used to induce plasticity, the imposed spike trains are regular and the relative timing between every pre- and postsynaptic spike is fixed. This is at odds with natural firing patterns observed in the cortex of intact animals, where cells fire irregularly and the timing between pre- and post-synaptic spikes varies. To investigate synaptic changes elicited by in vivo-like irregularly firing neurons at different rates and realistic correlations between pre- and post-synaptic spikes, we use numerical simulations and mathematical analysis of synaptic plasticity models. We concentrate on a calcium-based model [Graupner and Brunel 2012], and further consider a voltage-based model [Clopath et al 2010] and a spike-timing based model [Pfister and Gerstner 2006]. To allow for comparison, all models are fitted to plasticity results obtained in vitro [Sjostrom et al 2001]. We show that standard stimulation protocols overestimate the influence of spike-timing on synaptic plasticity. Using a simple modification of regular spike-pair protocols, we allow for neurons to fire irregularly. Such irregular spike-pairs reduce the amplitude of potentiation and depression obtained by varying the time difference between pre- and postsynaptic spikes. This protocol allows us to quantify the relative effects of firing rate and timing in natural firing patterns, and to predict changes induced by an arbitrary correlation function between pre- and post-synaptic spikes. We show that spike correlations change synaptic plasticity at low firing rates in all models; whereas their influence becomes negligible at high firing rates for the calcium-based model but remains significant for the other two models. Our findings yield predictions for novel experiments and help bridge the gap between existing results on synaptic plasticity and plasticity occurring under natural conditions.

P2.218

Dendritic tree confers nonlinearity in signaling pathway integration from dendrite to nucleus

Gervasi, N. (Paris)[1], Li, L. (Paris)[2], Girault, J.-A. (Paris)[2]
[1]Institut du Fer à Moulin, INSERM UMRS 839, Paris, France, [2]Institut du Fer à Moulin INSERM S 839, Paris, France

Neurons have particular shapes with extended dendritic trees covered with synapses collecting inputs from their network partners. The role of the geometry of the neuron in the integration of electrical signals has already been investigated. Another important question is related to the control of nuclear functions by cytonuclear signaling pathways, which are important for long-term neuronal adaptations. How are these inputs integrated at the nuclear level and how is this integration dependent on the shape of neurons? Moreover, do all inputs influence equally the information flow to the nucleus or are they weighted by their location?
Our model structure is the striatum that receives convergent motor, sensory and motivational signals from various brain areas. The GABAergic medium-sized spiny neurons (MSNs) of the striatum integrate multiple neuromodulatory inputs by spatio-temporal combination of their signaling pathways. To gain deeper insights in the specificity of the dendritic location in the generation of intracellular signaling activity from dentrite to the nucleus, we developed two parallel approaches: real time imaging of second messengers and kinase activity and computational modeling of cAMP/PKA pathway using different neuronal geometries.
Simulation results from the model with a single dendrite predict that the transfer of cAMP/PKA activity to nucleus depends linearly on the distance of dendritic inputs from the nucleus. Inputs that are initiated closer to the nucleus have a larger influence on nuclear signaling activity than more distal inputs, as expected. However, the linearity is entirely lost when simulation is based on a model with more realistic geometry. We identify specific regions of the dendritic tree that are predicted to have a stronger influence on the nucleus. Using FRET imaging of cAMP and PKA biosensors coupled with photorelease of caged cAMP in MSNs we are testing the existence of a nonlinear integration of cAMP/PKA pathway from dendrite to nucleus.
Nonlinear dendritic integration is a very interesting feature that could increase the computational power of neurons and could imply different capacities of various synaptic inputs to induce long-term neuronal plasticity depending on their location on the dendritic tree.

P2.219

Investigation of the impact of source estimation methods on brain connectivity analysis with magnetoencephalography (MEG)

Hincapie, A.S. (Lyon)[1,2], Kujala, J. (Aalto)[3], Cosmelli, D. (Santiago de Chile)[2], Mery, D. (Santiago de Chile)[2], Jerbi, K. (Lyon)[4]
[1]Lyon Neuroscience Research Center (CRNL), Dycog, Lyon, France, [2]Pontificia Universidad Catolica de Chile, Santiago de Chile, Chile, [3]Aalto University, Brain Research Unit, O.V. Lounasmaa Laboratory, Aalto, Finland, [4]Lyon Neuroscience Research Center (CRNL), Lyon, France

To reduce ambiguity in MEG connectivity analysis is highly desirable to study interaction at the level of the neuronal sources, as their signal can be detected by multiple MEG sensors. The identification of the neuronal sources from the signals detected by the sensors is an inverse problem and is solved using analytical methods. This inverse problem is highly underdetermined and numerous methods have been developed to solve it. However, as two of the most successful methods used in MEG imaging, the Minimum Norm and the Beamforming, have different assumptions, the choice of the method to use can also lead to ambiguity in this analysis. Yet, while comparative studies between the methods have focused on their spatial performance, hardly any study has systematically focused on comparing their connectivity results.
The main objective of this project is to comparatively study the performance of the inverse methods most used in the area for connectivity analysis of neural sources. To achieve this goal, we have built a framework to ensure that the methods are compared under the same anatomical specifications and

functional parameters. In addition we have used simulated sources to analyze the performance of the methods given different characteristics of the signals (signal to noise ratio, location of the sources, level of correlation between the sources, amplitude, number and shape of sources). The use of the common framework as well as the differences in performance of the methods allows us to do the implementation of a reliability index to simultaneously evaluate the results obtained by the methods. The differences found in the performance depending on the characteristics of the signals and the reliability index implemented here can be used as a helpful tool to guide method selection and reduce ambiguity in the interpretation of MEG connectivity results.

P2.220

A multiplicative learning rule for auditory discrimination behavior

Bathellier, B. (Vienna)[1], Tee, S.P. (Vienna)[1], Hrovat, C. (Vienna)[1], Rumpel, S. (Vienna)[1]
[1]*IMP, Vienna, Austria*

Most learning models are based on additive learning rules in which the synaptic update does not explicitly depends on the current weight of the synapse. In contrast, recent observations of temporal fluctuations of dendritic spine volume (a proxy of synaptic efficacy) show that synaptic weight updates in the brain strongly depend on the current weight of the synapse and suggest that biological learning rules may in fact be multiplicative. However, behavioral signatures of this fundamental discrepancy are lacking so far.
We recently observed that individual mice learning an auditory Go/NoGo discrimination task often showed sigmoid-like learning curves, with a long initial “delay phase” where the performance stays at chance level followed by a steep increase of the performance. To understand this phenomenon, we used a reinforcement learning model equivalent to a circuit in which a set of excitatory “sound” neurons projects with plastic synapses onto a decision unit. For this architecture, we could show analytically and confirm by simulations, that when the learning rule is additive no “delay phase” can exist. In contrast, the same model endowed with a multiplicative learning rule can easily reproduce learning curves of individual mice, provided that the initial synaptic weights are low enough.
Hence, we reasoned that the “delay phase” should vanish if we could put the mice in a situation where the connections relevant for learning a certain task have already a high efficacy. Such a situation can be obtained in a reversal experiment where the Go and NoGo sounds are swapped after overtraining the mice on the initial task which should result in strengthening of the relevant connections. In line with the model, we observed no “delay phase” for a reversal experiment.
Altogether, these observations support the idea that the brain uses multiplicative learning rules affecting the dynamics of learning-related behaviors.

P2.221

A biophysical cortical column model to study the effect of GABAA receptor modulation on the VSD signal

Chemla, S. (Marseille)[1], Chavane, F. (Marseille)[1]
[1]*Team InVibe, Institut de Neuroscience de la Timone, CNRS and Aix-Marseille Université, Marseille, France*

Voltage-Sensitive Dye (VSD) imaging produces a mesoscopic, multi-component, signal that results from membrane depolarization dependent fluorescence integrated over a large population of cells. We have previously shown that the contributions of each signal´s components are dynamic and stimulus-dependent (Chemla and Chavane, 2010a). However, these contributions may also be strongly

affected by the network state, such as in anesthetized vs. awake preparations. To study the impact of the network state on the VSD signal, we investigated the effect of the GABAA-mediated IPSCs decay time
constant, tauG, on the global VSD response to a given input stimulus. Importantly, tauG is not only affected by most anesthetics but also by the VSDs themselves (Mennerick et al., 2010). We therefore measured systematically the effect of tauG on our model response to stimulus of increasing strength. Our results suggest that tauG strongly modulates the temporal profile of the VSD signal: an increase in the time constant generates significant decrease in VSD plateau amplitude and modulations in time constants, semi-saturation contrast and inhibition contribution. On the other hand, the type of modulation exerted by VSDs on tauG have a small although significant influence on the population response, and this for most arousal states, changing by around 1-10% most of the VSD signal properties or contributions. We conclude that VSD responses shape and the dynamic contribution of excitation and inhibition to the VSD signal are strongly affected by the arousal state of the animal but only moderately by VSD interaction with GABAA receptor.

P3.001

Reduction of Parvalbumin interneuron number and inhibitory transmission in area CA2 of the hippocampus in the 22q11 mouse model of schizophrenia

Chevaleyre, V. (Paris)[1], Piskorowski, R. (Paris)[2], Stark, K. (New York)[3], Mukai, J. (New York)[3], Siegelbaum, S. (New York)[3], Gogos, J. (New York)[3]
[1]Uiversité Paris Descartes, CNRS UMR 8118, Paris, France, [2]Uiversite Paris Descartes, CNRS UMR 8118, Equipe ATIP/Avenir, Paris, France, [3]Columbia University, New York, United States

An imbalance between excitation and inhibition is observed to occur in several pathologies, and more specifically, a reduction in interneuron number has been observed in area CA2 in the hippocampi of schizophrenic patients in post-mortem studies. Our previous work has demonstrated that inhibition in area CA2 of the hippocampus can act as a gate to control the ability of CA3 to excite CA2 neurons, and thus alter information flow. Motivated by findings in human issue, we asked whether inhibition in area CA2 might also be affected in a mouse model of schizophrenia. We used a line of mice that have a chromosomal micro-deletion (22q11) equivalent to the deletion in humans that leads to DiGeorge syndrome, a developmental disorder that results in the onset of schizophrenia in 30% of diagnosed patients. We have found that the 22q11 deletion (Del) mice have fewer Parvalbumin (PV) expressing interneurons in CA2, with no differences found in areas CA1 and CA3. In addition, inhibitory transmission in CA2 recruited by CA3 inputs was decreased while excitatory transmission from cortical inputs and CA3 was not altered. Interestingly, similar to disease onset in humans, these differences did not manifest until the mice were between 8 and 12 weeks old. Furthermore, CA2 pyramidal cells of Del mice were more hyperpolarized and had a lower membrane resistance. As a consequence, CA2 pyramidal cells in Del mice displayed fewer action potentials in response to CA3 or cortical input stimulation. Finally, the decrease in inhibition in Del mice impaired the activity-dependent increase in the excitatory drive between CA3 and CA2 resulting from a long-term depression of PV inhibitory synapses in CA2. These results indicate that both information transfer and synaptic plasticity are altered in Del mice. Thus, this mouse line is a promising model for examining the cellular mechanisms underlying the changes occurring in the hippocampus during the onset of schizophrenia in humans.

P3.002

Mutations in the MT-related protein KIF2A cause neuronal migration defects in human and mouse

Broix, L. (Paris)[1,2], Poirier, K. (Paris)[1,2], Saillour, Y. (Paris)[1,2], Lebrun, N. (Paris)[1,2], Khalaf-Nazzal, R. (Paris)[3,4,5], Pedespan, J.M. (Bordeaux)[6], Legrez, V. (Meaux)[7], Bahi-Buisson, N. (Paris)[1,2,8], Francis, F. (Paris)[3,4,5], Chelly, J. (Paris)[1,2]
[1]INSERM U 1016, Paris, France, [2]Institut Cochin, Université Paris Descartes, Paris, France, [3]INSERM UMR-S 839, Paris, France, [4]Institut du Fer à Moulin, Paris, France, [5]Université Pierre et Marie Curie, Paris, France, [6]Service de Pédiatrie, CHU Pellegrin, Bordeaux, France, [7]Pédiatrie et urgences

pédiatriques, CH de Meaux, Meaux, France, [8]Unité de Neurologie Pédiatrique, Hôpital Necker Enfants Malades, Université Paris Descartes, Paris, France

Malformations of cortical development (MCD) in human include a spectrum of cortical abnormalities that represent frequent causes of intellectual disability, psychomotor delay and of severe epilepsy. MCDs are known to result from alterations of proliferation and migration of neuronal cells. In recent years, it was shown that the cytoskeleton plays a critical role in cortical development. For instance, mutations in genes encoding α-tubulin subunits (*TUBA1A*, *TUBA8*) and β-tubulins (*TUBB2B*, *TUBB3, TUBB5*) were identified in patients with MCD. These findings stressed the importance of the microtubule (MT) network in these pathologies.
Through a large scale sequencing approach based on exome sequencing of patients with posterior predominant pachygyria and microcephaly, two *de novo* mutations were discovered in the *KIF2A* gene. KIF2A is a member of the kinesin-13 family, which rather than moving along MTs with cargoes, has a special function of depolymerizing MTs.
We analysed the consequences of the newly discovered disease-causing KIF2A mutations in two ways. First, we assessed the deleterious effects of KIF2A mutations *in vivo* by analysing the cellular localization of overexpressed mutant KIF2A proteins in COS-7 and human fibroblast cells. Instead of the expected diffuse punctiform cytoplasmic pattern, KIF2A mutants showed a predominant co-localization with MTs. Similar results were observed in fibroblasts derived from the mutated patient. Altogether, our *ex vivo* studies argue in favour of a dominant-negative effect of the mutation.
Secondly, we investigated the association between *Kif2A* knockdown and MCDs by analysing the consequences of *Kif2a* RNA interference on cortical radial neuronal migration in mice using *in utero* electroporation. Brain sections of E18.5 embryos electroporated at E14.5 with *Kif2a* shRNAs revealed a significant fraction of electroporated cells in the subventricular zone/intermediate zone, suggesting that *Kif2a* down-regulation induced a significant arrest of migrating cells. We conclude that Kif2a plays a critical role in neuronal migration during mouse corticogenesis.
These data reinforce the importance of MT-related proteins in cortical development and strongly suggest that KIF2A-dependent processes contribute to the pathogenesis of MCD.

P3.003
Impaired GABAB-receptor function in adult-born olfactory bulb interneurons

Valley, M. (Paris)[1], Lledo, P.-M. (Paris)[1]
[1]*Institut Pasteur, CNRS URA 2182 - Unité perception et mémoire, Paris, France*

Neuronal regeneration occurs naturally in a few restricted mammalian brain regions, but its functional significance remains unclear. Recently it was shown that acute optogenetic stimulation of adult-born neurons but not neurons born during early post-natal development could accelerate the rate of learning an olfactory discrimination task. To investigate synaptic mechanisms that may underlie this phenotype we compared the GABAergic synaptic outputs of granule cell interneurons born at different ages in the mouse olfactory bulb *in-vitro*. We found that adult-born interneurons born around p60 lacked GABAB neuromodulation of spike-evoked GABA release while interneurons born just after birth at p6 exhibited GABAB sensitivity that declined with age. This functional defect was not due to a lack of GABAB-R1 protein because adult-born neurons exhibited a paradoxical increase in GABAB-R1 immunoflourescent density towards the dendrite interior compared to post-natal born granule cells indicating that a loss of GABAB function may relate to a mis-localization of the receptor. Together, these results suggest that adult neurogenesis produces a population of functionally unique synapses in the olfactory bulb that release GABA more reliably in response to spiking activity.

P3.004

Role of RhoGTPases and microtubule associated proteins in neuronal migration and axon maintenance

Bloch-Gallego, E. (Paris)[1], Couesnon, A. (Paris)[2], Chaimowicz, C. (Paris)[2], Backer, S. (Paris)[2], Chaverot, N. (Paris)[2], Bernard, V. (Paris)[3], Molgo, J. (Gif sur Yvette)[4]
[1]Institut Cochin, INSERM U 1016, CNRS UMR 8104, Univ Paris Descartes, Paris, France, [2]Institut Cochin, Paris, France, [3]INSERM Unité 952, CNRS UMR 7224, Université Pierre et Marie Curie, Paris, France, [4]Institut de Neurobiologie Alfred Fessard, Gif sur Yvette, France

During embryonic development, neuronal growth cones integrate guidance cues that are conveyed to both actin and microtubule (MT) cytoskeletons and ensure axon outgrowth and pathfinding. We had previously shown that Netrin-1, that is secreted, by the floor plate, was chemoattractive for both axons and nuclei of precerebellar neurons (PCN) in the hindbrain. We have demonstrated the respective and distinct roles of Netrin-1 and its receptor DCC in the oriented migration of PCN.
RhoGTPases are factors of interest to mediate the conversion of extracellular cues to modifications of the cytoskeleton. We have investigated their roles in tangential neuronal migrations, as well as in the acquisition of mature and organized structures in neuronal pools of the central nervous system. We demonstrated that Trio-GEF could regulate the clustering of precerebellar and motor neurons through the activation of Rac1. We have recently further investigated the possible role of a matricellular protein - CCN1 -that we discovered to be upregulated downstream RhoGTPases activation in young PCN. Its role in the development of the nervous system had been poor characterized so far.
In parralel, we have investigated the role of a MT associated protein, CLIP3. Unlike other members of the CLIP family (Cytoplasmic Linker Protein), *clip3* is specifically expressed in the nervous system and its invalidation in mouse leads to a lethal phenotype at birth. *Clip3* expression was found to be restricted to late developmental stages and to specific neuronal pools, in particular several nuclei of motoneurons (MNs). *Clip3*$^{KO/KO}$ newborns die from respiratory failure, and show deficits in neuromuscular transmission. Macroscopic observations, confocal imaging and electron microscopy brought the conclusion that *Clipr-59* deficiency affects axon maintenance but not axon guidance toward muscle targets. Thus, CLIPR-59 is involved in the stabilization of specific motor axons at the NMJ during mouse late embryogenesis and its role is crucial for mouse perinatal development.

P3.005

Mapping the gene regulative networks of cortical development

Borello, U. (Bron)[1,2], Madhavan, M. (Cincinnati)[3,4], Campbell, K. (Cincinnati)[3,4], Dehay, C. (Bron)[1,2]
[1]Institut Cellule Souche et Cerveau, INSERM U 846, Bron, France, [2]Université de Lyon, Université Lyon I, Lyon, France, [3]Cincinnati Children's Hospital Medical Center, Division of Developmental Biology, Cincinnati, United States, [4]University of Cincinnati College of Medicine, Cincinnati, United States

The transcriptional network regulating cortical development remains relatively unknown. Molecules secreted by the different forebrain signaling centers establish the expression gradient of specific transcriptional factors, which are necessary for the patterning and differentiation of the distinct structural and functional domains of the cerebral cortex.
To get new insights in the transcriptional regulation of corticogenesis, we have focused on the role of Sp8, a zinc finger transcription factor involved in early patterning and arealization of the cortex (Borello et al, Cereb Cortex 2013).
In order to decipher the molecular mechanisms of Sp8 regulation, we have used a new genetic system to over-express Sp8 specifically in the forebrain at different time points during development. Gain-of-function approach allowed us to examine the role of Sp8 in proliferation, survival and differentiation of cortical progenitors and, importantly, its relationship with Fgf signaling. We have identified a novel regulatory feedback loop at the core of the molecular mechanism controlling patterning and growth of the developing cortex.
Given the fundamental role of Sp8 in patterning the developing cortex and maintaining the balance between neural progenitor cell renewal/differentiation, we are studying the network of genes upstream

and downstream of Sp8. We are implementing a genome wide analysis of the Sp8 downstream genes in the gain-of-function as well as loss-of-function Sp8 mouse mutants. In parallel, we are studying the enhancers regulating Sp8 expression during corticogenesis to identify the gene upstream of Sp8.
A comparative analysis of the molecular mechanisms regulating gene expression between mouse and non-human primates will allow us to understand how evolution shaped the primate brain.
Financial support: Neurodis foundation, LabEX CORTEX.

P3.006

Synaptic activity dependent localization of Tau protein

Frandemiche, M.-L. (La Tronche)[1], De Seranno, S. (La Tronche)[1], Rush, T. (La Tronche)[1], Dollmeyer, M. (La Tronche)[1], Lanté, F. (La Tronche)[1], Buisson, A. (La Tronche)[1]
[1]*Institut des Neurosciences, Equipe 12, La Tronche, France*

Tau is a microtubule associated protein which is mainly known to stabilize microtubules in axons. However, it emerged that tau could participate in dendritic events. Tau plays a pivotal role in toxicity of amyloid beta at the synaptic level and is associated with fyn. This suggests new functions of tau in an Alzheimer's disease (AD) context (Ittner et al., 2010; Hoover et al., 2010). Here, we exposed neurons (DIV 14) to a synaptic activation using bicuculline (50 µM), a GABA-A receptor antagonist, in the presence of a voltage-gated K+-channel blocker 4-aminopyridine (2.5 mM). We performed confocal live imaging on cortical neurons transfected with human-tau-GFP and Life actin-RFP, a peptide that selectively binds to the main cytoskeletal component of post-synaptic excitatory synapses, filamentous actin. In resting conditions, tau was mostly localized in axons and dendritic shafts. However 30% of identified synapses displayed a weak tau-mediated fluorescence. Following synaptic activation, more than 70% of the synapses displayed an intense increase in tau fluorescence. We verified tau localization in spines by performing synaptosomal fractionation to isolate “PSD-enriched” and “non-PSD-enriched” fractions from our cultured neurons. Western blot analysis of endogenous tau proteins revealed a preferential localization of tau in the non-PSD fraction in resting conditions, whereas synaptic activation promoted tau translocation into the post-synaptic density compartment (2-fold increase vs. controls). Exposure to 100 nM of amyloid β oligomers for 15 min induced a mislocalization of tau into the spines both during resting conditions and synaptic activation and observed with confocal imaging and on fractionation samples. These results highlight a new function of tau in the synapse. Deciphering mechanisms underlying the newly discovered function of tau could lead to a better understanding of the synaptotoxic phenomenon observed in AD.

P3.007

T-type calcium channels are involved in cerebellar long term potentiation

Binda, F. (Strasbourg)[1], Uebele, V.N. (Whitehouse Station)[2], Renger, J.J. (Whitehouse Station)[2], Poulain, B. (Strasbourg)[1], Isope, P. (Strasbourg)[1]
[1]*CNRS, INCI, Strasbourg, France,* [2]*Merk & Co., Inc., Whitehouse Station, United States*

T-type channels (Ca_V3 family) are low threshold voltage-gated calcium channels activated by small membrane depolarization and characterized by fast activation and inactivation kinetics. Neuronal T-type channels generate low threshold spikes that can be responsible for burst firing and oscillatory behavior. In the cerebellum, Purkinje neurons (PCs) express T-type calcium channels in the soma and dendrites with the $Ca_V3.1$ isoform being the most represented as shown by the almost complete elimination of T-type currents and calcium transient in PCs from $Ca_V3.1$ knockout mice (Hildebrand et al. 2009). At sub-cellular level, electron microscopy revealed a preferred localization of $Ca_V3.1$ at dendritic spines at parallel fibers (PF) synapses. Interestingly, burst of low intensity PF stimulation

induce fast calcium transients in spine, transients that are mainly mediated by $Ca_V3.1$ openings. At these synapses, raise of intracellular calcium is a key step in the establishment of long term plasticity whose direction strongly depends on the reached calcium concentration. While a strong increase in intracellular calcium leads to long term depression (LTD), potentiation (LTP) is characterized by a lower calcium threshold (Coesmans et al. 2004). T-type calcium channels openings following PF stimulation may contribute to the establishment of this low threshold and therefore be fundamental for LTP at these synapses. Interestingly, mGluR1 activation is required for LTP (Wang et al 2009) and it was shown to potentiate T-Type currents in PCs.
Role of T-type calcium channels in LTP was investigated by patch clamp experiments on acute cerebellar slices. We showed that LTP was induced by high frequency burst of PF stimulation and it was effectively prevented by bath application of the specific T-type calcium channels antagonist TTA-P2 or the specific mGluR1 antagonist JNJ16259685. These results revealed a new and fundamental role of T-type calcium channels in the establishment of long term plasticity.

P3.008

The adhesion-GPCR BAI3 coordinates dendritogenesis and synaptogenesis during neuronal development

Sigoillot, S. (Paris)[1], Usardi, A. (Paris)[1], Lanoue, V. (Paris)[1], Talleur, M. (Paris)[1], Iyer, K. (Paris)[1], Isope, P. (Strasbourg)[2], Vodjdani, G. (Paris)[3], Selimi, F. (Paris)[1]
[1]CIRB - Collège de France, CNRS UMR 7241, INSERM U 1050, Team Mice, Molecules and Synapse Formation, Paris, France, [2]INCI, CNRS UPR 3212, Strasbourg, France, [3]CRICM, UPMC, Inserm UMRS 975, CNRS UMR 7225, Paris, France

Dendritogenesis and spinogenesis constitute key steps in the formation of a functional neuronal network involving a remodeling of the actin cytoskeleton, in particular by RhoGTPases like Rac1. However, how this remodeling is regulated by extracellular signals is not yet fully understood.
The adhesion-GPCRs Brain Angiogenesis Inhibitors (BAI) have several extracellular domains with potential recognition/adhesion properties and are highly expressed in the brain during development. BAI1 is known to modulate the RhoGTPase Rac1 in non-neuronal cells through its interaction with the protein ELMO1. In this context, we have shown that BAI3 is localized in actin-rich structures such as dendrites and spines in hippocampal neurons and that BAI3 knockdown in these neurons or in Purkinje cells leads to an increase in the length and complexity of their dendritic arborization *in vitro*. Our data also revealed that the regulation of dendrite morphogenesis by BAI3 is partially dependent on BAI3's interaction with ELMO1 *in vitro*.
To analyze the role of BAI3 *in vivo*, we first generated transgenic mice expressing a dominant negative form of BAI3 specifically in Purkinje cells that allowed us to show that BAI3 controls dendritogenesis in a cell autonomous manner. Second, using injection of lentivirus in cerebellum of neonatal mice, we confirm that the knockdown of BAI3 in Purkinje cells leads to perturbations of dendrite differentiation. We also observe a decrease in spine and synapse formation in Purkinje cells after knockdown of BAI3. We are currently performing a detailed analysis of the role of BAI3 in synaptogenesis both *in vitro* and *in vivo*. Interestingly, the secreted proteins C1ql, homologues of complement proteins, interact with BAI3 and can modulate synaptogenesis *in vitro*. In this context, our data show that C1ql1 is secreted by one of the two excitatory afferents of the Purkinje cells and can modulate synaptogenesis on these neurons.
Taken together, our results show a new function of BAI proteins as coordinators of the development of neuronal architecture with the establishment of the proper connectivity on a given neuron. Given the link between BAI3 and some symptoms of schizophrenia, our data provide new insights in the study of neurodevelopmental disorders.

P3.009

Microtubule dynamics and post-translational modifications in zebrafish spinal motor axon guidance

Ten Martin, D. (Paris)[1,2], Fassier, C. (Paris)[1,2], Hazan, J. (Paris)[1,2]
[1]*Université Pierre et Marie Curie Paris VI, Paris, France,* [2]*INSERM 952 CNRS 7224, Paris, France*

The formation of a functional nervous system depends on the accuracy of its network wiring during embryonic and postnatal development. All along embryogenesis, neuronal growth cones navigate through precisely defined paths to reach their proper targets. Axon extension and navigation then rely on the reorganisation of the microtubule (MT) and actin networks. Historically considered as a key player in axon extension, MTs have recently been shown to play an instructive role in axon guidance decisions, which sheds new light on the potential involvement of MT-associated proteins (MAPs) in these navigational processes. Our team project aims at deciphering the influence of some MAPs on zebrafish spinal motor neuron (SMN) development. To this aim, we are characterising the differential role and functional redundancy of some MT-associated AAA+ (ATPases Associated with diverse cellular Activities) proteins, including the microtubule-severing spastin and katanin, in SMN axon outgrowth. My PhD project specifically focuses on the involvement of katanin in SMN development. Loss-of-function analyses showed that *katanin* knockdown leads to a dramatic decrease in zebrafish larval mobility, which is tightly associated with severe defects in SMN axon pathfinding. These data thus revealed for the first time *in vivo* that the fine-tuning of katanin levels in SMN is critical for axon guidance processes. Furthermore, since both MT polyglutamylation and acetylation were shown to differentially influence katanin and spastin severing activity, we are now exploring a potential involvement of these MT post-translational modifications in SMN axon extension and navigation. Interestingly, my preliminary analysis of two MT polyglutamylation enzymes, TTLL6 and TTLL11, has shown that their knockdown during development of the zebrafish leads to obvious defects in spinal motor axon architecture, which are strikingly reminiscent of those resulting from *katanin* and *spastin* knockdown, suggesting that MT polyglutamylation similarly controls SMN axon outgrowth.

P3.010

Molecular mechanisms underlying Climbing Fiber/Purkinje Cell synapse formation and specificity

Iyer, K. (Paris)[1], Usardi, A. (Paris)[1], Talleur, M. (Paris)[1], Selimi, F. (Paris)[1], Doyle, J.P. (New York)[2], Heintz, N. (New York)[2]
[1]*CIRB - Collège de France, Paris, France,* [2]*The Rockefeller University, New York, United States*

To investigate the molecular pathways regulating synapse formation and maintenance, we use the olivo-cerebellar network as a model system, focusing on the Climbing Fiber/Purkinje Cell (CF/PC) synapse. We have performed a comparative transcriptomic analysis of different cell populations in this network to identify genes that are specifically enriched in the Inferior Olivary Nucleus (ION), followed by a comparison of these genes with the PC transcriptome to recognize ligand-receptor complexes potentially specific to the CF/PC synapse. The enrichment of candidate genes in the ION is then confirmed by quantitative RT-PCR and their specific localization is assessed by immunofluorescence labelling. We are also performing a functional screening of the candidate genes for their role in the formation and maintenance of the CF/PC synapse in cultured hindbrain explants as this model recapitulates, in vitro, the olivocerebellar network development and maturation. From the transcriptomic analysis, about 1700 genes were found to be specifically enriched in the ION, which included about 500 genes coding for membrane and adhesion proteins. From this group, we selected those genes with a high microarray fold change in the ION, existing or potential roles in synapse formation, and known modes of interaction with binding partners. We are currently focussing on Nectin-3 as a candidate gene. Nectins are immunoglobulin-like cell adhesion molecules that have been shown to play a role in the formation of specific hippocampal synapses (Mizoguchi et al., J Cell Biol 2002) and regulation of adhesive cellular patterns at various types of cell-cell junctions. In our study, additional to in situ hybridization data available online, we confirmed the consistent localization

of Nectin-3 in the ION during development, by immunostaining. We are now proceeding to functionally analyze the possible role of Nectin-3 in regulating the formation and development of ION-originating CF synapses, and in defining CF synaptic innervation territory on Purkinje cells.

P3.011

Unraveling new molecular pathways controlling synapse specificity in the olivo-cerebellar system

Usardi, A. (Paris)[1], Doyle, J.P. (New York)[2], Schmidt, E.F. (New York)[2], Goyenvalle, C. (Paris)[3], Talleur, M. (Paris)[1], Heintz, N. (New York)[2], Selimi, F. (Paris)[1]
[1]Equipe Mice, Molecules and Synapse Formation, CIRB-Collège de France, Paris, France, [2]Laboratory of Molecular Biology, Howard Hughes Medical Institute, The Rockefeller University, New York, United States, [3]Equipe Développement et Vieillissement du Système Nerveux, UMR NPA 7102 CNRS et Univ. P. & M. Curie, Paris, France

Synaptogenesis is a crucial step for the development of a functional brain and is highly specific in terms of partner selection and choice of innervating territory. Defects in this process are linked to neurodevelopmental disorders such as intellectual disabilities and autism. To identify new signaling pathways controlling these processes, we are using the olivo-cerebellar network as a model system since it is characterized by two excitatory afferents - the parallel fibers (PF) which arise from the granule cells (GC) and the climbing fibers (CF) which arise from the inferior olivary neurons (ION) - that have clear differences in terms of their physiology and form synapses on separate dendritic territories on Purkinje cells (PC). We used the bacTRAP strategy to produce a genetically modified mouse that expresses a GFP-tagged ribosomal subunit only in IONs and identify the gene expression profile of this neuronal population. Thus, we have compared this profile to the one previously obtained for GCs and PCs. We have isolated about 500 genes that are highly expressed by IONs, but not in GCs, and that code for membrane proteins that have a potential in regulating connectivity. Amongst these genes we have identified EWI-3, a member of the EWI subfamily of Ig-like proteins. By immunohistochemistry, EWI-3 is detected in specific neuronal populations in the adult mouse brain. Interestingly, the expression of EWI-3 is very dynamic during development: it is highly expressed in the cerebellum at early stages and it disappears with age. EWI proteins are known to be major partners of tetraspanin proteins, which are linked to the actin cytoskeleton and have diverse physiological functions. Using database mining and analysis of mRNA expression levels, we identified tetraspanin 7 (TSPAN7) as a gene highly expressed during cerebellar development. Using affinity-purification, we showed that EWI-3 interacts with TSPAN7 in the developing mouse cerebellum *in vivo*. Mutations in TSPAN7 are associated with X-linked non-syndromic intellectual disability and autism, and TSPAN7 can regulate maturation of excitatory synapses. We are currently investigating the possibility of the EWI/TSPAN complex as a new signaling pathway regulating the proper development of the olivo-cerebellar network.

P3.012

Role of Wnt and Sonic hedgehog signalling on telencephalic differentiation of human pluripotent stem cells

Nicoleau, C. (Evry)[1,2], Bugi, A. (Evry)[3], Bonnefond, C. (Evry)[3], Varela, C. (Evry)[3], Peschanski, M. (Evry)[1,2], Perrier, A. (Evry)[1,2]
[1]INSERM UMR 861/UEVE/AFM, Evry, France, [2]UEVE U 861, I-STEM, AFM, Evry, France, [3]CECS, I-STEM, AFM, Evry, France

Human pluripotent stem cells (hPSCs) can be directed to form neural plate cells (R-NSC) in adherent culture condition through the synergistic inhibition of both activin/nodal and BMP pathway. During mammalian development, Wnt signaling and Hedgehog signaling mediate rostro/caudal of and dorso/ventral patterning of neural plate cells. Here, we explored the role of these pathways in the context of telencephalic induction and telencephalic dorso-ventral patterning of hPSCs-derived R-NSCs.
To investigate the impact of canonical Wnt-signaling we used Wnt agonist (WNT3a) or antagonist (DKK-1; XAV-939) early during neural induction of hPSCs. Expression of neural, telencephalic and non-telencephalic markers were monitored by QRT-PCR at day 10 and 20 of differentiation. RNA level of telencephalic markers (*SIX3*, *FOXG1*, *OTX2)* are significantly higher in R-NSC generated with DKK-1 or XAV-939 while the expression level of more caudal markers (*OTX1*, *LMX1A)* is reduced suggesting that inhibition of WNT signaling promotes telencephalic commitment.
Next, we explored the impact of Shh and Wnt signaling on dorso/ventral patterning of hPSC-neural derivatives *in vitro*. We alternatively treated hPSC-derivatives with cyclopamine (Shh antagonist) or recombinant Shh at increasing concentrations (day 10-28) or with increasing doses of XAV (day 1-20). SHH treatment induced enhanced ventral telencephalic differentiation as shown by increases in mRNA level and cells number positive for ventral telencephalic markers (GSX2, NKX2.1) and reduction in mRNA level of two cortical markers (*EMX1*, *TBR2)*. Analyses conducted at day 20 on telencephalic cultures treated with XAV showed that XAV dose dependently enhances the expression of ventral telencephalic markers and inhibits dorsal markers.
Together these results demonstrate that optimal production of GSX2+ LGE progenitors from hPSC is achieved *via* early and sustained inhibition of Wnt-pathway and moderate and late activation of Shh pathway. LGE progenitors terminally differentiated both *in vitro* and *in vivo (*HD rat model) into neurons expressing DARPP32, Calbindin, CTIP2, FOXP2 and DRD2, markers of authentic medium spiny neurons. Assessment of the therapeutic potential of such hPSC-derived population for HD cell therapy is ongoing.

P3.013

Mature Purkinje cells require the retinoic acid-related orphan receptor alpha (RORα) to maintain climbing fiber monoinnervation and other adult characteristics

Dusart, I. (Paris)[1], Chen, X.R. (Paris)[1], Heck, N. (Paris)[2], Lohof, A. (Paris)[1], Rochefort, C. (Paris)[1], Morel, M.-P. (Paris)[1], Wehrle, R. (Paris)[1], Doulazmi, M. (Paris)[1], Marty, S. (Paris)[3], Cannaya, V. (Paris)[1], Avci, H.X. (Paris)[1], Mariani, J. (Paris)[1], Rondi-Reig, L. (Paris)[1], Vodjdani, G. (Paris)[4], Sherrard, R. (Paris)[1], Sotelo, C. (Alicante)[5]
[1]UPMC University, CNRS UMR 7102, Paris, France, [2]UPMC University, CNRS UMR 7224, INSERM UMRS 952, Paris, France, [3]Institute of Biology of Ecole Normale Supérieure, Paris, France, [4]CRICM - Centre de Recherche de l'Institut du Cerveau et de la Moelle Epinière, Paris, France, [5]Instituto de Neurosciencias, Universidad Miguel Hernandez, Alicante, Spain

Neuronal maturation during development is a multiple step process regulated by transcription factors (TFs). During cerebellar development, the TF RORa (retinoic acid-related orphan receptor alpha) is necessary for Purkinje cell (PC) maturation. The expression of this TF is maintained in adulthood. To identify the role of RORa in mature PCs, we used Cre-lox genetic tools in vivo that delete this TF specifically from PCs between postnatal days 10 - 21, i.e. when PCs have already completed major steps of maturation.
Up to 14 days, no differences were detected between mutant and control PCs: both become monoinnervated by climbing fibers extending all along well-developed dendrites. After 3 weeks on mutant mice, the somata and dendrite of PCs are atrophic with almost complete disappearance of spiny branchlets, and there is some PC death, associated with ataxia. The innervation pattern of surviving mutant PCs is abnormal. Rora -deleted PCs showed several immature characteristics in their climbing fiber (CF) innervation. Notably, multiple functional CF innervation was re-established on these mature PCs, similar to that seen during early development. These CF contacts were found on the soma and proximal dendrites of the rora-deleted PCs, a morphology characteristic of early postnatal

development. This morphological modification of CF contacts could be induced even later, using lentivirus mediated depletion of rora from adult PCs.
The late postnatal expression of RORa cell-autonomously regulates the maintenance of PC dendritic complexity and the CF innervation status of the PC (dendritic versus somatic contacts, and mono- versus multi-innervation). Thus, our data show that the state of differentiation of adult neurons are under the control of TFs and that in the absence of such TFs adult neurons lose their mature characteristics to acquire some of an earlier developmental stage.

P3.014

Molecular control of postnatal forebrain neurogenesis by microRNAs

De Chevigny, A. (Marseille)[1], Follert, P. (Marseille)[1], Coré, N. (Marseille)[1], Hennen, E. (Marseille)[1], Béclin, C. (Marseille)[1], Cremer, H. (Marseille)[1]
[1]*Institut de Biologie du Développement de Marseille, Contrôle Moléculaire de la Neurogenèse, Marseille, France*

During postnatal and adult neurogenesis in the olfactory system of mammals,regionalized neural stem cells in the subventricular zone (SVZ) along the walls of the lateral ventricles permanently generate neuronal precursors. These new neuroblasts perform long distance migration via the rostral migratory stream (RMS) to reach the olfactory bulb (OB) where they differentiate into interneurons with different neurotransmitter phenotypes positions and connectivity. We aimed at identifying molecular mechanisms that control
(1) regionalization and determination of the stem cell pool in the SVZ and
(2) differentiation of a stem cell into a mature neuron.
Increasing evidence reveals that microRNAs function during the entire process of neurogenesis, from neural stem cell self-renewal and fate determination to neuronal maturation, synaptic formation, and plasticity. microRNAs mediate fine-tuning of protein expression through post-transcriptional gene silencing by sequence-dependent binding of target mRNAs. Therefore they are required to finely control expression of key regulatory genes during these processes. To investigate the role of microRNAs in OB neurogenesis, we performed a large scale analysis of microRNA expression in different neurogenic tissues that provided a uniquely detailed picture of microRNA expression in space and time. We identified several microRNAs specifically enriched in different regions in the SVZ that could be involved in regionalization of the neural progenitors. Other microRNAs were expressed in specific tissues of the neurogenic sequence such as the olfactory bulb suggesting a role in neuron maturation and/or integration. From this screen, we showed that miR-7a controlled expression of the transcription factor Pax6 in lateral progenitors to fine-tune dopaminergic differentiation. In addition, we identified the miR-200 family of microRNAs as strong candidates to control the induction of neuronal differentiation in the OB.

P3.015

Early evidences of brain plasticity after hemodialysis session in chronic renal failure patients assessed using BOLD-fMRI

Belaich, R. (Fez)[1,2], Boujraf, S. (Fez)[1,2], Housni, A. (Fez)[1,2], Batta, F. (Fez)[1,2,3], Maaroufi, M. (Fez)[3], Tizniti, S. (Fez)[3], Skalli, T. (Fez)[3], Errasfa, M. (Fez)[4]
[1]*Faculty of Medicine and Pharmacy, University of Fez, Clinical Neuroscience Laboratory, Fez, Morocco,* [2]*Faculty of Medicine and Pharmacy, University of Fez, Department of Biophysics and Clinical MRI Methods, Fez, Morocco,* [3]*University Hospital of Fez, Nephrology Department, Fez,*

Morocco, [4]Faculty of Medicine and Pharmacy, University of Fez, Pharmacology Department, Fez, Morocco

Introduction: The oxidative stress is a known factor contributing to long-term complications of dialysis. Studies have shown the involvement of hemodialysis "HD" membrane in the genesis of oxidative stress (OS). Hence the goal of this study was to assess the early evidences of the impact of HD by the Helixone membrane on brain plasticity in hemodialysis patients suffering of chronic renal failure. Both BOLD-fMRI and serological approaches were used.
Materials and methods: 12 male volunteers following chronic HD for more than 6 months were recruited. Diabetic, smoking and patients with episodes of infection or treatment with iron or erythropoietin injection were excluded. The MDA marker of OS was assessed in the blood using TBARS method before and after HD sessions. Similarly, the BOLD-fMRI was performed before and after HD using motor paradigm immediately before and after HD sessions; the fMRI data was processed using SPM8 package.
Results and conclusion: The biological results showed that HD increases the OS in these patients. [MDA before HD= 3,550 ± 0,580µM vs. MDA after HD =9,899± 8,367µM; p=0,002].
BOLD-fMRI revealed significant activation of the motor cortex, the BOLD signal in the activated site is inversely correlated with level of OS.
The HD seems to raise the inflammatory state of the brain tissue reflecting an increased OS, while it was expected to decrease considering the removal of free radicals responsible of OS by HD procedure. In the meantime, functional brain reaction demonstrated a reorganization of brain functional activity to assume the inflammatory and oxidative stress enhanced by HD process. This reveals the plasticity of the brain induced by the OS occasioned by the HD. Hence, particular care must be paid to HD patients considering the long term impact on general health and brain tissues in particular.
Keywords: BOLD-fMRI, oxidative stress, hemodialysis, Helixone, total MDA

P3.016

Distinct features of CX3CR1 expressing cells within the postnatal subventricular zone/rostral migratory stream

Xavier, A. (Rio de Janeiro)[1], Menezes, J.R.L. (Rio de Janeiro)[1], Nedergaard, M. (Rochester)[2]
[1]Federal University of Rio de Janeiro, Neuroanatomy/Anatomy, Rio de Janeiro, Brazil, [2]University of Rochester Medical Center, Center for Translational Neuromedicine, Rochester, United States

This study characterizes distinct features of CX3CR1 expressing microglia within subventricular zone (SVZ), the major postnatal neurogenic niche, where interneuron precursors are continuously generated and migrate through the rostral migratory stream (RMS) towards the olfactory bulb. Sections obtained from transgenic mice expressing the enhanced green fluorescent protein (eGFP) under the chemokine receptor CX3CR1 promoter at postnatal ages P1, P7, P14 and P60 were immunostained for glial and neuronal markers. Neuroblasts were also labeled with thymidine analog bromodeoxyuridine (BrdU; 100mg/kg, i.p.),1 hour and 5-7 days prior to histological analysis (n=3 animals each age). During neonatal periods, confocal analysis depicts SVZ/RMS microglia displaying typical amoeboid morphology, but at later stages the immature/migratory morphology differs from cortical ramified cells. Despite displaying distinct morphology, these cells do not label for the dendritic cell marker CD11c. Microglia appose to $BLBP^+/GFAP^+$ processes, indicating that they could establish cellular/molecular interactions with SVZ stem cell lineage. Despite the close distribution, neuroblasts do not seem to be phagocytized along the SVZ/RMS, as we do not observe colocalization of CX3CR1-$eGFP^+/TREM2^+$ with $DCX^+/BrdU^+$. In contrast, phagocytic activity occurs at glomerular layer, where neurons are continuously replaced in adult rodents. There, CX3CR1-$eGFP^+/TREM2^+$ cells are co-labeled with neuroblasts markers ($DCX^+/BrdU^+$). Furthermore, ELISA analysis of culture media obtained from P1, P7 and P30 mice reveals that the anti-inflammatory cytokine interleukin (IL) 10 and its activator IL6 are present in the SVZ at all ages, differing from cortical cells, which only secrete these cytokines at P7.The presence of CX3CR1-$eGFP^+$ cells with distinct characteristics under physiological conditions in the SVZ/RMS could reflect intrinsic mechanisms of this neurogenic niche

controlling its macrophage population, that should be addressed for a consistent understanding of adult neurogenesis.

P3.017

Hippocampal synaptogenesis and dendritic spine density enhancement after perinatal asphyxia

Saraceno, G.E. (Buenos Aires)[1], Galeano, P. (Buenos Aires)[1], Caceres, L.G. (Buenos Aires)[2], Romero, J.I. (Buenos Aires)[1], Holubiec, M. (Buenos Aires)[1], Logica, T. (Buenos Aires)[1], Muñiz, J.A. (Buenos Aires)[1], Guelman, L. (Buenos Aires)[1], Castilla, R. (Buenos Aires)[1], Brocco, M. (Buenos Aires)[3], Capani, F. (Buenos Aires)[1]
[1]ININCA, School of Medicine, University of Buenos Aires (UBA), Cytoarchitecture and Neuronal Plasticity Laboratory, Buenos Aires, Argentina, [2]CEFyBO-CONICET, School of Medicine University of Buenos Aires, Buenos Aires, Argentina, [3]Instituto de Investigaciones Biotecnológicas, UNSAM-CONICET, Buenos Aires, Argentina

Even though the underlying causes of neurodevelopmental disorders (NDDs) are known to onset before birth, symptoms may not appear until months or years later. It is thought that dysfunction of neuronal connectivity could play a central role in the pathophysiology of NDDs. Perinatal asphyxia (PA) is an important risk factor for several NDDs of presumed multifactorial etiology since it affects the accurate establishment of neural circuits during a period of apparent normal development. The lack of knowledge of the underlying mechanisms of this dysfunction, prompted us to investigate the morphological changes in the neuronal cytoskeleton induced by PA in a murine model. Consequences of PA on neuronal and glial CA1 hippocampal components, which are involved in the establishment of neuronal circuits, were analyzed at 30 days of age. Results indicated that asphyctic rats showed an increment of NeuN abnormal staining in *stratum radiatum* of the hippocampal CA1 area. No changes were observed in GFAP+ astrocytes morphology, neither in MAP2+ dendrites morphology, nor in the phosphorylation status of medium and heavy neurofilaments (NF H/Mp). Regarding the consequences of PA on dendritic spines, an increase in mushroom-type protrusion was observed using confocal and electron microscopy. Real-time PCR assays revealed an over-expression of β-actin mRNA, while Western blot analysis showed higher β-actin protein levels in synaptosome fractions of asphyctic animals. In addition, these alterations were accompanied by an increase in the expression of M6a, a protein involved in spinogenesis and synaptogenesis. When we analyzed the possible signaling pathways involved in synaptogenesis induced by PA, PI3K/Akt/GSK3 pathway was activated. Surprisingly, asphyctic animals showed better cognitive performance than control rats. Taken together, these results strongly support the hypothesis that abnormal synaptogenesis during brain development induced by PA may contribute to the etiology of the NDDs.

P3.018

Hippocampal short term synaptic plasticity and short term memory are impaired in mice deficient for Melanin concentrating hormone neurons

Le Barillier, L. (Lyon)[1], Léger, L. (Lyon)[1], Fort, P. (Lyon)[1], Luppi, P.H. (Lyon)[1], Malleret, G. (Lyon)[1], Salin, P. (Lyon)[1]
[1]CNRS UMR 5292 INSERM U 1028 CRNL UCBL1, SLEEP Team, Lyon, France

It is well known that paradoxical sleep (PS) facilitates memory. However, the cellular and molecular mechanisms underlying such facilitation are still unknown. Previous works demonstrated that synaptic plasticity is modulated during PS in the hippocampus, a structure which plays a key role in learning

and memory processes. As Melanin Concentrating Hormone (MCH) is supposed to be specifically released during PS and might enhance memory retention, we hypothesize that MCH could be a molecular target that supports the memory function of PS. The aim of our work was to demonstrate whether MCH modulates both hippocampal-dependent synaptic plasticity and memory by combining behavioural and electrophysiological approaches.
We characterized synaptic plasticity at the Schaffer collaterals to CA1 pyramidal cells synapses (SC-CA1) in brain slices from MCH/ataxin3 mice in which MCH neurons disappear progressively at adult age (about three months). We found that the early phase of both LTP and LTD was impaired in MCH/ataxin3 mice as compared to their wild-type controls. As this deficit was associated to a blockade of post-tetanic potentiation (PTP), a common presynaptic form of short term plasticity known to participate to the immediate phase of LTP in SC-CA1 synapses, it showed that short term plasticity was impaired in these mice. This alteration is specific as paired-pulse facilitation which is another form of short-term plasticity, was normal in these mice.
In a Morris water maze task, we observed that MCH/ataxin3 mice were initially slower than controls to learn the spatial location of an hidden platform, although they did not show long-term memory deficits. They also showed a habituation deficit during an open-field task. These behavioral results suggest an alteration of short term memory in MCH/ataxin3 mice.
Altogether, these results suggest that MCH may facilitate short term memory by facilitating short term plasticity in the hippocampus, and point to a possible new role of PS in hippocampal short term plasticity and memory.

P3.019

Identification of multiple subsets of ventral interneurons and differential distribution along the anteroposterior axis of the developing spinal cord

Francius, C. (Paris)[1], Harris, A. (Brussels)[2], Hendricks, T.J. (La Jolla)[3], Stam, F.J. (La Jolla)[3], Barber, M. (Paris)[4], Kurek, D. (Rotterdam)[5], Grosveld, F.G. (Rotterdam)[5], Pierani, A. (Paris)[4], Goulding, M. (La Jolla)[3], Clotman, F. (Brussels)[2]
[1]Institut du Fer à Moulin (IFM), INSERM UMRS 839, Université Pierre & Marie Curie, Paris, France, [2]Université catholique de Louvain, Institute of Neuroscience, Laboratory of Neural Differentiation, Brussels, Belgium, [3]Molecular Neurobiology Laboratory, the Salk Institute for Biologicals Studies, La Jolla, United States, [4]CNRS UMR 7592, Institut Jacques Monod, Université Paris Diderot, Paris, France, [5]Erasmus MC Stem Cell Institute, Department of Cell Biology, Erasmus Medical Center, Rotterdam, Netherlands

The spinal cord contains neuronal circuits that coordinate rhythmic motor activities and are designated Central Pattern Generators (CPGs). The CPG circuits consist in motor neurons and in multiple interneuron cell types, many of which are derived from four distinct cardinal classes of ventral interneurons, called V0, V1, V2 and V3.
While significant progress has been made on elucidating the molecular and genetic mechanisms that control ventral interneuron differentiation, little is known about their distribution along the antero-posterior axis of the spinal cord and their diversification. Here, we report that V0, V1 and V2 interneurons exhibit distinct organizational programs at brachial, thoracic and lumbar levels of the developing spinal cord. We demonstrate that each cardinal class of ventral interneuron can be subdivided into several subsets according to the combinatorial expression of different sets of transcription factors, and that these subsets are differentially distributed along the anteroposterior axis of the spinal cord.
This comprehensive molecular profiling of ventral interneurons provides an important resource for investigating neuronal diversification in the developing spinal cord and for determining the contribution that specific interneuron subsets make to CPG circuits and motor control.

P3.020

Role of Reelin in structural and functional plasticity during postnatal development of the prefrontal cortex

Iafrati, J. (Marseille)[1,2], Orejarena, M.J. (Marseille)[1,2], Lassalle, O. (Marseille)[1,2], Bouamrane, L. (Marseille)[1,2], Chavis, P. (Marseille)[1,2]
[1]*INSERM U 901, INMED, Marseille, France,* [2]*Université de la Méditerranée UMR 901, Aix-Marseille 2, Marseille, France*

Throughout the lifespan, neuronal networks in the central nervous system undergo dynamic rearrangements that take place when the structure and function of the individual synapses change. These synaptic changes occur during the normal brain development and they are triggered by environmental stimuli and experiences. In addition, psychiatric disorders are often associated with profound modifications of the morphology and function of synapses (van Spronsen and Hoogenraad, 2010; Penzes et al., 2011).
Reelin is a large secreted glycoprotein of the brain extracellular matrix. Reelin has been implicated in the maturation of dendritic spines and recent studies from our group show that reelin regulates the homeostasis of glutamate receptor of the NMDA subtype (Campo CG et al., 2009; Groc L et al., 2007; Sinagra M et al., 2005).
Reelin dysfunction has been also proposed to contribute to the etiology of psychiatric disorders. Schizophrenic patients exhibit prefrontal cortex (PFC) dysfunctions and present reduced levels (50%) of reelin in the PFC. However the role of reelin in maturation and plasticity of glutamate connectivity in the PFC has never been investigated. To study the impact of decreased levels of reelin, we took advantage of the Heterozygote Reeler Mice (HRM) which exhibits reduced reelin levels.
We found that long term potentiation, a leading synaptic model for memory storage, is abolished in the PFC of HRM but intact in wild-type mice. The impairment of long-term potentiation in HRM is correlated with changes in glutamatergic receptors content and dendritic spines density in the PFC. Further project development will focus on studying the morphology of dendritic spines in PFC neurons of HRM.
We propose that decreased reelin content causes alterations in structural, functional and behavioral development of prefrontal circuits.

P3.021

RAD51, a protein linked to human congenital mirror movements, is expressed in the murin corticospinal tract during neurodevelopment

Welniarz, Q. (Paris)[1], Shum, M. (Paris)[2], Meneret, A. (Paris)[1], Wherlé, R. (Paris)[2], Morel, M.-P. (Paris)[2], Dusart, I. (Paris)[2], Roze, E. (Paris)[1]
[1]*INSERM U 975, CNRS UMR 7225, Université Pierre et Marie Curie-Paris 6, UMR S 975, Paris, France,* [2]*CNRS UMR 7102, Université Pierre et Marie Curie-Paris 6, Paris, France*

Mirror movements are involuntary movements of one side of the body that accompany and mirror intentional movements on the opposite side and mainly involve the distal upper limbs. *RAD51* mutations in humans have been linked to congenital mirror movements, a rare genetic disorder characterized by mirror movements without additional neurologic defects. Patients with *RAD51* mutations present an aberrant ipsilateral corticospinal tract component, which might be due to an abnormal decussation of the corticospinal tract at the junction between the medulla and the spinal cord. Moreover, heterozygous mutations in *DCC* (deleted in colorectal carcinoma), the gene encoding the receptor for netrin-1, may also result in mirror movements. Impairment of DCC/netrin-1 signaling, which promotes attraction and guidance of developing axons toward the midline, results in altered axonal fiber crossing with abnormal ipsilateral connections.
Surprisingly, *RAD51* is known to be involved in DNA repair by homologous recombination. The involvement of *RAD51* in mirror movements reveals a totally unexpected role of this gene in neurodevelopment: it might be involved in the axonal guidance of the corticospinal tract at the pyramidal decussation.

To understand the involvement of RAD51 in congenital mirror movements, we characterized RAD51 spatio-temporal expression pattern in the mouse brain and spinal cord during development. In the cortex of newborn mice, RAD51 was mainly found in the cell soma in the subplate (SP) and, in lesser amounts, in cortical layer V. Strikingly, RAD51 was detected at P2 (Post natal day 2) within the corticospinal tract from the cortex to the spinal cord: it was expressed in the internal capsule, in ventral position within the hindbrain, and eventually, at the pyramidal decussation. These results suggest that RAD51 deficiency could specifically alter the decussation process. Further ongoing experiments will determine whether RAD51 is expressed in neurons or in glial cells and clarify its subcellular location.

P3.022

In vivo use of transistor for electrical recording and glucose sensing

Doublet, T. (Marseille)[1,2,3], Khodagholy, D. (Gardanne)[2], Welch, M.E. (Ithaca)[4], Ismailova, E. (Gardanne)[2], Quilichini, P. (Marseille)[1], Ghestem, A. (Marseille)[1], Herve, T. (Gardanne)[3], Ober, C. (Ithaca)[4], Malliaras, G. (Gardanne)[2], Bernard, C. (Marseille)[1]
[1]INSERM UMR 1106, INS, Marseille, France, [2]Centre Microélectronique de Provence, Ecole Nationale Supérieure des Mines de Saint-Etienne, BioElectronic, Gardanne, France, [3]Microvitae, Gardanne, France, [4]Department of Materials Science and Engineering, Cornell University, Ithaca, United States

Neuronal firing and field oscillations are major readouts of brain function *in vivo*. Numerous devices have been designed to record single units and field potentials, on the surface of the brain (e.g. grids) or *in situ* (e.g. silicon probes), including in human for diagnosis purposes. These electrodes have enabled major scientific and clinical advances. However, a technological jump is now needed to increase the biocompatibility of these devices (a pre-requisite for future brain-machine interface), improve their signal to noise ratio and allow micro scale multimodal recordings.
We now present such devices.
We developed new microelectrodes which record neuronal activities in vivo and which can be functionalize for glucose sensing. Those electrodes use OECT (organic electrochemical transistor) as recording site. The results show an increase in the signal to noise ratio of the neuronal activity, allowing the recording on the surface of the brain of oscillations, which could only be recorded with depth implantable electrodes.
Furthermore, we also developed glucose sensor using OECT. This electrode show a more sensitive and powerful glucose sensing than the catalytic electrode. The first use of PGMA nanobrushes attached to organic materials here demonstrated allow a local covalent docking of the glucose oxidase without altering the conductivity of the transistor.
These new probes may constitute the basis for future local multimodal recording.

P3.023

Multicolor mosaic analysis in the developing central nervous system with Brainbow strategies

Loulier, K. (Paris)[1], Barry, R. (Paris)[1], Mahou, P. (Palaiseau)[2], Le Franc, Y. (Wilrijk)[3,4], Matho, K.S. (Paris)[1], Supatto, W. (Palaiseau)[2], Fouquet, S. (Paris)[1], Beaurepaire, E. (Palaiseau)[2], Morin, X. (Paris)[5], Livet, J. (Paris)[1]
[1]Centre de Recherche Institut de la Vision, Inserm Umr S 968, Upmc - Paris 6, dépt. de Développement, Paris, France, [2]Ecole Polytechnique, CNRS UMR 7645, INSERM U 696, Laboratory for Optics and Biosciences, Palaiseau, France, [3]University of Antwerp, Laboratory of Theoretical Neurobiology and Neuroengineering, Dept. Biomedical Science, Wilrijk, Belgium, [4]e-Neuroscience Consulting Network, Paris, France, [5]Institut de Biologie de l'Ecole Normale Supérieure, CNRS UMR 8197, INSERM U 1024, Paris, France

Understanding how individual neural progenitors contribute to the architecture of the developing central nervous system (CNS) during development remains challenging. In order to resolve multiple neighboring progenitors and their descendants, we have generated new Brainbow constructs expressing an expanded palette of trichromatic markers (red, yellow and cyan fluorescent proteins) addressed to specific subcellular compartments. We introduce these transgenes by electroporation in the embryonic mouse forebrain and chicken spinal cord. This method yields focal, semi-sparse combinatorial labeling sustained into adulthood. Coupling this approach with genome-integrative transposon vectors allows us to label progenitors over several rounds of cell division, and to track neural stem cells and their descendants from embryonic to adult stages. We have also generated transgenic mice with the new Brainbow constructs, which permit precocious labeling of neural progenitors at stages and locations where electroporation cannot be performed. We use a recently developed multicolor two photon-imaging method to visualize the distribution of clonally related cells over depth of several hundred of microns in CNS samples. Color contrast and marker combinations allow us to resolve neighboring progenitors and their descendants. Finally, we have further developed the Brainbow system to modulate in vivo the function of candidate proteins. To test this approach, we have generated a Brainbow construct that expresses a dominant negative form of the mitotic spindle orienting protein LGN (aka GPSM2) along with the cyan fluorescent protein. Genomic expression of this transgene creates a genetic mosaic in which the status of cells regarding LGN perturbation is color-coded. This approach allows for tracking both perturbed and non-perturbed neighboring cells within the same sample. It will be of interest to characterize cell autonomous and non cell-autonomous actions of candidate effectors in neural progenitors. The multicolor strategies presented will also be applicable in various animal models to investigate the clonal architecture of intact tissues and the signaling pathways modulating their development.

P3.024

How MCPH1 impacts the morphogenesis of the cerebral cortex and its final size: towards a better understanding of the mechanisms responsible for microcephalies

Journiac, N. (Paris)[1,2], Gressens, P. (Paris)[1,2], Nardelli, J. (Paris)[1,2]
[1]*Hopital Robert Debré, Inserm U 676, Paris, France,* [2]*Université Paris Diderot, Paris, France*

Genetic microcephalies (MCPH) are rare diseases, with recessive autosomal transmission, characterized by severe reduction of cerebral volume leading to mental retardation and occasional epilepsy. Mutations in several genes have been linked to microcephaly but the developmental pathways affected are far from being understood. To gain insight into these pathways we focused our studies on the Mcph1 gene, which regulates DNA-damage response and the division of neuroprogenitors by coupling the centrosome cycle with entry into the M phase. However, how Mcph1 influences the final neurogenesis outcome is not understood. First, we established the expression pattern of Mcph1 in mice during telencephalon development. Mcph1 was most expressed in the ventricular zone (VZ) until E13,5 and more discreetly in the preplate. The expression in the VZ decreased markedly from E14.5 and onwards.
Functional studies have been performed in Mcph1 conditional mutant mice. *Mcph1$^{lox/lox}$; Nestin:cre* mice present a decrease in the cortical plate thickness, thus mimicking microcephaly. To delineate more specifically the consequences of Mcph1 inactivation in the telencephalon, we have also included *Mcph1$^{lox/lox}$; Emx1$^{kicre/+}$* mice in our studies. In both contexts, the progenitor populations are not significantly affected at early stages, as assessed by BrdU, Tbr2 and phospho-Histone3 immunostaining. Furthermore, we addressed whether Mcph1 inactivation could specifically impact the production of the Cux2-expressing progenitors that are fated to give rise to layer IV-II neurons in the cortical plate and to pioneer neurons located in the preplate. We found that Mcph1 excision induced a significant decrease in Cux2 progenitors. Consistently, we observed a clear depletion of pioneer neurons in the preplate and in layers IV to II.
We are currently conducting experiments to further evaluate how Mcph1 impacts the production of Cux2 progenitors, and to determine how preplate pionner neurons are eventually involved in this process.

P3.025

Characterization of microcephalin 1 (MCPH1) expression in the human developing dorsal telencephalon

Cipriani, S. (Paris)[1,2], Verney, C. (Paris)[1], Guimiot, F. (Paris)[3], Delezoide, A.-L. (Paris)[4], Adle-Biassette, H. (Paris)[1,2,5], Gressens, P. (Paris)[1,2,6], Nardelli, J. (Paris)[7]
[1]Inserm U 676, Paris, France, [2]Univ Paris Diderot, Sorbonne Paris Cité, UMRS 676, Paris, France, [3]Hôpital Robert Debré, Paris, France, [4]Hopital Robert Debré, Paris, France, [5]Lariboisière Hospital, APHP, Paris, France, [6]Centre for the Developing Brain, King's College, St Thomas' Campus, London, United Kingdom, [7]Centre for the Developing Brain, King's College, St Thomas' Campus, Paris, France

MCPH1 is implicated in DNA damage repair and cell-cycle regulation containing three terminal BRTC domains. *MCPH1* mutation induces an aberrant neurodevelopmental disorder known as autosomal recessive primary microcephaly (MCPH). MCPH is characterized by reduced cortical size and mental retardation. In mice, *Mcph1* mutations have been shown to reduce cerebral cortex size and to induce premature differentiation of neuroprogenitors. However, the role of MCPH1 in human cortical development remains to be investigated. In this study, we explored MCPH1 expression in the human developing dorsal telencephalon with particular regard for its spatial and temporal evolution. Histological studies were performed in human embryos and fetuses aged between gestational week (GW) 6 and 30. MCPH1 was stained by the mean of two antibodies directed against the central region (antibody A) or the terminals BRCT domains (antibody B) of the protein. With both antibodies MCPH1 was mostly localized in nuclear and perinuclear regions. However, antibody A labeled neuroprogenitors of ventricular zone (VZ) and outer sub-VZ (oSVZ), in opposition to antibody B that surprisingly labeled post-mitotic neurons of deep cortical plate (CP) and upper subplate (SP). This suggests selective expression of two MCPH1 forms. MCPH1 staining appeared at very early developmental stage (GW6) with decreasing expression gradient along the medio-lateral axis. At later stages (GW7-GW8), the gradient disappeared. Double labeling studies showed that $MCPH1^+$ cells did not express the proliferation marker Ki67, but post-mitotic markers such as calretinin and reelin. At later stages (GW11-20), CP and SP enlargement were associated with MCPH1 expression increase from the ventro-lateral to dorso-medial cortex. Co-labeling with cortical layer markers showed that MCPH1 was mostly expressed in SP and migrating neurons of layer II-VI, indicating a different origin for these neurons. At GW22, MCPH1 expression was drastically reduced and it was no longer detectable at later stages. Taken together these data indicate that MCPH1 function in cortical development is not restricted to proliferation/differentiation control of neuroprogenitors, but that MCPH1 might also play a role in neuronal maturation and/or migration.

P3.026

The presynaptic machinery acts as a band-pass filter of synaptic activity at the cerebellar granule cell to Purkinje cell connection

Doussau, F. (Strasbourg)[1], Valera, A. (Strasbourg)[1], Poulain, B. (Strasbourg)[1], Isope, P. (Strasbourg)[1]
[1]CNRS UPR 3212, INCI, Strasbourg, France

The mossy fiber to granule cell pathway is one of the major sensorimotor inputs of the cerebellar cortex. In vivo experiments showed that mossy fiber input codes are preserved at the granule cell to Purkinje cell synapse, the output stage of the cerebellar cortex. Since a high variability in the pattern of discharge of the granule cells was observed (from a few Hz to hundred of Hz), understanding signal integration in the whole physiological range in Purkinje cells is fundamental to apprehend information processing in the cerebellum.
Our recent studies have shown that granule cells are able to faithfully convey information during short burst of activity at frequency up to 1 KHz (Valera et al, 2012). At the presynaptic level, this implies that

the active zone is adapted for an ultrafast replenishment of release-ready synaptic vesicles and to the recruitment of reluctant release sites.
Here we investigated the interplay between several forms of short term synaptic plasticity that occurs when the rate of stimulation is rapidly shifted from low to high frequencies. Experimentally, synaptic responses were elicited and recorded in acute cerebellar slices in mice by stimulating parallel fibers and by monitoring evoked EPSCs on Purkinje cells using patch-clamp techniques. We showed that during sustained low-frequency stimulation (LFS) at 2 Hz, EPSCs rapidly faded away. At rest, recovery from LFD takes several minutes after the end of LFS. Since the kinetic and the amplitude of LFD are strongly affected by a calcineurin blocker, this synaptic depression is the consequence of an activity-dependent dephosphorylation of proteins involved in neurotransmitter release. Strikingly, LFD can be temporarily relieved by applying short bursts of stimulation at high frequency (100 Hz) and this recovery can be partially abolished if LFD is elicited by triple pulses at 2 Hz rather than single pulses at 2 Hz. We propose that a frequency-dependent recruitment of new release sites and/or reluctant synaptic vesicles permits the rapid and transient recovery of synaptic transmission at depressed synapses. We suggest then that the granule cell to Purkinje cell synapse could act as a temporal band-pass filter of mossy fiber inputs.

P3.027

White matter development in children with idiopathic localization-related epilepsy

Ciumas, C. (Lyon)[1], Saignavongs, M. (Lyon)[1], Ilski, F. (Lyon)[1], Heckemann, R.A. (Lyon)[2,3], Herbillon, V. (Lyon)[1], Laurent, A. (Lyon)[1], de Belelscize, J. (Lyon)[1], Panagiotakaki, E. (Lyon)[1], Hannoun, S. (Lyon)[3], Sappey Marinier, D. (Lyon)[3], Ostrowsky-Coste, K. (Lyon)[1], Ryvlin, P. (Lyon)[1]
[1]INSERM U 1028, CNRS UMR 5292, Centre de Recherche en Neuroscience de Lyon, Translational and Integrative Group in Epilepsy Research (TIGER), Lyon, France, [2]The Neurodis Foundation, Lyon, France, [3]CERMEP – Imagerie du Vivant, Lyon, France

Background: Epilepsy is a common neurological condition, frequently diagnosed in children and adolescents. Among the various existing forms, localization-related epilepsies of unknown or genetic cause (cryptogenic and idiopathic epilepsies) are the most prevalent. No lesions are usually detected on conventional magnetic resonance imaging (MRI) examination. However, abnormalities at the cellular level affecting the neuronal microstructure may be associated with these syndromes. Only few studies focused on rather mixed syndromes reported changes in white matter architecture in children with recent onset epilepsies. Conversely, there are no studies examining childhood localization related epilepsies early in its course and the neurodevelopmental course is not adequately defined. However, abnormalities at the cellular level affecting the neuronal microstructure may be associated with these syndromes, particularly in one of the most common forms of epilepsy in children - benign epilepsy with centro-temporal spikes (BECTS). The main hypothesis was to assess whether focal discharges in BECTS result in structural abnormalities undetectable on the conventional MRI examination, notably of the white matter.
Methods: In this study, we explored the white matter in 25 children suffering from BECTS) and in 24 age- and sex matched controls. We applied a voxel based morphometry technique to explore the integrity of white matter. For this purpose we used a region-of-interest approach as well as voxel-based analysis to detect regions with significant group difference. Secondly, we investigated whether epilepsy related clinical variables might contribute to any possible alterations of white matter.
Results: Significant differences of DTI-derived parameters (fractional anisotropy (FA) and mean diffusivity (MD)) were observed between BECTS and healthy groups mainly within the areas that surround left rolandic sulcus. Furthermore, decreased fractional anisotropy and increased mean diffusivity around these areas was prevalent in the sub-group of patients with duration of epilepsy was more than 12 month.
Conclusions: White matter integrity is compromised in BECTS. DTI seems sensitive enough to detect structural changes of the white matter undetectable with conventional MRI.

P3.028
Gabaergic interneuron odyssey during cerebellar development

Cadilhac, C. (Montpellier)[1], Bachy, I. (Montpellier)[1], Jahannault-Talignani, C. (Montpellier)[1], Ango, F. (Montpellier)[1]
[1]*Institut de Génomique Fonctionnelle, Montpellier, France*

Neural circuits formation and function imply the fine coordination of multiple developmental processes such as cell migration and differentiation, cell type recognition and targeting and eventually synapse formation and maturation.
Mouse cerebellum develops post-natally and thus provides us a powerful model to analyse those processes. Cerebellar circuit complétion occurs within 3 weeks after birth and gives rise to a prototypical 3 layers organization : The external granule cell layer (eGCL) - the molecular layer (ML) - the internal granule cell layer (iGCL). In this context, GABAergic interneurons development has been described to occur in an inside-out manner but the molecular and cellular mechanisms of their migration remain poorly elucidated.
Here, we used a multidisciplinary approach to better understand GABAergic interneurons migration in developing cerebellum. In particular, we present a live imaging study that unravels a tangential migration path that was never described before in cerebellum.

P3.029
The Coffin-Lowry syndrome-associated protein RSK2 regulates neurite outgrowth through phosphorylation of PLD1 and synthesis of phosphatidic acid

Ammar, M.R. (Strasbourg)[1]
[1]*INCI Strasbourg, Strasbourg, France*

More than 80 human X-linked genes have been associated with mental retardation (MR) and deficits in learning and memory. However, most of the identified mutations induce limited visible morphological alterations in brain organization and the molecular bases underlying the neuronal clinical features remain elusive. We show here that neurons cultured from mice deficient for ribosomal S6 kinase 2 (*Rsk2*), a model for the Coffin-Lowry syndrome (CLS), exhibit a significant delay in development in a pattern very similar to that shown by neurons cultured from phospholipase D1 (*Pld1*) knockout mice. We found that gene silencing of *Pld1* or *Rsk2* as well as acute pharmacological inhibition of PLD1 or RSK2 strongly impaired neuronal growth factor (NGF)-induced neurite outgrowth from PC12 cells. NGF triggered RSK2-dependent phosphorylation of PLD1 leading to its activation and the synthesis of phosphatidic acid at the site of neurite growth. NGF-induced neurite outgrowth was severely inhibited in PC12 cells silenced for RSK2 expression, but this phenotype could be rescued by expression of a phosphomimetic PLD1 mutant, revealing that PLD1 is the major target for RSK2 in neurite formation. Additionally, TIRF microscopy experiments revealed that RSK2 and PLD1 positively control fusion of VAMP-7 vesicles at the site of neurite outgrowth. We propose that the loss of function mutations in *RSK2* that leads to CLS and neuronal deficits are related to defects in neuronal growth due to impaired RSK2-dependent PLD1 activity resulting in reduced VAMP-7 vesicle fusion rate and membrane supply.

P3.030
Developmental effects of fluoxetine on the serotonergic system

Martinelli, E. (Paris)[1], Ledonné, F. (Paris)[1], Gaspar, P. (Paris)[1]
[1]*Institut du Fer à Moulin INSERM S 839, Paris, France*

Serotonin (5-HT) has an important role as a trophic factor during development, including neuronal proliferation, migration, differentiation, and synaptogenesis. Many of these regulatory roles persist after birth. Selective 5-HT reuptake inhibitors (SSRIs) are among the most frequently prescribed

treatments for psychiatric conditions such as, anxiety and depression, and achieve a therapeutic effect by increasing serotonergic tone. In rodents, exposure to SSRIs during early postnatal period alters emotional behavior during adulthood, and predisposes the animal to depression and anxiety-like behaviors. The underlying mechanisms are not well understood, but there appears to be a critical period from postnatal day 4 to 21 (P4 to P21), during which this effect occurs. Our study aims to better characterize the morphological and biochemical changes in response to SSRI exposure during this critical period that are implicated in the disturbed behavior in adulthood. 5-HT neurons are generated early in development and reach their targets between embryonic days 11 to 15 in the mouse. The neurons that produce 5-HT are located in the raphe nuclei of the brainstem and innervate targets throughout the central nervous system. For our study, we have chosen to focus on maturation in the prefrontal cortex and hippocampus. We measured the density of 5-HT fibers in these regions in mice at different postnatal ages during the critical period and adulthood. The 5-HT fiber morphology was visualized after immunolabelling and DAB revelation, and relative axon fiber density was quantified. The density of 5-HT fibers increases most significantly between the ages of P14 and P21. At day P21 the 5-HT fiber density is mature and is maintained through adulthood. To measure the effects of SSRI exposure during this critical period, mice pups will receive daily subcutaneous injections of fluoxetine (10 mg/ kg). The density of fibers in the prefrontal cortex and hippocampus will be quantified at P21. Our results may provide an understanding of the anatomical changes due to exposure to fluoxetine during early postnatal period, and may give important information to reevaluate the safety of SSRI use during pregnancy and infancy.

P3.031

Presynaptic release from retinal axons is important for their refinement

Rebsam, A. (Paris)[1], Assali, A. (Paris)[1], Bennis, M. (Marrakesh)[2], Kaeser, P. (Stanford)[3], Südhof, T. (Stanford)[3], Gaspar, P. (Paris)[1]
[1]Institut du Fer à Moulin, INSERM U 839, Paris, France, [2]Cadi Ayyad University, Marrakesh, Morocco, [3]Howard Hughes Medical Institute, Stanford University, Stanford, United States

Projections of Retinal Ganglion Cells (RGCs) in their main targets, the dorsal Lateral Geniculate Nucleus (dLGN) and the Superior Colliculus (SC), are organized in eye-specific domains and with precise topography. In both cases, projections are initially intermingled and are refined into their final territories during the first postnatal weeks. Alteration of spontaneous neural activity in the retina disrupts both eye-specific segregation and retinotopy. However, the cellular mechanisms linking neural activity to map refinement remain poorly understood.
Here we examine the role of presynaptic release during development in the refinement of retinal projections to the dLGN and SC. To distinguish between the synaptic and non-synaptic effects of activity blockade, we perturbed specifically the presynaptic release at retinal terminals. This was obtained by a conditional deletion (Cre/LoxP system) of *Rim 1* and *2* in RGCs, using a SERT-Cre (serotonin transporter) mouse line. Although SERT expression was reported to be restricted to ipsilateral RGCs, we found that most RGCs show effective recombination in SERT-Cre mice. The removal of Rim proteins is known to strongly reduce calcium-dependent neurotransmitter release, without affecting spontaneous release. Our tracing studies indicated that Rim conditional double knock out (Rim cDKO) mice have defects in eye-specific segregation in the dLGN but no major topographic defects in the SC. This result suggests that segregation but not gross topography involves calcium dependent synaptic release. Interestingly, ipsilateral projections in the SC do not form patches and extend more laterally in Rim cDKO compared to control mice at P27, suggesting that ipsilateral projections could be more sensitive to the perturbed presynaptic release. Focal injections and electroporations in the retina are in progress to assess the organization of axon arbors and presynaptic terminals in both contralateral and ipsilateral axons.
Our results show that synaptic release of RGCs is important for eye-specific segregation and for the organization of ipsilateral projections.

P3.032

Synaptic role of microtubule associated protein MAP-6 (STOP) in dendritic spine plasticity by actin dynamic regulation

Peris, L. (Grenoble)[1], Bisbal, M. (Grenoble)[1], Seggio, M. (Grenoble)[1], Elie, A. (Grenoble)[1], Brocard, J. (Grenoble)[1], Bosc, C. (Grenoble)[1], Deloulme, J.C. (Grenoble)[1], Delphin, C. (Grenoble)[1], Arnal, I. (Grenoble)[1], Andrieux, A. (Grenoble)[1]
[1]*Grenoble Institut des Neurosciences, U 836 UJF CEA CHU, Grenoble, France*

Alterations of cytoskeleton have been involved in several neurodegenerative pathologies. Our laboratory has provided one of the first experimental evidence for a role of microtubules in mental functions by developing a mouse model deficient for a microtubule associated protein MAP-6 (known as STOP).
MAP-6-null mice were described as animal model of schizophrenia. They exhibited alterations of brain functions with neurotransmission anomalies associated with severe behavioral deficits that react positively to antipsychotic drugs such as neuroleptics. These behavioural deficits could be associated with synaptic defects affecting both short-term and long-term synaptic plasticity. However little is known about the biological function and the cellular targets of MAP-6 protein in neurons.
The proposal of this work is to evaluate cytoskeleton function in dendritic spine formation in MAP-6-deficient neurons and to analyze the role of MAP-6 proteins in synaptic plasticity, all events that are defective in neuronal pathologies. Here we show that the different MAP-6 isoforms are developmentally regulated. At early stages, E-MAP-6 localizes mainly to the axon. Later, N-MAP-6 localizes to the axon, dendrites and dendritic spines. MAP-6 null neurons have less dendritic spines (*in vivo* and *in vitro*), and the overexpression of the main MAP-6 isoforms rescues dendritic spines number. Using knockdown and overexpression experiments we show that MAP-6 proteins are important for dendritic spines formation and stabilization. The overexpression of different MAP-6 constructs allowed us to determine the minimal region of MAP-6 responsible of this activity (MAP-6 5R). At cellular level, MAP-6 5R colocalizes with actin cytoskeleton and might be responsible of MAP-6-actin interaction previously described. Biochemical analysis with purified MAP-6 and actin are being done in our laboratory to confirm this observation.
This work provide a better understanding of how a microtubule associated protein, MAP-6, is involved in synaptic plasticity probably by regulating actine dynamics in dendritic spines. Finding targets that modulate cytoskeletal dynamics might provide successful treatment of pathologies where synaptic connectivity is dysfunctional, like schizophrenia.

P3.033

***Arc* expression identifies the neuronal ensemble within the lateral amygdala recruited during fear conditioning**

Gouty-Colomer, L.-A. (Rotterdam)[1], Hosseini, B. (Rotterdam)[1], Colomer, C. (Rotterdam)[2], Marcelo, I. (Rotterdam)[1], Schreiber, J. (Rotterdam)[2], Houweling, A. (Rotterdam)[2], Elgersma, Y. (Rotterdam)[2], Kushner, S.A. (Rotterdam)[1]
[1]*ERASMUS - University Medical Center, Dept of Psychiatry, Rotterdam, Netherlands,* [2]*ERASMUS - University Medical Center, Dept of Neuroscience, Rotterdam, Netherlands*

The biology of memory is among the most fundamental questions in modern neuroscience. Fear conditioning produces a profound memory with a robust behavioral phenotype (cued freezing) gated by synaptic changes in the lateral amygdala. However, further progress has been limited by the sparse encoding of fear memory, since only a minority of neurons (10-20%) undergo synaptic plasticity during fear conditioning. Moreover, a similar proportion of neurons express the immediate-early gene Arc after fear conditioning, consistent with its well-known role in memory consolidation. We used Arc-dVenus reporter mice to visually-identify cells and electrophysiologically characterize neuronal ensembles specifically recruited during fear learning. Stereological quantification and fluorescence

analysis of dVenus positive cells show that a single tone-shock pairing is sufficient to induce a qualitative change in the population of Arc-expressing cells in the lateral amygdala. Increasing the number of pairings induces a further increase fluorescence intensity of dVenus+ neurons.
Interestingly, Arc+ neurons display specific intrinsic properties independent of sensory stimulation or learning, as shown by their increased excitability compared to their Arc- neighbors. Importantly, fear learning specifically induces a strong potentiation of thalamic afferent synapses, selectively in the Arc+ cell population. These cells express increased amplitude of evoked excitatory postsynaptic currents, without modification of the paired-pulse ratio, suggesting postsynaptic changes highly specific to fear conditioning.
In conclusion, we show that sensory stimuli activate Arc+ cells exhibiting particular intrinsic properties. In contrast, fear conditioning induces the establishment of a specific and defined population of Arc-expressing cells that selectively undergo strong synaptic potentiation. Taken together our results suggest that dVenus expression can be used to identify neuronal ensembles functionally participating in the encoding of a fear memory.

P3.034

The transcription factor Meis1 controls cardiac sympathetic innervation and synaptogenesis independently of noradrenergic specification

Marmigère, F. (Montpellier)[1], Bouilloux, F. (Montpellier)[1], Thireau, J. (Montpellier)[2], Karam, S. (Montpellier)[2], Richard, S. (Montpellier)[2]
[1]U 1051, INSERM, Montpellier, France, [2]U 1046, Inserm, Montpellier, France

The autonomic sympathetic nervous system regulates involuntary physiological functions such as respiration, cardiac rhythm, sudation or salivation. These neurons are contacted by preganglionic motor neurons located in the medial spinal cord and project peripherally to their target organs in the whole body. During embryonic development, a complex cross regulatory network of transcription factors is responsible for early noradrenergic specification of these neurons mainly by promoting the expression of Tyrosine Hydroxylase (TH), a key enzyme of monoamine biosynthesis. NGF (Nerve Growth factor) signaling through TrkA receptor serves as a major long range survival and axonal growth factor for these neurons, whereas short distance axonal guidance is mediated by Sema3a through activation of Neuropilin-1 (Nrp-1) receptor. Whereas the transcriptional cascade governing noradrenergic specification has been extensively studied, transcription factors regulating axon guidance and growth of sympathetic neurons remain to be discovered.
We show that the transcription factor Meis1 is essential for target innervation by sympathetic neurons during embryonic development. Specific Meis1 ablation in neural crest cells did not affect early proliferation and noradrenergic specification. Instead, Meis1 governs a large program of axonal growth and in its absence, sympathetic neurons failed to reach their terminal peripheral targets. We conclude that Meis1 is part of a transcriptional program uncoupling sympathetic neurons axonal growth and synaptogenesis from neurotransmitters specification.
Finally, most of the mice deleted with Meis1 in the PNS massively died at birth. Surviving animals exhibited sudden and unpredictable death within the three first months following birth. Because the Meis1 human locus has been identified as a risk factor for sudden cardiac death, we investigated cardiac functions in young adult $HtPA^{CRE}/Meis1^{LoxP/LoxP}$ mutant mice. We demonstrate that $HtPA^{CRE}/Meis1^{LoxP/LoxP}$ displayed spontaneous bradycardia and severe sino- and atrio-ventricular conduction defects. These results strongly suggest that altered Meis1 expression and/or function in human could have serious harmfull consequences from chronotropic incompetence to death according to the imposed stress.

P3.035

Semaphorin 3B mediated extracellular signalling regulates neural progenitors division

Castellani, V. (Villeurbanne)[1], Arbeille, E. (Villeurbanne)[1], Sanyas, I. (Villeurbanne)[1], Bozon, M. (Villeurbanne)[1], Kindbeiter, K. (Villeurbanne)[1], Moret, F. (Villeurbanne)[1]
[1]*CGphiMC - Univ. Claude Bernard Lyon I, CNRS UMR 5534, Bât.Mendel 741, Villeurbanne, France*

The orientation of cell division has a major impact on tissue tissue architecture, and cell fate choices. During CNS development in vertebrates, the growth of the neural tube and the generation of neuron and glial cells results from the proliferation of neural progenitors organized in a neuroepithelium closed around a central canal. The orientation of the mitotic spindle with respect to the apico-basal axis is important for the conservation of the integrity of the neuroepithelium and influences the fate of daughter cells. Previous studies mainly focused on the role of intracellular factors, like LGN, Numa and small G proteins in this orientation. In contrast, contribution of extracellular cues to the orientation of the mitotic spindle has been fewly explored so far. Given that the central canal is a source of major developmental factors like morphogens, we wondered whether extracellular diffusible signals could also be present which would contribute to the orientation of neural progenitor divisions. We show that dorsally open neural tubes from E10.5 mice maintained in short term culture displayed a strong increase in the percentage of oblique divisions compared to un-open ones. Lumen-derived signals are thus required for neural progenitors to achieve planar divisions in the mouse spinal neuroepithelium at the onset of neurogenesis. Among these signals, we identified Sema3B, a secreted member of the Semaphorin family involved in axon guidance and cytoskeleton remodeling. First, we detected Sema3B mRNA in floor plate cells which suggests that it could be secreted in the lumen of the spinal cord. Second, short term exposure of open neural tubes to exogenous Sema3B restored planar divisions in a large population of spinal progenitors. Third, invalidation of the Sema3B gene resulted in a decrease in the percentage of planar divisions for profit of oblique divisions in E10.5 spinal progenitors without alteration of progenitor number or polarity. These results thus reveal that beyond its role as morphogen-releasing organizer, the floor plate also provides extracellular signaling which controls the orientation of neural progenitor division.
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P3.036

Manipulating serotonin supply and its effect on adult hippocampal neurogenesis

Klempin, F. (Berlin)[1], Beis, D. (Berlin)[1], Mosienko, V. (Berlin)[1], Kempermann, G. (Dresden)[2], Bader, M. (Berlin)[1], Alenina, N. (Berlin)[1]
[1]*MDC for Molecular Medicine, Berlin, Germany,* [2]*Center for Regenerative Therapies Dresden (CRTD), Dresden, Germany*

In the adult brain, neural stem cells reside in a particular niche formed by a microenvironment that promotes neuronal development. One of the intrinsic soluble neurogenic factors is serotonin (5-HT, 5-Hydroxytryptamine). This neurotransmitter has attracted much attention in the context of theories linking major depression to failing adult neurogenesis. We recently developed a mouse model deficient in serotonin-synthesizing enzyme, tryptophan hydroxylase 2 (TPH2) (*Tph2*$^{-/-}$ mice). These mice are selectively depleted in brain serotonin and exhibit extreme aggression accompanied by reduced anxiety and a depression-like state. Here we took advantage of this genetic loss-of-function model to directly test the action of the neurotransmitter in the modulation of adult hippocampal neurogenesis. We have conducted experiments to determine proliferation and differentiation of dentate gyrus precursor cells in *Tph2*$^{-/-}$ mice and their littermates at 3 different ages (P42, P80, and 1 year-old) *in vivo*. Given the significant number of studies proposing serotonin as positive neurogenic factor, we have been surprised to find that the *Tph2*$^{-/-}$ mouse has no measurable change in baseline proliferation detected by BrdU incorporation at any of the investigated time points. We suggest that lack of serotonin is compensated by other mechanisms that maintain normal rates of precursor cell proliferation. Indeed, additional validation of our results at cell level in *Tph2*$^{-/-}$ mice revealed increased proliferation accompanied with apoptosis of Sox2-positive type-2a progenitor cells. Whether lack of

serotonin has a direct and acute effect on the hippocampal niche or leads to an enduring adaptation at cell and receptor levels needs to be investigated. These experiments will add to our understanding of the pharmacology of serotonin action that could lead to alternative therapeutic strategies in depression or age-related cognitive decline.

P3.037

Type 1 metabotropic glutamate receptor 1 gates GluD2 channels at parallel fiber to Purkinje cell synapse

Ady, V. (Paris)[1], Perroy, J. (Montpellier)[2], Tricoire, L. (Paris)[3], Piochon, C. (Paris)[3], Dadak, S. (Montpellier)[2], Chen, X. (Paris)[3], Dusart, I. (Paris)[3], Fagni, L. (Paris)[4], Lambolez, B. (Paris)[3], Levenes, C. (Paris)[5]
[1]CNRS - Université Paris Descartes UMR 8119, Laboratoire de Neurophysique et Physiologie, Paris, France, [2]Institut de Génomique Fonctionnelle, Montpellier, France, [3]Université Pierre et Marie Curie-Paris 6, Paris, France, [4]Institut de Génomique Fonctionnelle, Paris, France, [5]Université Paris Descartes, Paris, France

The receptor GluD2 is a member of ionotropic glutamate receptor (iGluR) family that is almost exclusively expressed in Purkinje cells of the cerebellum. It is now understood that GluD2 regulates the formation and plasticity of parallel fiber to Purkinje cells synpase, through the extracellular amino-terminal domain and the cytoplasmic carboxy-terminal domain, respectively. However, Whether GluD2 operates as a functional ion channel has thus been a long-standing still unresolved question. Our study shows that the type 1 metabotropic glutamate receptor (mGlu1) gates the GluD2 channel. In human embryonic kidney cells (HEK293), the activation of recombinant mGlu1 elicits a current that relies on coexpression of GluD2 and has properties similar to that of GluD2-currents. In cerebellar Purkinje cells, the mGlu1-dependent current at parallel fiber synapses is reduced in mice lacking GluD2 channel pore. In both HEK293 and Purkinje cells, the mGluR1-dependent current is reduced by the expression of the dominant-negative mutant subunit GluD2 V617R. These results show that GluD2 has evolved from the iGluR family into a channel indirectly gated by glutamate through mGlu1 to sustain a slow excitatory transmission. The present work inaugurates physiological and pharmacological studies on this receptor family, offering new perspectives on receptors crosstalks, neuronal signaling and synaptic transmission.

P3.038

Functional characterization of projection-specific output neurons of mouse presubiculum

Fricker, D. (Paris)[1], Simonnet, J. (Paris)[1], Lofredi, R. (Paris)[1], Eugene, E. (Paris)[1], Cohen, I. (Paris)[1], Miles, R. (Paris)[1]
[1]CRICM - Centre de Recherche de l'Institut du Cerveau et de la Moelle Epinière, UPMC / INSERM UMR S 975 / CNRS UMR 7225, Paris, France

The Presubiculum is part of the temporal lobe circuits for spatial information processing. We previously described the functional neuroanatomy of this transitional cortical area between hippocampus and neocortex (Simonnet et al. 2012). Presubiculum is organized in six layers, with different layers containing distinct neuronal populations. Principal neurons in superficial layers (II/III) and deep layers (V/VI) are regular spiking, and we discovered a population of intrinsic bursting pyramidal neurons in layer IV. Multimodal information from several upstream brain regions converges in Presubiculum, which contains head direction cells, grid cells, and border cells (Taube et al. 1990, Boccara et al. 2011).Thalamic nuclei convey vestibular information, while visual information is relayed from visual and retrosplenial cortices. These signals are integrated within the presubicular microcircuit to give a

specific output signal to downstream regions. It has been shown that the Anterodorsal Nucleus of the thalamus (ADN) and the Lateral Mammilary Nucleus (LMN) receive information from distinct presubicular neuronal populations (Yoder and Taube, 2011). The specific features of signals carried by these two pathways are not known.
Stereotaxic injection of retrograde tracers (retrobeads) into either LMN or ADN revealed two specific neuronal subpopulations in the presubiculum. Stained neurons were recorded in the slice preparation using the patch clamp technique, to examine electrophysiological and morphological properties. All retrogradely labeled neurons after a LMN injection were layer IV intrinsic bursting neurons. Non-labeled neurons in layers IV were regular spiking. Labeled neurons following ADN injections were regular spiking neurons in deep layers of the presubiculum.
We conclude that projection neurons targeting LMN and AND are two distinct populations with specific neuronal properties. It will be interesting to examine how output signal specificity may be generated within the presubicular microcircuit.

P3.039

A new look into mechanisms underlying mGluR4 actions at parallel fiber - molecular layer interneuron synapses in the rat cerebellum

Bessiron, T. (Orsay)[1], Mclean, H. (Orsay)[1], Daniel, H. (Orsay)[1]
[1]*IBBMC, Pharmacologie et Biochimie de la Synapse, cnrs 8619, Orsay, France*

mGluR4 receptors are important modulators of synaptic transmission in the cerebellar cortex. Located on Parallel Fiber (PF) terminations, their activation reduces the liberation of Glutamate at the PF-PC synapse. PF also contact other cell types including Molecular Layer Interneurons (MLI), namely Stellate Cells and Basket Cells. Given the important role of MLI in controlling PC firing, we first determined that PF terminals at these synapses express functional mGluR4. We then took advantage of certain properties of this synapse in order to further understand the molecular mechanisms by which mGluR4 reduces Glutamate release.
Contrary to the PF-PC synapse which responds to single supra-threshold stimuli with liberation of Glutamate from multiple release sites, PF-MLI synapses respond with fewer release sites (Crowley et al., 2007), leading to smaller responses with variable amplitudes and latencies. Interestingly, these properties, which could be linked to the composition of the exocytosis machinery, may be responsible for glutamate release failures and which can be readily quantified.
Pharmacological activation of mGluR4 with L-AP4, a broad-spectrum group III mGluR agonist, reversibly increased the Failure rate at PF-MLI synapses, indicative of possible effects on exocytotic mechanisms. Using a paired-pulse paradigm, we aimed to assess whether or not varying the extracellular calcium concentration could modulate the effect of L-AP4 on evoked (e) presynaptic calcium transients, eEPSC amplitude, Paired-Pulse Facilitation (PPF) and Failure rate. We show that while the extent of L-AP4 depression of evoked presynaptic calcium transients depends little on extracellular calcium concentration, reducing extracellular calcium potentiates the L-AP4 depressant effect on eEPSC amplitude and increases the PPF, while increasing extracellular calcium potentiates the effect of L-AP4 on the Failure rate.
This suggests that mGluR4 activation not only inhibits calcium influx, but may also directly modify exocytotic mechanisms. This hypothesis is supported by a recent biochemical study from our group (Ramos et al., 2012) revealing interactions between native mGluR4 and various components of this exocytosis machinery in the cerebellum.

P3.040

Internalization and down-regulation of the ALK receptor in neuroblastoma cell lines upon monoclonal antibodies treatment

Vigny, M. (Paris)[1], Mazot, P. (Paris)[2], Cazes, A. (Paris)[3], Janoueix-Lerosey, I. (Paris)[4]

[1]INSERM/UPMC, Paris, France, [2]INSERM U 839/UPMC, Paris, France, [3]Institut Curie, Centre de Recherche, Inserm U 830 Institut Curie, Paris, France, [4]Institut Curie, Centre de Recherche, Inserm U 830, Paris, France

Activating mutations of the full length ALK receptor, with two hot spots at positions F1174 and R1275, have been characterized in sporadic cases of neuroblastoma. Using stably transfected NIH3T3 cells expressing mutated ALK at position F1174 or R1275, we recently established that the constitutive kinase of the mutated receptors impaired their trafficking, with retention in intracellular compartment. Strikingly intracellular miss-localized receptors appeared much less phosphorylated than the cell surface pool (Mazot et al. 2011). Here, we report similar basal patterns of ALK phosphorylation between the neuroblastoma IMR-32 cell line which expresses only the wild-type receptor (ALK^{WT}) and the SH-SY5Y cell line which exhibits a heterozygous ALK F1174L mutation and expresses both ALK^{WT} and ALK^{F1174L} receptors. We demonstrated that this lack of detectable increased phosphorylation in SH-SY5Y cells is a result of intracellular retention and proteasomal degradation of the mutated receptor. As a consequence, in SH-SY5Y cells, plasma membrane appears strongly enriched for ALK^{WT} whereas both ALK^{WT} and ALK^{F1174L} were present in intracellular compartments. We further explored ALK receptor trafficking by investigating the effect of agonist and antagonist mAb (monoclonal antibodies) on ALK internalization and down-regulation, either in SH-SY5Y cells or in cells expressing only ALK^{WT}. We observe that treatment with agonist mAbs resulted in ALK internalization and lysosomal targeting for receptor degradation. In contrast, antagonist mAb induced ALK internalization and recycling to the plasma membrane. This study provided novel insights into the mechanisms regulating ALK trafficking and degradation, showing that various ALK receptor pools are regulated by proteasome or lysosome pathways according to their intracellular localization. The lack of detectable phosphorylation of the intracellular mutated receptor in SH-SY5Y cells may rely on a permanent dephosphorylation by specific tyrosine phosphatases. This point is currently under investigation.

P3.041

Thalamocortical inputs from the orexin-sensitive rhomboid nucleus elicit long-lasting recurrent excitation in parietal cortical circuits

Hay, A. (Paris)[1], Lambolez, B. (Paris)[1]
[1]Université Pierre et Marie Curie Laboratoire Neurobiologie des Processus Adaptatifs, Equipe Réseau Cortical et Couplage Neurovasculair, Paris, France

The thalamus is the major information gateway to the cerebral cortex. Thalamic “specific” sensory nuclei send transient, stimuli-driven, signals to their cognate sensory cortical areas and elicit brief responses time-controlled by local circuit mechanisms. Conversely, intralaminar and midline thalamic nuclei are thought to convey tonic information on behavioral and environmental conditions but little is known on their effects on cortical circuits. These nuclei have been viewed as a “non-specific” system of widespread and diffuse thalamocortical (TC) projections regulating arousal and. However, more recent studies show that different non-specific nuclei target distinct cortical areas and serve distinct functions. A clear example of this anatomo-functional specialization is the midline rhomboid nucleus (Rh). Rh is a preferred target of hypothalamic orexins, which promote arousal and vigilance, and projects to bilateral parietal and somatosensory cortices. Growing evidence implies the Rh in multimodal sensory processing and in spatial memory, in accord with the functional specialization of its cortical target areas.
Here, we performed selective optogenetic stimulation of Rh TC fibers in slices of parietal and somatosensory cortices and characterized light-evoked responses using patch-clamp recordings. We found that light-induced synaptic glutamatergic currents involved both AMPA and NMDA receptors and exhibited pronounced depression upon trains of stimuli. Rh projections innervated both excitatory and inhibitory cell types in all supra- and infragranular layers but targeted to a lesser extend layer II/III interneurons and virtually avoided layer IV. Rh TC inputs elicited a low level of feedforward inhibition due to a weak feedforward recruitment of FS interneurons. As a consequence, Rh TC inputs induced

prolonged recurrent excitation of the cortical network that summated to trigger action potentials with a large jitter.

P3.042

Structural dynamics and heteromerization properties of metabotropic glutamate receptors

Scholler, P. (Montpellier)[1,2], Doumazane, E. (Montpellier)[1], Fabre, L. (Montpellier)[1,2], Donsimoni, G. (Codolet)[2], Roux, T. (Codolet)[2], Zwier, J.M. (Codolet)[2], Trinquet, E. (Codolet)[2], Pin, J.-P. (Montpellier)[1]
[1]Institut de Génomique Fonctionnelle - CNRS UMR 5203, INSERM U 661, Univ. Montpellier, Montpellier, France, [2]Cisbio Bioassays, Codolet, France

A fine tuning of neurotransmitter receptors activation mechanism is essential for their physiological properties. Allosteric effects, either inside the receptor itself upon ligand binding, or upon action of partner proteins, are a key point. For example, metabotropic glutamate receptor subtype 2 (mGlu2) is under investigation to find allosteric modulators that may display antipsychotic activity. The eight mGluRs (1-8), members of the super-family of G protein coupled receptors (GPCR), are complex machineries composed of several domains interacting together through allosteric processes. They are stable dimers and both subunits contain an extracellular domain (ECD), where glutamate and orthosteric ligands bind, and a transmembrane domain (TMD) responsible for downstream signaling and allosteric modulations. How these two domain domains communicate together, and how this leads to receptor activation remain poorly understood.
Here we describe a powerful time-resolved FRET (TR-FRET) approach which allows, all directly at the surface of live cells:
1) to study conformational changes of the extracellular domain upon ligand binding,
2) to screen for new orthosteric or allosteric ligands independently of a signaling pathway in a HTS compatible manner, and
3) to analyze the formation of heterodimers and their conformational changes upon activation.
Briefly, we show that a large relative movement of the ECDs is a key step in the dimeric receptor activation, as revealed by changes in the intramolecular FRET efficacy. The effect of specific mutations, allosteric modulators binding in the TMD, or the effects of the G protein are also investigated for their influence on the ECD conformations. Also, using a new multiple labeling approach, we show the formation of heterodimeric mGluRs, and their existence is further confirmed by biochemical and complementation studies. The analysis of the activation mechanism of the mGlu2-4 heterodimer with our conformational sensor reveals an asymmetrical functioning. All these results bring much information on the activation mechanism of mGluR, and demonstrate that our TR-FRET strategy is a new powerful approach to study the formation and the activation mechanisms of class C GPCRs in live cells, offering possibilities to find new ligands.

P3.043

Synaptic properties of nucleus tractus solitarii neurons are target specific

Strube, C. (Marseille)[1], Gackière, F. (Marseille)[1], Saliba, L. (Marseille)[1], Penalba, V. (Marseille)[1]
[1]Université Aix-Marseille - CNRS, CRN2M, UMR 7286, Marseille, France

Located in the brainstem, the nucleus tractus solitarius (NTS) is the major gateway through which visceral primary afferent information enters the brain, is integrated, and ultimately relayed towards other brain structures. It elaborates two types of messages: those sent to the caudal ventrolateral medulla (CVLM) giving rise to autonomic reflexes and those transmitted to the brain via ascending pathways which includes indirect projections via the parabrachial nucleus (PBN) involved in visceral

perception and participating to self-awareness. Second order NTS neurons possess both AMPA and NMDA receptors (AMPAR, NMDAR). These ionotropic receptors underly currents with different biophysical properties and can thus give rise to different excitatory neurotransmission modes. Therefore, we decided to determine the relative contribution of AMPA and NMDA receptors to excitatory post-synaptic currents (EPSCs) in NTS neurons projecting to CVLM or to PBN, after identification by retrograde labeling.
Most of the recorded neurons display EPSCs with both AMPAR and NMDAR components (iAMPA and iNMDA). Miniature EPSCs recorded from a single neuron reveal that iNMDA amplitude is strongly correlated to iAMPA amplitude, suggesting that all the synapses recorded from the same neuron have a similar proportion of NMDAR / AMPAR. However, iNMDA / iAMPA ratio recorded at +40 mV is highly variable from one neuron to another, ranging from 0 to almost 1. More interestingly, the value of this ratio recorded in CVLM-projecting-neurons is significantly smaller than in PBN-projecting-neurons. Moreover, iAMPA recorded at -70 mV displays different amplitude and kinetics, depending on the projection pathway. These differences suggest that synaptic processing and plasticity could involve different mechanisms in neurons participating to autonomic reflexes and visceral perception.

P3.044

New developments to study neuronal circuits using recombinant rabies virus technology

Frick, A. (Bordeaux)[1], Haberl, M.G. (Bordeaux)[1,2], Silva, S.M. (Bordeaux)[3], Ginger, M. (Bordeaux)[1,3], Mulle, C. (Bordeaux)[3], Conzelmann, K.-K. (Munich)[4]
[1]Neurocentre Magendie, Bordeaux, France, [2]University of Zürich, Zürich, Switzerland, [3]CNRS UMR 5297, Interdisciplinary Institute of Neuroscience, Bordeaux, France, [4]Gene Centre Munich, Ludwig Maximilian University, Munich, Germany

An understanding of how the brain processes information requires knowledge of the architecture of its underlying neuronal circuits, as well as insights into the relationship between architecture and physiological function. A range of sophisticated tools is needed to acquire this knowledge, and recombinant rabies virus (RABV) is becoming an increasingly important part of this essential toolbox. The glycoprotein deleted (DG) RABV has permitted a range of approaches for the study of defined neuronal circuits. Here we report a novel RABV DG vector that infects cell bodies permitting anterograde tracing of fine-detailed neuronal structure. We will show how this technique can be applied in various brain regions in combination with a variety of imaging/analysis tools to gain knowledge of neuronal morphology and connectivity. This vector can be readily combined with retrograde labeling or mono-trans-synaptic tracing, complementing the existing toolbox for dissecting brain anatomy and physiology.

P3.045

Glial Expression of Split-ends regulates sensitivity in *Drosophila* Parkinson disease models

Goubard, V. (Lyon)[1], Querenet, M. (Lyon)[1], Mollereau, B. (Lyon)[1], Davoust, N. (Lyon)[1]
[1]Ecole Normale Supérieure de Lyon, CNRS UMR S 239-Biologie Moléculaire de la Cellule, Lyon, France

Parkinson's disease (PD) is the second most prevalent neurodegenerative disease after Alzheimer disease. It is characterized by the selective loss of dopaminergic neurons in the *substantia nigra pars compacta*. Dopaminergic signaling deregulation leads to resting tremor and postural instability. Mutations in PARK genes such as Pink1, as well as environmental cues like the use of the pesticide Paraquat, have been shown to trigger PD. Although several key players have been identified and

characterized in rodent models of PD, little is known about the mechanisms that regulate survival and death of dopaminergic neurons.
Drosophila is another model of choice to understand the mechanisms of the Parkinsonian pathology. Most of the genes involved in familial PD have been studied in *Drosophila* PD models. We used *Drosophila* to identify new genes involved in dopaminergic neuronal cell death induced by Paraquat treatment.
From a candidates-based genetic screen, we identified Split-ends (spen), a nuclear protein expressed in both neuronal and glial cells in *Drosophila* adult brains. Spen is the *Drosophila* homolog of human SHARP. Fly mutant carrying a loss of function mutation in *spen* exhibits a dominant increased sensitivity to Paraquat treatment. Importantly, specific depletion of spen in glial cells by RNAi also renders flies more sensitive to Paraquat, while neuronal depletion of spen does not affect sensitivity to Paraquat. Furthermore, spen expression is modulated upon Paraquat treatment. Our results suggest a specific role of spen in glial cell in Paraquat resistance. Furthermore, we showed that spen and pink1 interact genetically in that spen mutant enhances the down-turned wing and collapsed thorax phenotypes observed in pink1 mutant.
Together, our experiments suggest that the expression of spen is required for cellular resistance in pharmacological and genetic models of PD. In *Drosophila* brain, spen is required in glial cells for resistance to Paraquat treatment, which suggests a role of glial cells in neuron survival.

P3.046

Lack of AHNAK affects adhesion, motility, and mechanical properties of Schwann cells

Nothias, F. (Paris)[1], Soares, S. (Paris)[1], Féréol, S. (Créteil)[2], Fodil, R. (Créteil)[2], Bartolami, S. (Montpellier)[3], Taxi, J. (Paris)[1], Tricaud, N. (Montpellier)[3], von Boxberg, Y. (Paris)[1]
[1]CNRS-UMR 7224, INSERM U 952, UPMC, Team2: Axon Regeneration & Growth, Paris, France, [2]University Paris East Creteil (UPEC), INSERM U 955, CNRS ERL 7240, IMRB, Créteil, France, [3]U 1051, INSERM, Equipe Avenir, Molecular Mechanisms of Myelination/Demyelination, Montpellier, France

Proper function of SC in intact and regenerating peripheral nerve relies on their polarized interaction with axons, and with basal lamina. Perturbation of these interactions due to absence or mutation of basal lamina component laminin, or its receptors in SC, leads to severe defects in axon sorting and myelination, and is the cause for peripheral neuropathies and muscular dystrophies. AHNAK, a 700 kDa protein with a large central part composed of repeat units, has been studied mainly in epi- and endothelial cells and muscle, but its precise function remains largely unknown. We previously reported that AHNAK is constitutively expressed in SC, and that siRNA-interference with *ahnak* expression in primary cultured SC cells affected their morphology and adhesion on laminin, concomitant with altered distribution and expression of laminin receptor ß-dystroglycan. Here, we show by electron microscopic analysis of sciatic nerve from AHNAK-ko mice that myelination, and enwrapping of non-myelinated sensory fibers are affected, suggestive of abnormal fiber sorting. *In vivo*, acute shRNA silencing of *ahnak* in myelinating SC of mouse sciatic nerve during postnatal development leads to a drastic reduction of SC elongation on axons, i.e. shorter internodes, the consequence of which would inevitably be impaired nerve impulse conduction. Based on these observations, and the co-immunoprecipitation of ß-dystroglycan with AHNAK, we postulated that AHNAK should play a role in SC motility, which we confirmed by demonstrating that in a migration assay, AHNAK-ko SC exhibited a migration velocity decreased by 35-40% compared to wildtype SC. Furthermore, atomic force microscopy analysis revealed that mechanical properties of AHNAK-ko SC differ from those of wildtype.
Our data suggest that AHNAK serves as a scaffold protein that by linking the SC basal lamina to the cortical actin cytoskeleton, participates in the transduction of external signals into morphofunctional changes within the SC, and may therefore be involved in the pathophysiology of peripheral neuropathies and muscular dystrophies.

P3.047

Quantitative neurotransmitter detection on the AniRA-neurochem technological platform

Marinesco, S. (Lyon)[1], Meiller, A. (Lyon)[2], Debilly, G. (Lyon)[2], Parrot, S. (Lyon)[2], Denoroy, L. (Lyon)[3], Renaud, B. (Lyon)[2]
[1]Univ. Claude Bernard, Inserm U 1028 - Cnrs Umr 5292 - Crnl, phys. Intégrée Du Système D'éveil, Lyon, France, [2]Univ. Claude Bernard, Inserm U 1028 - Cnrs Umr 5292 - Crnl, Plate-forme technologique AniRA-Neurochem, Lyon, France, [3]Univ. Claude Bernard, Inserm U 1028 - Cnrs Umr 5292 - Crnl, Team BioRan, Lyon, France

Detecting neurotransmitters and quantifying their concentrations is important for understanding the signaling processes taking place in the central nervous system. The AniRA- NeuroChem technological platform is devoted to the *in vivo, in vitro and ex vivo* study of neurotransmission, by quantifying the amounts of neurotransmitter and metabolic molecules in different brain areas. It presently offers the following services:
- Microanalysis of neurotransmitters by capillary electrophoresis with laser-induced fluorescence detection or capillary liquid chromatography with electrochemical detection in brain microdialysates, cell cultures or tissues. Neurotransmitters presently analyzed by capillary electrophoresis are excitatory (glutamate, aspartate) and inhibitory (GABA) amino acids. Capillary liquid chromatography can quantify serotonin or catecholamine contents in minute brain samples. These separative micro-methods are especially suited for analyzing microdialysates or whole tissue homogenates sampled from the brain of laboratory animals;
- In situ bio-sensors measurement of endogenous molecules in freely-moving or anaesthetized rodents. Nitric oxide (NO) and its derivatives can be monitored *in vivo* using carbon fiber microelectrodes covered with nickel-porphyrin and Nafion. Enzymatic biosensors are platinum microelectrodes covered with poly-phenylenediamine and a specific oxidase enzyme allowing the recognition of neurotransmitters and metabolites such as glucose, lactate, D-serine and glutamate;
The AniRA-Neurochem facility can collaborate with French or foreign laboratories outside Lyon by quantifying neurotransmitters on frozen samples, or by shipping biosensors.

P3.048

Altered surface interplay between NMDA and dopamine receptors in a neuropsychiatric disorder

Jezequel, J. (Bordeaux)[1], Mikasova, L. (Bordeaux)[1], Bouchet, D. (Bordeaux)[1], Rogemond, V. (Lyon)[2], Honnorat, J. (Lyon)[2], Groc, L. (Bordeaux)[1]
[1]Institut Interdisciplinaire de Neurosciences, CNRS UMR 5297, Bordeaux, France, [2]Lyon Neuroscience Research Center, INSERM U1028, CNRS UMR 5292, Lyon, France

Glutamatergic N-Methyl-D-Aspartate receptors (NMDAR) play a key role in many physiological processes. A disrupted balance between the NMDAR and dopamine receptor signaling has been involved in major neuropsychiatric disorders. The most described synaptic autoimmune encephalitis is associated with autoantibodies directed against extracellular domains of the NMDAR, leading patients to develop severe psychotic symptoms. Although NMDAR are the primary target of these antibodies, the cellular and molecular pathway(s) that conduct to their dysfunction remain to be fully understood. Here, we investigated the impact of such autoantibodies on the surface trafficking of NMDAR and dopamine receptors using a combination of high resolution nanoparticule imaging and immunocytochemistry. We report that anti-NMDAR antibodies from patients with encephalitis strongly disturb the surface content and trafficking of NMDAR in cultured hippocampal neurons. Surprisingly, anti-NMDAR antibodies also perturb the surface dynamics of the dopaminergic D1 receptor, which is known to directly interact with the GluN1 subunit of the NMDAR. This suggests that an altered trafficking of the primary target of autoantibodies, i.e. against NMDAR, potentially triggers a "domino effect" on associated membrane receptors. Together, these data show that an altered surface

trafficking of NMDAR, induced by autoantibodies from patients with neuropsychiatric symptoms, acutely modifies the surface organization of NMDAR, as well as partner receptors such as dopamine D1 receptors.

P3.049

Optogenetic coactivation of Purkinje cells controls climbing fiber discharge in the cerebellar cortex

Chaumont, J. (Strasbourg)[1], Guyon, N. (Paris)[2], Valera, A. (Strasbourg)[1], Marcaggi, P. (Paris)[2], Gautheron, V. (Paris)[3], Reibel-Foisset, S. (Strasbourg)[1], Popa, D. (Paris)[2], Dieudonné, S. (Paris)[2], Dugué, G. (Paris)[2], Stephan, A. (Illkirch)[4], Barrot, M. (Strasbourg)[1], Cassel, J.-C. (Strasbourg)[5], Dupont, J.-L. (Strasbourg)[1], Doussau, F. (Strasbourg)[1], Poulain, B. (Strasbourg)[1], Selimi, F. (Paris)[3], Léna, C. (Paris)[2], Isope, P. (Strasbourg)[1]
[1]Institut des Neurosciences Cellulaires et Intégratives, CNRS UPR 3212, Dépt neurotransmission et sécrétion neuroendocrine, Strasbourg, France, [2]Institut de Biologie de l'Ecole Normale Supérieure, CNRS UMR 8197, Paris, France, [3]Center for Interdisciplinary research in biology - College de France, UMR 7241 - U 1050, Paris, France, [4]Institut de Génétique Moléculaire et Cellulaire, CNRS UMR 7104 INSERM U 964, Université de Strasbourg, Illkirch, France, [5]Laboratoire d'imagerie et de Neurosciences Cognitives, CNRS UMR 7237, Université de Strasbourg, Strasbourg, France

The cerebellum plays a central role in sensori-motor integration in vertebrates. We set out to develop a strain of mice in which optogenetic methods would allow a selective stimulation of Purkinje cells, the sole output of the cerebellar cortex. We then developed a new strain of transgenic mice carrying a channelrhodopsin2-YFP fusion protein under the L7-pcp2 promoter which is exclusively expressed in Purkinje cells. A BAC containing the L7/pcp2 gene (186D18) was modified to drive the expression of a cDNA coding for the fusion channelrhodopsin2-eYFP. After microinjection of the construct in male pronucleus, a high level of ChR2-YFP in almost 100% of Purkinje cells was observed in one founder line. *In vitro*, LED photostimulation at 460 nm elicited currents of several nanoamps in Purkinje cell in whole cell mode, but no currents were induced in other cell types of the cerebellar cortex. If short flashes (2 ms) reliably elicit action potentials both *in vitro* and *in vivo*, longer illumination (100-500 ms) can drive Purkinje cells up to 200 Hz. Using *in vivo* recordings, we set out to study the effect of Purkinje cell photostimulation on their identified targets. This optical genetically-targeted stimulation technique will allow us to modulate the pattern of activity of Purkinje cells and evaluate the effect of these precise spatio-temporal alterations on cerebellar microcircuit. Purkinje cells target deep cerebellar neurons, the output of the cerebellum, including an inhibitory GABAergic feedback to olivary cells. This nucleo-olivary connection is in register with the modular organization of the olivo-cerebellar network, suggesting that Purkinje cells may phasically control their own olivary afferents. Tetrode recordings in the deep cerebellar nuclei demonstrated that focal stimulations of Purkinje cells strongly inhibit spatially-restricted sets of cerebellar nuclear neurons. Such stimulations also trigger precisely-timed climbing fiber input signals in the stimulated Purkinje cells. Therefore, our results demonstrate that Purkinje cells phasically control the discharge of their own olivary afferents and thus might participate in the regulation of cerebellar motor learning.

P3.050

The distinct signaling pathways involved in the urotensin II-induced migration and adhesion of glioma cells

Lecointre, C. (Mont Saint Aignan)[1], Joubert, J.-E. (Mont Saint Aignan)[1], Le Joncour, V. (Mont Saint Aignan)[1], Desrues, L. (Mont Saint Aignan)[1], Brulé, C. (Montpellier)[2], Perzo, N. (Sherbrooke)[3], Schouft, M.-T. (Mont Saint Aignan)[1], Jarry, M. (Mont Saint Aignan)[1], Proust, F. (Mont Saint Aignan)[1,4], Leduc,

R. (Sherbrooke)[3], Prézeau, L. (Montpellier)[2], Gandolfo, P. (Mont Saint Aignan)[1], Castel, H. (Mont Saint Aignan)[1]
[1]Laboratory DC2N, Inserm U 982, IRIB, University of Rouen, Mont Saint Aignan, France, [2]Institut de Génomique Fonctionnelle, University of Montpellier 1 and 2, Montpellier, France, [3]Department of Pharmacology, Faculty of Medicine and Health Sciences and Sherbrooke Pharmacology Institute, University of Sherbrooke, Sherbrooke, Canada, [4]Neurosurgery, Neuroradiology and Anesthesiology and Critical Care Departments, Rouen University Hospital, Rouen, France

High grade glioma represent the most frequent primitive cerebral tumor in adult. They are characterized by aggressive invasiveness and neoangiogenesis, two phenomena responsible of the high level of disease recur. Urotensin II (UII) is the most potent vasoactive neuropeptide and regulates astrocyte activities[1-3], promotes neoangiogenesis from brain microvessels and/or acts as a chemoattractant, stimulating migration of endothelial progenitor cells. The aim of the present study was to identify the UII-associated signaling pathways involved in glioma development.
Through the development of a series of signaling assays based on BRET and TR-FRET, we assayed for the ability of UII to couple different G protein pathways (G_s, G_q, $G_{i/o}$, $G_{12/13}$) in HEK cells expressing human UT. In particular, UII dose-dependently activated G_q, $G_{i/o}$ and G_{13}, and regulated cAMP production and PIPs metabolism. We then demonstrated that glioma cell lines U87 and SW1088 and fresh glioma explants expressed UT (and UII), but in these tumoral cells, UII failed to mobilize calcium and to modify cell cycle and proliferation. Gradients of UII, exhibited chemoattracting behavior and dose-dependent stimulation of glioma and HEK-UT cell migration. The UII-mediating chemotaxic effect was independent on $G_{i/o}$, but significantly reduced by a PI3K inhibitor and totally abolished by ROCK inhibition. In contrast, homogenous higher concentrations of UII drastically blocked cell motility of glioma cells and HEK-UT, stimulated glioma cell-matrix adhesion (cell adhesion assay) and cell-cell adhesion (cloning ring assay), through $G_{i/o}$. These mechanisms involved stress fiber formation and vinculin accumulation from 10 min to 1h after treatments. In glioma cells it was shown that UII promotes Rho activation and fails to stimulate phosphorylation of ERK1/2 or Akt. Together, these observations suggest that UII regulates high grade glioma, likely promoting *i)* a gradient-induced directional cell migration through a G_{13}/Rho/Rock and PI3K pathways and *ii)* a homogenous high UII concentration-evoked cell-matrix adhesion through a $G_{i/o}$ protein.
*[1]Castel H et al. (2006) J Neurochem **99,** 582-595.*
*[2]Jarry M et al. (2010) Biochem. J. **428(1)**; 113-24.*
*[3]Desrues L et al. (2012) PLoS One. **7(5)**; e36319.*

P3.051

Characterization of neurodegeneration in a novel apoptosis inducing factor (AIF)-mutant mice

Dias Florencio Leite, G. (Paris)[1,2], Baritaud, M. (Paris)[1], Delavallée, L. (Paris)[1], Cabon, L. (Paris)[1], Brunelle-Navas, M.-N. (Paris)[1], Susin, S.A. (Paris)[1], Krantic, S. (Paris)[1]
[1]Centre de Recherche des Cordeliers, INSERM U 872 - Equipe 17, Physiopathologie des maladies oculaires, Paris, France, [2]Université Paris Descartes, Paris, France

X-gene linked protein Apoptosis-inducing factor (AIF) plays a vital, physiological role in mitochondrial respiration. In pathological conditions, including neurodegeneration in Alzheimer's and Parkinson's diseases, AIF is translocated to nucleus where it triggers caspase-independent apoptosis via direct interaction with DNA. Notably, AIF deficiency or mutations have been involved in mitochondrial impairment. The relative contribution of AIF-related mitochondrial impairment and nuclear DNA degradation to apoptosis is still not resolved because AIF mediates both functions. We have recently generated a novel AIF mutant mice where AIF / DNA interaction and subsequent apoptosis are abolished. This model therefore provides a unique possibility to address selectively the role of mitochondrial impairment in cell death.
Though hemizygous males and homozygous females die in utero, the heterozygous females are viable. These females present with marked, but varying phenotypes, ranging from slight growth retardation to severe, age-related neurodegeneration, associated with hydrocephaly and lack of one eye. Western blot data indicate that, when compared to age-matched controls, heterozygous females

display reduced AIF expression in olfactory bulb and cerebellum by the age of 2- and 5- months, respectively. By contrast, AIF expression is increased in the cerebral cortex of mutants at 2 months. These differences in AIF content may reflect the increased susceptibility of cerebellum and olfactory bulb to AIF-related oxidative stress, and explain why the cortex is more resistant and affected only in older animals. Immunohistochemical labeling of microglia and astrocytes with CD11b/Iba1 and GFAP, respectively point to morphological signs of glial activation. These signs suggests a neuroinflammation, at least in the animals with severe neurodegeneration (i.e. in hydrocephalus). The characterization of underlying molecular mechanisms by which AIF is involved in neuronal death via age-related mitochondrial impairment is crucial for development of more efficient therapies for neurodegenerative diseases.

P3.052

Cell surface assembly of class 3 semaphorin receptors

Jourdan, C. (Montpellier)[1], Kniazeff, J. (Montpellier)[1], Ango, F. (Montpellier)[1]
[1]Institut de Génomique Fonction - Univ. de Montpellier 1 & 2, Cnrs Umr 5203 - Inserm U 661, dépt. de Neurobiologie, Montpellier, France

Secreted class 3 semaphorins (sema3) modulate a wide variety of neuritis behavior during central nervous system formation, including attraction, repulsion and branching. Sema3 family function is triggered through binding to specific receptor complexes that include the obligate co-receptors neuropilins (1 and 2) and a specific plexin-A (1 to 4). Interestingly sema3 binding to neuropilins dimers was shown to lack transduction signaling and specific recruitment of plexin-A is necessary to ensure proper function of the receptor complexes. Yet, the molecular and cellular mechanisms of class 3 semaphorin receptors assemblies are not known. For instance, do heteromeric form of sema receptors are assembled and ready to respond to semaphorins? Or, is it the ligand that defines the receptor assembly? To answer these questions we used time-resolved FRET to analyse cell-surface protein-protein interaction in live cells. First, we confirmed that neuropilin 1 and 2 form homo and hetero dimers at the cell surface without ligands. We found that plexin-A1,2 and 4 formed constitutive heterodimers with neuropilin 1, but not plexin-A3. In addition we found that mutant plexin-A1-4 with constitutive activity formed heterodimers with neuropilin1, suggesting that plexinA3 and neuropilin1 association might be regulated by sema3A, or at least that sema3A induce a conformational change leading to the detection of a fret signal between plexinA3 and neuropilin1. We are now testing the role of sema3A in this association as well as the specific role of plexins in the sema3A signaling pathway. Understanding the basic principles of sema class 3 receptors assembly will be pivotal to understand how ligand binding translates into specific pathways of cellular signaling.

P3.053

VGLUT3 is differentially expressed in various strains of laboratory mice: impact on anxiety and drug behavioral responses

Ramet, L. (Paris)[1], Sakae, D.Y. (Paris)[1], Henrion, A. (Créteil)[2], Poirel, O. (Paris)[1], Jamain, S. (Créteil)[2], El Mestikawy, S. (Paris)[1,3], Daumas, S. (Paris)[1]
[1]INSERM U 952, CNRS UMR 7224 Université Pierre et Marie Curie, Pathophysiologie des maladies du SNC, Paris, France, [2]Inserm U 955, Psychiatrie Génétique, Créteil, France, [3]McGill University, Montreal Neurological Institute, Département de Psychiatrie, Montréal, Canada

Glutamate is stored in vesicles by vesicular glutamate transporters named VGLUT1-3. VGLUT1 and -2 are involved in canonical glutamatergic transmission. In contrast, VGLUT3 is present in neurons using other classical transmitters: i) subpopulations of GABAergic interneurons in the cortex and the

hippocampus, ii) serotoninergic neurons in the raphe and iii) cholinergic interneuron in the striatum. We have previously shown that C56BL/6 mice that no longer express VGLUT3 (VGLUT3-KO) display anxiety-associated phenotype and modified striatal-related behaviours (such as higher spontaneous and cocaine-induced locomotor activity or haloperidol-induced catalepsy). Moreover, several studies have shown phenotypic differences in inbred mouse strains for both anxiety and behavioural responses elicited by drugs. Given the involvement of VGLUT3 in these phenotypes, the purpose of this study was to investigate strain differences in VGLUT3 expression levels.
Five inbred mouse lines were chosen according to their contrasted anxiety and drugs sensitivity: C57BL/6, C3HeN, DBA/2J, 129/Sv, and Balb/c. We analyzed the level of VGLUT3 expression in different brain areas involved in reward or mood regulation such as dorsal or ventral striatum, hippocampus, prelimbic cortex.
Our preliminary results show that these 5 inbred mouse strains express very different levels of VGLUT3. This difference of expression does not seem to be linked to mutations of the *vglut3* gene promotor. Furthermore, mice behaviour in the open-field, elevated plus-maze, spontaneous- and cocaine-induced locomotor activity is under investigation and will be correlated to VGLUT3 levels in different brain areas.
By using these mice lines expressing very different level of VGLUT3 protein, we hope to better understand the contribution of the *vglut3* gene in specific behaviours such as anxiety and sensitivity to substance of abuse.

P3.054

The role of Pyk2 and Nir1 in retinal synaptic neurotransmission

Cammas, L. (San Francisco)[1], Bouquet, C. (San Francisco)[1], Schlessinger, J. (New Haven)[2], Beggs, H. (San Francisco)[1], Copenhagen, D. (San Francisco)[1]
[1]University of California, Department of Ophthalmology, San Francisco, United States, [2]Yale University School of Medicine, Department of Pharmacology, New Haven, United States

Purpose: The retina is composed of a hierarchical circuit of neurons where photoreceptors phototransduce the light stimulus into an electrical signal that modulates glutamate signaling at the first retinal synapse. The cone photoreceptors, that mediate daylight and color vision, establish synaptic connections with cone bipolar cells. Rods connect to rod bipolar cells. Dysfunction of the cones or their post-synaptic pathways results in incurable blinding diseases such as age-related macular dystrophies and cone dystrophies (CORD). Only one gene, Nir1, has been found mutated in CORD5, its function however remains unknown. It binds the protein Pyk2 and the mutation that causes CORD5 is located in the Pyk2-binding domain of the Nir1 gene. Pyk2 is a non-receptor tyrosine kinase that participates in the assembly of signaling complexes and is involved in synaptic biology. To understand the role of Nir1 and Pyk2 in cone signaling we are (i) generating a mutant mouse model for CORD5 where the Nir1 gene is deleted and (ii) analyzing the retinal function of a Pyk2 deleted mouse.
Methods: The mutant mouse for Nir1 is generated by inserting a polyvalent transgene in the Nir1 gene. Visual functions of mice are explored using electroretinography (ERG) methods that allow for specific detection of rod and cone activity as well as postsynaptic neuron activity. Retinal samples are analyzed using microscopy, histological and biochemical methods.
Results: We show that Pyk2 mediates cone signaling at the first retinal synapse. Pyk2 deleted mice display a specific ERG defect indicating that Pyk2 is involved in cone-mediated vision only, not in rod-mediated vision. Moreover, cone phototransduction is normal in $Pyk2^{-/-}$ mice, only the cone to bipolar synaptic neurotransmission is defective.
Conclusion: Pyk2's role in retinal synaptic biology is unique as there are no other mouse models that display a specific cone bipolar defect without affecting the rod bipolar cell. Moreover, previous studies of cone dystrophy hypothesized that defective cone phototransduction and/or cone degeneration was the primary disease causing defect. We suggest that synaptic defects could also cause symptoms of cone dystrophy.

P3.055

Regulation of autophagy by urotensin II: potential involvement in glial tumorigenesis

Castel, H. (Mont-Saint-Aignan)[1], Coly, P.-M. (Mont-Saint-Aignan)[2], Schouft, M.-T. (Mont-Saint-Aignan)[3], Lejoncour, V. (Mont-Saint-Aignan)[2], Céline, L. (Mont-Saint-Aignan)[4], Wurtz, O. (Mont-Saint-Aignan)[2], Gandolfo, P. (Mont-Saint-Aignan)[4], Morin, F. (Mont-Saint-Aignan)[4]
[1]Univ. de Rouen, INSERM U 982 - DC2N, Astrocytes et niche vasculaire, Mont-Saint-Aignan, France, [2]Inserm 982, Lab DC2N, Université de Rouen, Mont-Saint-Aignan, France, [3]Inserm 982, Lab DCN, Université de Rouen, Mont-Saint-Aignan, France, [4]Inserm 982, Mont-Saint-Aignan, France

Growth factors, but also vasoactive peptides, are endogenous compounds that have been shown to play an important role during tumorigenesis. We have previously demonstrated that the vasoactive peptide urotensin II (UII) and its receptor UT are expressed in several glioma cell lines. Moreover, we have found that UII stimulates tumor growth of heterotopycal xenografts of U87 glioma cells in nude mice. Autophagy is a catabolic process that involves the entrapment of cytoplasmic components within characteristic vesicles for their delivery to and degradation within lysosomes. This process is induced by cellular stress, such as nutrient deprivation or hypoxia. When overactivated, however, autophagy can lead to cell death. At times, autophagic cell death is used as an alternative to apoptosis to eliminate unwanted, damaged, or transformed cells. Consistent with this, tumorigenesis is associated with a downregulation in autophagy, and many genes that mediate the execution of autophagy have been shown to be tumor suppressors. Together, these data prompted us to investigate if the oncogenic activity of UII could involve regulation of the autophagic process.
Using the HEK-293 cell line, we found that UII reduced the autophagic activity elicited by hypoxia. We found that the reduction in the number of autophagosomes was due to inhibition of their biogenesis rather than modulation of their lysosomal clearance. Moreover, inhibition of autophagy by UII correlated with reduced levels of mRNAs encoding the pro-autophagic proteins Bnip3 and Redd1, which were previously described as tumor suppressors in xenograft mouse models. This effect of UII might involve post-transcriptional mechanisms, since UII strongly induced the expression of the microRNA-221, an “oncomiR” highly expressed in several human cancers, which has been found to downregulate Redd1 mRNA by targeting its 3'untranslated region. Collectively, these data lead us to propose that UII, by activating a signaling pathway involving the oncomiR-221 and its downstream targets, may accelerate tumor growth by reducing the autophagic capacity of cancer cells exposed to their hypoxic microenvironment.

P3.056

Activation of microglia impacts their directional motility

Avignone, E. (Bordeaux)[1,2], Lepleux, M. (Bordeaux)[1,2], Angibaud, J. (Bordeaux)[1,2], Nägerl, U.V. (Bordeaux)[1,2]
[1]Institut Interdisciplinaire de Neurosciences, UMR 5297 CNRS, Bordeaux, France, [2]Institut Interdisciplinaire de Neurosciences, Université Bordeaux 2, Bordeaux, France

Microglia cells are the resident macrophages of the central nervous system and are considered its first line of defense. Their morphology is characterized by numerous fine processes, which are strikingly dynamic. While these processes appear to randomly scan the parenchyma under physiological conditions (basal motility), they can rapidly move towards sites of acute injury or danger signals, like excessive levels of ATP (directional motility).
In addition to this more or less instantaneous response, microglia undergo manifold molecular and structural changes in pathological conditions, called activation, which unfold on a much longer time scale (several days). However, the functional impact of activation is poorly understood, in particular it

is unknown how activation might affect microglial ability to scan the brain or to respond to noxious stimuli.
To address this question we used the model of kainate (KA) induced status epilepticus, where activation of microglia has been described 48 hours after intraperitoneal injection of KA.
To characterize the morphological dynamics of microglia, we used time-lapse two-photon imaging in acute hippocampal slices from transgenically labeled mice ($CX3XR1^{+/EGFP}$). We examined baseline motility and two types of directional motility: in response to acute laser lesions, and to local application of an ATP analogue (P2Y12 receptor agonist) via a patch pipette.
Our experiments reveal strikingly different modes of directional motility, depending on the type of stimulus (laser or P2Y12 receptor agonist). In particular, differences regarding response velocity, affected area and processes synchronization were observed. Importantly, microglial activation accelerated the directional motility towards the pipette, but not to the laser lesion site, and did not affect basal motility.
In summary, our experiments indicate that microglial activation induced by status epilepticus differentially impacts the dynamic behavior of microglia in response to specific acute noxious stimuli, which may be an important feature of altered microglial behavior during pathophysiological conditions.

P3.057

Role of the extracellular-signal regulated kinase (ERK) pathway in the regulation of arc protein: implications for striatal structural plasticity and behavioral responses to cocaine

Salery, M. (Paris)[1]
[1]*Université Pierre et Marie Curie, Paris, France*

The characterization of the molecular and cellular basis of striatal plasticity is a major step in the understanding of mechanisms underlying long term adaptations mediated by addictive drugs. In the striatum, cocaine-induced ERK activation triggers several mechanisms important for neuronal plasticity induced by drugs of abuse. Our team has been instrumental to the identification of the transcriptional regulations mediated by nuclear substrates of ERK. In the present study we focused on the characterization of cytoplasmic ERK partners that could mediate local adaptations induced by cocaine
The somatodendritic-localized protein Arc is known to play a crucial role in synaptic plasticity, notably through a modulation of both AMPA receptors trafficking and actin cytoskeleton dynamics. Our ongoing work shows that Arc mRNA levels are increased in the striatum 30 minutes after a single injection of cocaine (20 mg/kg) and this is associated with an increase in Arc protein expression occurring 60 minutes after cocaine treatment. Our pharmacological studies reveal that this cocaine mediated increase in Arc expression is dependent on the activation of both dopamine D1 and glutamate NMDA receptors in an ERK dependent fashion. In order to assess whether ERK could regulate Arc expression in the cytoplasm at the translation level, we first analyzed the role of a cytoplasmic ERK target, the MAP-kinase interacting kinase 1 (Mnk-1), which is involved in the translation machinery. We observed a rapid increase (10 minutes) in the active (i.e. phosphorylated) form of Mnk-1 after cocaine treatment (20 mg/kg). Importantly, we show that cocaine-induced Mnk-1 activation dependent on ERK activity. Ongoing experiments include the analysis of Arc expression under pharmacological blockade of Mnk-1.
Data obtained so far show an induction of Arc expression, along with the activation of Mnk-1 in response to acute cocaine. This suggests that cytoplasmic regulation of Arc expression could trigger synaptic changes underlying cocaine-induced plasticity and behavior. The discovery of novel ERK-dependent dendritic molecular events important for structural and behavioral responses to cocaine will potentially help identifying novel potential therapeutic targets in the field of addiction.

P3.058

Gating of excitation in the dorsal raphe nucleus by GABA

Soiza-Reilly, M. (Boston)[1,2,3], Anderson, W.B. (Sydney)[4], Vaughan, C.W. (Sydney)[4], Commons, K.G. (Boston)[1,2]
[1]Boston Children's Hospital, Anesthesia, Boston, United States, [2]Harvard Medical School, Anesthesiology, Boston, United States, [3]Inserm U 839, Paris, France, [4]University of Sydney, Pain Management Research Institute, Sydney, Australia

Serotonin neurons located in the dorsal raphe nucleus (DR) regulate forebrain's activity involved in stress and emotional states. Additionally, GABA and glutamate neurotransmission modulate the excitability of DR neurons, ultimately influencing behavioral states including sleep/wake cycles, anxiety and aggression. Several evidences suggest a complex interaction between GABA and glutamate transmission within the DR, however how these circuits are organized as well as the possible neuroanatomical basis for this interaction remain vastly unknown. In this study, we examined the fine synaptic architecture of GABA and glutamate axons in the DR using a quantitative high-resolution immunofluorescence technique called array tomography, as well as immunoelectron microscopy. This analysis revealed a novel microcircuit feature within the DR involving GABA and glutamate axons organized in synaptic triads with a common postsynaptic target. Consistent with this, electrophysiological recordings showed that GABA gates glutamate release in the DR through a dual presynaptic mechanism involving both enhancement and inhibition of glutamate release mediated by GABA-A and GABA-B receptors respectively. This study uncovers a unique anatomical feature within DR's neuropil, providing new insight into how excitatory glutamate and inhibitory GABA transmission locally interact to modulate the excitability of DR neurons

P3.059

Pannexin1 are expressed in Zebrin II positive bands in the adult mouse cerebellum

Levenes, C. (Paris)[1], Ady, V. (Paris)[1], Bernard, V. (Paris)[2], Dubayle, D. (Paris)[1]
[1]CNRS - Université Paris Descartes UMR 8119, Paris, France, [2]Laboratoire de Physiopathologie des Maladies du SNC - INSERM U 952, CNRS UMR 7224, Université Pierre et Marie Curie, Paris, France

Pannexins, a class of membrane channels, are structural homologs of the invertebrate protein innexins and of the gap junction forming proteins connexins. However, they do not seem to form gap junction in neurons but rather to function as large pore ion channels at the surface of membranes. Their physiological role in the central nervous system is still unknown but they have been implicated in several pathological conditions such as ischemic, excitotoxic and ATP-dependent cell death, inflammasome activation. They are expressed in the cerebellum, particularly in Purkinje cells. Here, using immunochemistry staining, we show that Pannexin1 are not homogeneously distributed in the overall cerebellar Purkinje cells population. Rather, they are distributed along sagittal bands that match that of Zebrin II expression. As for Zebrin II proteins, sagittal section reveals their increasing expression from rostral to caudal lobules. Expression is rare in lobules I-II while lobules IX-X are strongly labelled all over the cerebellum. Electron microscopy of the cerebellar cortex reveals that Pannexin1 locate at cell membranes including spines and dendrites of Purkinje cells. In some cases, Pannexin1 was located at postsynaptic sites facing climbing fiber terminals. This heterogeneous distribution of Pannexin1 among Purkinje cells makes them new members of the large family of proteins distributed along Zebra stripes. This super familly includes ion channels, receptors, transporters and enzymes, that are all possible key members of the cerebellar activity. Pannexin1 are good candidates to participate to or underlie synchronized activity in the Zebra stripes of the cerebellar cortex.

P3.060

Increased locomotor activity leads to a calcineurin-dependent Pyk2 accumulation in the nucleus of dorsolateral striatum neurons

Coura, R. (Paris)[1], Faure, C. (Paris)[1], Marion-Poll, L. (Paris)[1], Montalban, E. (Paris)[1], Longueville, S. (Paris)[1], Terao, Y. (Paris)[1], Hervé, D. (Paris)[1], Girault, J.-A. (Paris)[1]
[1]*Inserm - U 839, Institut du Fer à Moulin (IFM), Université Pierre et Marie Curie, Paris, France*

Proline-rich tyrosine kinase 2 (Pyk2) is a non-receptor calcium-dependent protein-tyrosine kinase of the focal adhesion kinase family that is highly expressed in the forebrain. Pyk2 is activated in response to increases in intra-cellular calcium levels. Upon stimulation, Pyk2 undergoes rapid autophosphorylation at Tyr402 and can activate the Src family kinase and MAPK signaling pathways. Pyk2 has been involved in LTP and Src-dependent NMDAR potentiation. It has also been reported to bind PSD-95 in the postsynaptic density and to be critical for the induction of LTD. Furthermore, we have shown that Pyk2 undergoes a cytonuclear shuttling and accumulates in the nucleus under certain conditions, but the importance of this nuclear translocation in vivo is not known. Here, we focused on Pyk2 nuclear accumulation in the striatum. We found that 15 min after a cocaine injection (20 mg/kg, i.p.) Pyk2 immunoreactivity was increased in the nucleus of both striato-pallidal and striato-nigral medium-size spiny neurons of the dorsolateral striatum (DLS), but not in the other regions of the striatum. This accumulation was prevented in mice pretreated with cyclosoprin A, revealing the role of calcineurin in Pyk2 nuclear translocation, as previously observed in PC12 cells. In order to evaluate the implication of the glutamate pathway, we used either the NMDA antagonist MK-801 or the mGluR2 agonist LY379268. Cocaine-induced Pyk2 accumulation was prevented by LY379268, which is known to decrease glutamate release but was not altered by MK-801. Interestingly MK-801 alone or the muscarinic antagonist scopolamine which both increased locomotion induced the accumulation of Pyk2 in neuronal nuclei specifically in the DLS. We then examined whether increased locomotion was sufficient to induce this response. Forced exercise, in which mice were placed in a wheel and forced to move at the same speed as in response to cocaine injection, induced Pyk2 accumulation in the nucleus specifically in the DLS and this response was prevented by cyclosporin A. In conclusion, Pyk2 accumulates in the nucleus of neurons, specifically in the DLS, under various conditions of increased locomotor activity. This nuclear translocation is likely to be mediated by calcineurin activation.

P3.061

Changes in GABAergic transmission in the CA1 region of the mouse hippocampus after kainate-induced status epilepticus

Bidoret, C. (Paris)[1], Audinat, E. (Paris)[1]
[1]*Université Paris Descartes, U 603 Neurophysiologie et Nouvelles Microscopies, Interactions Neurone-Glie, Paris, France*

We previously showed that Status Epilepticus (SE) induced by systemic kainate injection in the Mouse induced a marked inflammation in the hippocampus (Avignone et al., 2008). This inflammatory reaction is characterized by a rapid upregulation of pro-inflammatory markers, an activation of microglia and astrocytes and a recruitment of leucocytes within the hippocampus (Levavasseur et al., in preparation). Here we investigate whether this inflammatory reaction was accompanied by changes in the inhibitory synaptic activity of the hippocampal network.
First, spontaneous and miniature inhibitory events recorded in CA1 pyramidal cells were increased from 2 days until 30 days after SE compared to vehicle injected animals. These new events were large and fast, suggesting that they arise at proximal synapses. Increased somatic inhibition of CA1 pyramidal cells has been previously observed in models of temporal lobe epilepsy after the latent period. Our data now indicate that these changes occur immediately after SE.
In addition, IPSCs evoked by electrical stimulation in the stratum pyramidale of CA1 showed also faster kinetics. However, this change was accompanied by an increase of the paired pulse ratio, suggesting a presynaptic modification of inhibitory transmission after SE. Furthermore, as already

described in the literature, we observed a loss of parvalbumine-positive basket cells after SE. Thus, we propose a compensatory function of surviving interneurons.
Because lymphocytes invade the hippocampal parenchyma after SE, we next investigate whether they contribute to the synaptic changes observed. For that purpose, we used the CD3epsilon KO mice in which the maturation of CD4+, CD8+ and TNK T lymphocytes is impaired. In these mice, the upregulation of inhibitory transmission was reduced. Therefore, our data suggest that inflammation, in particular the recruitment of T lymphocytes, contributes to a protective synaptic remodeling against hyper-excitability and the development of recurrent epilepsy.

P3.062
Implication of ATXN7 SUMOylation in degradation of nuclear aggregates

Marinello, M. (Paris)[1], Alves, S. (Paris)[1], Brice, A. (Paris)[1], Sittler, A. (Paris)[1]
[1]*Hôpital de la Pitié-Salpétrière, INSERM UMRS 975-CNRS UMR 7225-CRICM/NEB, Institut du Cerveau et de la Moelle Épinière, Paris, France*

Post-translational protein modifications are critical for the spatial and temporal regulation of signaling cascades. Recently, it was shown that protein modification by members of the SUMO (small ubiquitin-like modifier) protein family plays key roles in neuronal functions. Therefore, our team demonstrated that SUMOylation plays a role in Spinocerebellar ataxia type 7 (SCA7). SCA7 is a neurodegenerative disorder, whose pathology is caused by an expansion of a polyglutamine stretch in the protein ataxin-7 (ATXN7). Numerous ATXN7 positive nuclear inclusions, colocalizing with SUMO1 and SUMO2, were observed in cortex and cerebellar cells of SCA7 patients and mice. In a SCA7 cellular model we found that preventing SUMOylation of expanded ATXN7 leads to higher amounts of SDS-insoluble aggregates. These results demonstrate that SUMOylation influences aggregation of polyQ expanded ATXN7, which is likely a key event during progression of pathogenesis. In order to identify potential ATXN7 SUMOylation actors, we analyzed the expression and subcellular distribution of proteins that may be implicated in this pathway. We focused on the enzyme RanBP2, which we recently identified *in vitro* as SUMO E3 ligase for ATXN7. Our objective is to investigate if RanBP2, localized at the cytoplasmic filaments of the nuclear pore, couples the SUMO modification of ATXN7, with its entry into the nucleus. The first step was to determine the physical interaction and co-localisation between ATXN7 and RanBP2. Secondly, in order to confirm that RanBP2 was an actor in the SUMOylation of ATXN7, we used a siRNA approach to decrease its expression and evaluated the silencing consequences. Preliminary results show a deficit in the degradation of nuclear insoluble aggregates, pointing to the fact that RanBP2 could influence ATXN7 SUMOylation. The structural data for the SUMO E3 enzyme RanBP2 in complex with the other components of the SUMO pathway is now available, making possible the structure-based design of specific inhibitors or activators. For this reason, our aim is to elucidate the mechanisms regulating ATXN7 degradation via SUMOylation to identify new targets for therapeutic intervention.

P3.063
Identification of a dopaminergic circuit controlling startle-induced locomotion in the *Drosophila* brain

Riemensperger, T. (Paris)[1,2], Issa, A.-R. (Paris)[1], Pech, U. (Göttingen)[2], Coulom, H. (Paris)[1], Nguyễn, M.V. (Paris)[1], Cassar, M. (Paris)[1], Jacquet, M. (Paris)[1], Fiala, A. (Göttingen)[2], Birman, S. (Paris)[1]
[1]*Genetics and Physiopathology of Neurotransmission, ESPCI ParisTech, Neurobiology Unit, CNRS UMR 7637, Paris, France,* [2]*Molecular Neurobiology of Behaviour, Johann-Friedrich-Blumenbach-Institute, Georg-August-University of Goettingen, Schwann-Schleiden Research Center, Göttingen, Germany*

Like in humans, *Drosophila* locomotor ability progressively decreases with age and in certain pathological conditions. This decline can be monitored by a negative geotaxis assay that measures the climbing behavior of a fly in response to a gentle mechanical shock. Thus, this assay was previously used to demonstrate progressive locomotor deficits in adult flies when the human Parkinson's disease-associated protein α-synuclein, or its pathogenic mutant forms, were transgenically expressed in all *Drosophila* neurons (Feany and Bender, 2000). Here we have used this disease model to search for the brain neuronal pathways that control startle-induced locomotion in flies. We compared the effects of pathogenic mutant α-synuclein produced in *Drosophila* with various GAL4 drivers that express in large or small subsets of dopaminergic neurons. This led us to identify a group of 15 dopaminergic neurons in the anterior medial region of the brain whose dysfunction appears necessary and sufficient to induce progressive deficits in the negative geotaxis assay. These neurons innervate the tips of the mushroom body horizontal lobes and their synaptic projections shrink in the presence of α-synuclein. This suggests that activity of these dopaminergic neurons is required for an efficient control of startle-induced locomotion in flies. Thus, in *Drosophila* as in humans, motor deficits in Parkinson's disease conditions correlate to degeneration of a specific subset of brain dopaminergic neurons highly sensitive to α-synuclein toxicity.

P3.064

In vivo study of potential neurologic effects of nanomaterials after direct intracerebral exposure in mice

Bencsik, A. (Lyon)[1], Balvay, A. (Lyon)[1], Thieriet, N. (Maisons Alfort)[2], Lakhdar, L. (Lyon)[3]
[1]Anses, Unité MND, Lyon, France, [2]Anses, DER, Maisons Alfort, France, [3]Anses, PFEA, Lyon, France

The expanding development and production of engineered nanomaterials (ENMs) cover varied and extensive potential benefits in consumer products, food, drugs… The unique properties of ENMs have also raised concerns about the potential non intended consequences on human health and the environment. A potential risk for neurotoxicity arises if exposure leads to systemic absorption and distribution in the nervous system. This could support a possible link with the appearance of neurodegenerative diseases like Parkinson´s or Alzheimer's diseases. However, neurotoxicity remains poorly documented. Thus, to consider whether any brain exposure may trigger a neurotoxic effect we developed a first approach based on direct injection of nanoparticles (NP) into the brain of TgM83 mice, an experimental model of human neurodegenerative disease, using a stereotaxic apparatus. Nano-TiO_2 and -SiO_2 were selected because they are already used in several nanoproducts and we compared the neurologic effects of a single injection of these nano-TiO_2 and -SiO_2 on motor performance using a rotarod equipment - measured on a rotarod at 20 rpm or at an accelerating rod (from 4 to 40 rpm). Before and after injection motor activity is registered individually for each mouse exposed, once a week, for 4 weeks. Injections were performed under deep anesthesia into the ventricular compartment of the brain in order to favor the diffusion of NP into the entire brain. Besides, mice are culled at 1, 2, 3, and 4 weeks after exposure in order to study the time dependant effect on the histopathology of the brain (gliosis, inflammatory process, …).
The first results indicate that both ENMs studied are able to induce neurological effects after a direct intracerebral exposure. The histopathological analyses made it possible to visualize the site of injection, to characterize the cicatrization of the mechanical lesion induced by the needle during injection. Beyond the site of injection, the kinetic studies showed that, whereas gliosis progressively vanished, microglial activation grew throughout the brain suggesting an induction of a long lasting neuroinflammation. These results fitted well with the loss of motor performances.

P3.065

Mesoporous silica chips for effective 3D primary cell culture

Dreyfus, M. (Grenoble)[1], Bouamrani, A. (Grenoble)[1], Mombrun, A. (Grenoble)[1], Berger, F. (Grenoble)[1]
[1]*CEA Léti CLINATEC, Grenoble, France*

Despite decades of research in the field of cell culture, understanding the molecular mechanisms underlying pathologies still remain a challenge. Transposition of *in vitro* studies to *in vivo* animal models often results in a failure. It clearly appears that one of the main encountered problems resides in the inherent cell lines drifting from their original phenotype.
Therefore, it becomes necessary to impose new standards in cell culture studies by focusing on primary cultured cells with a limited number of passages. We developed new methodological tools and devices allowing *in vivo* sampling of biological tissues with a non-invasive approach. The method is based on the use of micro/nano patterned surfaces including mesoporous silica chips. Our approach offers an easy way to establish primary cultures of cells in their native micro environment. The implementation of several kinds of surgical instruments with the developed devices will open an access to a wide range of tissues, including areas where classical biopsies by resection are impossible.
Thus, we showed that beyond cell culturing opportunities, the cellular imprints obtained on the nanostructured surfaces can be analyzed in a multimodal approach, including two-photon microscopy imaging, immunocytochemistry, scanning electron microscopy and mass spectrometry. Altogether, polyomics analysis and early screening after sampling should strengthen the experimental data obtained in fundamental research. From one hand, this takes a growing importance to ensure that biological objects observed *in vitro* reflects the processes occurring in physiological or physiopathological conditions. From the other hand, improving *in vivo* sampling with minimally invasive devices will complement biopsy approaches for diagnosis procedures of human pathologies.
We now focus on the neural stem cell niche in the subventricular zone lining the walls of lateral ventricles. Using mesoporous silica chips, we developed a specific surgical tool allowing an in vivo harvesting of neural stem cells, opening the way for an effective establishment of primary neurospheres cultures.

P3.066

Vitamin D supplementation in a mouse model of Alzheimer's disease

Landel, V. (Marseille)[1,2], Cès, A. (Marseille)[1,2], Virard, I. (Marseille)[1,2], Loriod, B. (Marseille)[3], Féron, F. (Marseille)[1,2]
[1]*Aix Marseille Université, Marseille, France,* [2]*CNRS, UMR 7259, Marseille, France,* [3]*TAGC, INSERM U 1090, Marseille, France*

Similar to almost all tissues, the brain responds to vitamin D by regulating mRNA expression and protein synthesis. This steroid hormone exerts potent immuno-modulatory and neuroprotective effects and its deficiency has been associated to several neurodegenerative diseases, including Alzheimer's disease (AD). In this study we sought to assess the effects of vitamin D dietary supplementation in an AD mouse model (5xFAD) and to explore its mechanisms of action. The potential therapeutic benefit of vitamin D3 (cholecalciferol) was evaluated at the dose of 500 IU/kg/day for 4 months, during two time windows:
1) from weaning to young adulthood and
2) from young to advanced adulthood.
In the former case, vitamin D was delivered as a preventive drug, before the symptomatic phase; in the latter case, vitamin D was considered as a curative agent, delivered during the symptomatic phase. We then,
i) assessed memory deficits using the Y maze;
ii) quantified histological markers of AD, i.e. amyloid peptide accumulation and glial activation;
iii) studied the genes and metabolic pathways affected by vitamin D in the hippocampus and cortex, using pangenomic microarrays. Our first results indicate that vitamin D restores cognitive and memory

impairements when used as a therapeutic agent but not when delivered as a preventive drug. It also reduces amyloid plaque load and neuroinflammation. The transcriptome experiment is still in progress. This study provides credential to the clinical trial currently performed by a French team, based in Angers, which supplements AD patients' diet with vitamin D.

P3.067

Cognitive impairments in Parkinson's disease: assessing the role of apathy and depression

Quilodran, R. (Valparaiso)[1], Viñales, L. (Valparaiso)[2], Castilla, F. (Valparaiso)[3], Santos, I. (Valparaiso)[3], Marchant, I. (Valparaiso)[1], Procyk, E. (Bron)[4]
[1]Escuela de Medicina, Universidad de Valparaíso, Valparaiso, Chile, [2]Facultad de Ciencias, Universidad de Valparaíso, Departamento de Ingeniería Biomédica, Valparaiso, Chile, [3]Hospital Carlos Van Buren, Servicio de Neurología, Valparaiso, Chile, [4]SBRI Inserm U 846, Neurobiology of Executive Functions, Bron, France

Parkinson's disease (PD) has typically been considered to be a motor disorder secondary to dopamine depletion in the basal ganglia. Clinical evidence of cognitive impairments appearing even in the earliest phase has added critical information to characterize this syndrome. Particularly vulnerable to PD are a set of complex cognitive control mechanisms, i.e. executive functions that contribute to the continuous orchestration of behavior. Evidence from several studies suggests that apathy and depression are independent forms of PD expression that may affect cognition differentially. However, the specific contributions of apathy and depression to PD cognitive impairments remain misunderstood. We studied a population-based sample of PD patients to estimate the prevalence and characterize PD-related apathy and depression. In addition, we sought to evaluate whether apathy may reflect a primary behavioral disturbance affecting executive functioning independently from depression in PD.
55 patients (58% male, mean age 68.7 years ± 7.2) were recruited from the Department of Neurology at Carlos van Buren Hospital of Valparaiso and completed a comprehensive clinical and neuropsychological evaluation. Apathy and depression were evaluated using the Starkstein scale and the Beck Depression Inventory-II, respectively. Cognitive domains attention, memory, language, visuospatial abilities, and executive functions were examined using the Addenbrooke's Cognitive Examination test and the Problem Solving Task. As a measure of the relative health impacts of these PD-related disorders, we evaluated quality of life using the PDQ-39 test.
In our study, most participants had a mild/moderate PD with a mean duration of 4.7 ± 3.5 years. Apathy was diagnosed in 24/55 patients (43%). In 39.9% of the studied population, apathy coexisted with depression, whereas only 12.7% were apathetic without depression. Apathy was significantly associated with higher depression scores, lower cognitive functioning, and more severe motor symptoms. In agreement with previous reports, apathy appears to be a common disorder in PD population. Our preliminary results suggest that apathy but not depression is associated with impaired cognition in PD patients.

P3.068

Non human primate model of epilepsy: neural dynamics within the parieto-frontal system

Aboubakr, W. (Marseille)[1], Bartolomei, F. (Marseille)[1], Chauvel, P. (Marseille)[1], Régis, J. (Marseille)[1], Boussaoud, D. (Marseille)[1]
[1]Institut de Neurosciences des Systèmes, INSERM UMR 1106, Marseille, France

Focal epilepsies are characterized by the genesis of seizures in a local neuronal network, which constitutes the epileptic zone (EZ). It is poorly understood how an EZ is formed and how it can remain

focal or expand to a larger network. Our study aims to develop a parietal epilepsy model in the macaque monkey, in order to advance our understanding of epileptogenesis. Our aim is to understand the neuronal dynamics of the epileptic brain at different scales, using micro- and macroscopic neuronal recordings. Epilepsy was induced by an intracortical injection of alumina gel in the parietal cortex (area PEc), known to cause in macaques epileptic seizures close to those observed in human epilepsy. Electrodes with 10 contacts each were implanted in two parietal areas (MIP, PEip), the supplementary motor area (SMA) and the dorsal premotor cortex (PMd). Five of the 10 contacts were designed to record global activity (SEEG), and 5 for single unit activity.
This report is based on analysis of SEEG data. Results of daily recording sessions in a macaque monkey at rest show that after a latency of about 5 weeks after the injection, interictal spikes appear in the parietal cortex close to the injection site and in the nearby area MIP. Few days later, interictal spikes extend to the premotor regions, and the first seizure occurred at the 41th day. Analysis of signal interdependencies (non linear correlation) both locally within each cortical region, and between distant parietal and premotor electrodes, shows that the development of epilpesy is paralleled by two phenomena:
1) a pronounced increase in parieto-premotor synchrony,
2) the shift from a bi-directional relationship to a predominant uni-directionality with the parietal cortex driving the premotor cortex.
In parallel, the monkey displayed hypotonia and motor deficits of the contralateral hemibody after the first seizure, during 2 weeks.
These preliminary results confirm that the development of epilepsy is coincident with the propagation of neuronal changes from a local network to distant connected brain regions. Current work is seeking to replicate these findings in additional monkeys.
This work was performed in collaboration with Alcis SARL (Besançon) which provided the electrodes and a CIFRE fellowship to W.A.

P3.069

From DBS to deep brain bio-harvesting: a strategy for brain polyomic investigations

Bouamrani, A. (Grenoble)[1], Zaccaria, A. (Grenoble)[2], El Atifi, M. (Grenoble)[2], Benabid, A.L. (Grenoble)[1], Chabardes, S. (Grenoble)[3], Berger, F. (Grenoble)[1]
[1]CEA-LETI Clinatec, Grenoble, France, [2]GIN Inserm U 836, Team 7, Grenoble, France, [3]CHU de Grenoble, Neurosurgery, Grenoble, France

Deciphering the complex molecular mechanisms and cellular events responsible for neurological diseases is steadily growing in biomedical consideration. The emergence of poly-omics techniques in the neurology field, particularly neurogenomics and neuroproteomics, holds the promise of significant progress in the identification of important pathways and mechanisms involved in brain neuropathologies. However, limited access to patients' functional brain tissue samples and the restriction to post-mortem material have impeded relevant molecular annotation of neurodegenerative or psychiatric diseases. Therefore, a technological breakthrough providing specific micro/nano devices and strategies for relevant brain tissue poly-omics explorations is essential.
Given that deep brain stimulation (DBS) is the only non-lesional approach to explore functional brain locations, we first described an intracerebral fingerprint approach for molecular investigation of neuropathological brain tissue samples from patients undergoing DBS surgery. Scanning electronic microscopy and two-photon revealed the presence of brain micro-samples harvested on the stylet used during chronic electrode implantation. We demonstrated the feasibility of in depth proteomic and transcriptomic analyses on such brain tissue fingerprint samples. The identification of molecular pathways previously described in neurological disorders as well as several biological mechanisms associated with the CNS confirmed the cerebral origin of the sample.
To further improve the tissue fingerprint approach, we developed a dedicated surgical tool based on surface micro/nano-structuration technology. This silicon-based device directly implementable on routine surgical tools enabled a spatially organized brain tissue harvesting and increased the biocapture capacity.

Availability of fresh neuropathological human brain tissues compatible with poly-omic explorations provides an unprecedented opportunity to better understand the mechanisms underlying neurodegenerative or psychiatric diseases. This tissue fingerprint approach is simple, low cost and readily translatable in routine neurosurgery, and could complement postmortem human brain banks with original and valuable fresh samples at the time of disease progression.

P3.070

Effects of the genetic invalidation of 5-HT_3 receptors on chronic citalopram-induced 5-HT_{1A} autoreceptor desensitization in mice

Martin, V. (Paris)[1], Mongeau, R. (Paris)[2], Hamon, M. (Paris)[1], Lanfumey, L. (Paris)[1]
[1]INSERM U 894, Paris, France, [2]EA 4475 Pharmacologie de la circulation cérébrale, Université Paris Descartes, Paris, France

The antidepressant/anxiolytic action of chronic treatment with selective serotonin reuptake inhibitors (SSRI) is, at least partly, dependent on the functional desensitization of 5-HT_{1A} autoreceptors located on dorsal raphé (DR) serotonergic neurons. It has been recently proposed that the pharmacological blockade of 5-HT_3 receptors could accelerate SSRI-induced 5-HT_{1A} autoreceptor desensitization, thereby promoting the therapeutic efficacy of SSRI. In order to assess this hypothesis, we investigated (i) the phenotype of constitutive 5-HT_3 receptor knock-out (KO) mice using validated paradigms relevant to “depression-like” and “anxiety-like” behaviors, and
(ii) the kinetics of 5-HT_{1A} autoreceptor desensitization during chronic SSRI treatment in 5-HT_3 KO vs wild-type (WT) mice, using an *in vitro* electrophysiological approach.
In basal conditions, 5-HT_3 KO mutants spent more time than paired WT mice in active social interactions and in the open arms of the elevated plus maze. In the forced swim test, the immobility time of mutants was less than that of WT mice, while no differences between both genotypes were observed in the tail suspension test. A 14-day citalopram treatment (20 mg/kg i.p. /day) produced a similar decrease in the potency of the 5-HT_{1A} receptor agonist ipsapirone to dose-dependently inhibit the firing of DR 5-HT neurons in 5-HT_3 KO and WT mice. Further investigations at 3 and 7 days after starting citalopram treatment indicated that ipsapirone (60 nM) was equally effective to inhibit DR 5-HT neuron firing in brainstem slices from 5-HT_3 KO and WT mice. These results show that the time-course and amplitude of 5-HT_{1A} autoreceptor desensitization during chronic citalopram treatment were similar whether or not mice express 5-HT_3 receptors.
It can therefore be concluded that, under our conditions, the genetic invalidation of 5-HT_3 receptors does not accelerate the desensitization of somatodendritic 5-HT_{1A} receptors that progressively develops during chronic SSRI treatment. However, the anxiolytic- and antidepressive-like phenotype of 5-HT_3 KO mice further support the idea that 5-HT_3 receptors play an important role in the modulation of depression- and anxiety-related behaviors and may be a pertinent target for treating anxio-depressive disorders.

P3.071

Ischemic penumbra prediction using local blood oxygen saturation quantification by MRI

Barbier, E. (Grenoble)[1], Moisan, A. (Grenoble)[1,2], Coquery, N. (Grenoble)[1], Bouzat, P. (Grenoble)[1,3], Rémy, C. (Grenoble)[1], Detante, O. (Grenoble)[1,4]
[1]Grenoble Institut des Neurosciences U 836, Grenoble, France, [2]Unité mixte de thérapie cellulaire, Grenoble, France, [3]CHU de Grenoble, Grenoble, France, [4]Unité Neuro-Vacsulaire - Neurologie, CHU de Grenoble, Grenoble, France

Therapy in acute ischemic stroke can only be effective in presence of salvageable tissue in the brain ischemic area. However, the ability to distinguish areas of non-salvageable tissue (ischemic core, necrosis) from those of salvageable tissue (penumbra) remains a goal of diagnostic imaging to select patients who might benefit from thrombolysis. Ischemic penumbra is defined by MRI as the mismatch between the areas with abnormal diffusion (predictive of the necrotic core) and those with diminished perfusion. However, this method remains controversial as many comparative studies reported an overestimation of necrosis and penumbra comparing to TEP imaging. Quantitative MRI methods to measure the local blood oxygen saturation appears a promising tool to better discriminate penumbra and necrosis.
Methods: Twelve Wistar rats underwent a focal cerebral ischemia by occlusion of the right middle cerebral artery (MCAo). MR experiments were performed at 4.7T using a volume / surface cross coil configuration. Blood volume fraction (BVf), apparent coefficient diffusion (ADC) and local blood oxygen saturation (lSO2) were measured once the MCA was occluded. The entire MRI protocol lasted 25 min per animal and began 30 minutes after MCAo. BVf was computed from the change in T2* measured before and after injection of iron oxide particles. SO_2 was computed from the difference between 1/T2 and 1/T2* measured prior to iron oxide injection. Measures were performed in regions of interest (ROI) manually delineated on both ADC map (decreased ADC ROI) and lSO2 map (lSO2 < 40%, Hypoxia ROI).
Results: We observed a mismatch between the decreased ADC ROI defined on the ADC map and the hypoxia ROI delineated on the lSO2 map. Mean volume decreased ADC ROI was larger than Hypoxia ROI (60,9±57,1 mm3 vs 32,5±39,3 mm3) with similar ADC values (591±74 µm².s-1 vs 614±81 µm².s-1). Hypoxia ROI were always included in the decreased ADC ROI. BVf and lSO_2 values were smaller in Hypoxia ROI (BVf=1,5 ±0,5%, lSO2=28,4±5,4%) compared with decreased ADC ROI (BVf=2,3±0,9%, lSO2=48,1±10,1%).
Conclusion: Hypoxia ROI could reflect necrosis (ischemic core) whereas the mismatch decreased ADC / Hypoxia could represent the ischemic penumbra with maintained blood flow allowing a sufficient oxygenation.

P3.072

Interferon-beta induces clearance of mutant ataxin-7 and improves locomotion in SCA7 knock-in mice

Sittler, A. (Paris)[1], Chort, A. (Paris)[1], Alves, S. (Paris)[1], Marinello, M. (Paris)[1], Dufresnois, B. (Paris)[1], Dornbierer, J.G. (Paris)[1], Tesson, C. (Paris)[1], Latouche, M. (Paris)[1], Baker, D. (Cambridge)[2], Barkats, M. (Paris)[3], El Hachimi, K.H. (Paris)[1], Ruberg, M. (Paris)[1], Janer, A. (Paris)[1], Stevanin, G. (Paris)[1], Brice, A. (Paris)[1]
[1]CRICM UMR_S INSERM 975, CNRS 7225, UPMC Univ, Paris, France, [2]Biogen Idec, Cambridge, United States, [3]INSERM U 974, CNRS 7215, UPMC, Paris, France

We showed previously, in a cell model of spinocerebellar ataxia SCA7 that interferon-beta induces the expression of promyelocytic leukemia protein (PML) and the formation of PML nuclear bodies that degrade mutant ataxin-7, suggesting that the cytokine, used to treat multiple sclerosis, might have therapeutic value in SCA7. We now show that interferon-beta also induces PML-dependent clearance of ataxin-7 in a preclinical model, $SCA7^{266Q/5Q}$ knock-in mice, and improves motor function. Interestingly, the presence of mutant ataxin-7 in the mice induces itself the expression of endogenous interferon beta and its receptor. Immunohistological studies in brains from two SCA7 patients confirmed that these modifications are also caused by the disease in humans. Interferon-beta, administered intraperitoneally three times a week in the knock-in mice, was internalized with its receptor in Purkinje and other cells and translocated to the nucleus. The treatment induced PML expression and the formation of PML nuclear bodies and decreased mutant ataxin-7 in neuronal intranuclear inclusions, the hallmark of the disease. No reactive gliosis or other signs of toxicity were observed in the brain or internal organs. The performance of the $SCA7^{266Q/5Q}$ knock-in mice was significantly improved on two behavioral tests sensitive to cerebellar function: the Locotronic test of locomotor function and the beam-walking test of balance, motor coordination and fine movements, which are affected in patients with SCA7. In addition to motor dysfunction, $SCA7^{266Q/5Q}$ mice present abnormalities in the retina as in patients: ataxin-7-positive neuronal intranuclear inclusions which were

reduced by interferon-beta treatment. Finally, since neuronal death does not occur in the cerebellum of SCA7$^{266Q/5Q}$ mice, we showed in primary cell cultures expressing mutant ataxin-7 that interferon-beta treatment improves Purkinje cell survival. This treatment might apply to all polyglutamine diseases in which PML associates, in cell nuclei, with the mutant proteins responsible for the disease.

P3.073

Intermediate filament disorganization in neurons derived from giant axonal neuropathy mouse model

Scrivo, A. (Montpellier)[1], Bomont, P. (Montpellier)[1]
[1]*INM - INSERM U 1051, Montpellier, France*

Giant axonal neuropathy (GAN) is a fatale neurodegenerative disorder affecting both the peripheral and central nervous systems and inducing a generalized disorganization of the cytoskeletal Intermediate Filament (IF). GAN is caused by recessively inherited mutations in the gene encoding gigaxonin, a new BTB-Kelch protein that is shown to be low abundant and expressed throughout the nervous system. Proposed as a substrate adaptor of E3 ubiquitin ligase complexes, gigaxonin participates in the degradation of its partners by the Ubiquitin Proteaseome System (UPS) but its direct function as an E3 ligase component still awaits to be demonstrated.
In attempts to decipher the key role(s) of gigaxonin in sustaining neuron survival and IF integrity, we generated a knock out mouse model for GAN. The GAN mice exhibit motor and sensory deficits characteristics of the human pathology but to a milder extend and do not present any overt neurodegeneration. Nevertheless, we demonstrated that depletion of gigaxonin in mouse induces a severe alteration of IF architecture, as revealed in the human pathology. This includes a spatial disorganization of neurofilaments and an increase abundance of all three neurofilament subunits in neuronal tissues. To further dissect the mechanisms leading to IF aggregation, we developed an *in vitro* model for GAN, the GAN cortical neurons.

P3.074

Frontal-amygdala alterations during emotional conflict processing in euthymic bipolar patients: a word-face Stroop fMRI study

Favre, P. (Grenoble)[1], Polosan, M. (Grenoble)[2,3], Pichat, C. (Grenoble)[1], Bougerol, T. (Grenoble)[2], Baciu, M. (Grenoble)[1]
[1]*Laboratoire de Psychologie et de NeuroCognition, UMR CNRS 5105, Université Pierre Mendès France, Grenoble, France,* [2]*Centre Hospitalier Universitaire de Grenoble, Pôle Psychiatrie et Neurologie, Grenoble, France,* [3]*Institut des Neurosciences de Grenoble, Equipe 'Fonctions Cérébrales et Neuromodulation', Université Joseph Fourier, Grenoble, France*

Objectives: Acute episode in bipolar disorders (BD) are mainly characterized by emotion regulation impairment, which is also present during remission periods. This impairment may thus represent a trait marker of the disease and better understanding its cerebral correlates could improve diagnosis and treatment of this disorder. This fMRI study aims to assess fronto-limbic functioning in euthymic bipolar patients (EBP) with a task specifically designed to involve emotional regulation mechanisms.
Methods: Fourteen EBP and 13 matched healthy subjects (HS) were scanned while performing a word-face emotional Stroop task. This paradigm, adapted from Etkin et al (2006), allowed us to dissociate the generation and monitoring of emotional conflict (low vs. high conflict trials), from its resolution (high vs. low conflict trials). Between-group analyses were made to assess group differences in activation. Region of interest (ROI) analysis focused on amygdala activation was also

conducted, as we had specific hypotheses for its involvement in mood disorders. Additionally, correlations analyses between clinical variables and cerebral activation in EBP were tested.
Results: Whole-brain analyses reveal significant decrease activation in EBP compared to HS during emotional conflict monitoring in the right middle frontal gyrus (BA 46), in bilateral superior frontal gyri (BA 10) and also in bilateral middle temporal gyri (BA 21), precuneus (BA 23, 30, 32) and supramarginal gyri (BA 40). The ROI analysis reveals significant increase activation in the left amygdala in EBP compared to HS during emotional conflict resolution. Moreover a significant negative correlation between left amygdala mean activity and the age of the disease onset was found.
Conclusions: Deficit in prefrontal activations in EBP suggests a dysfunction in cognitive control during emotional conflict monitoring. The abnormal activation of the amygdala during emotional conflict resolution in EBP might be secondary to the failure of these cognitive control processes. Dysfunction of the emotion regulation network may thus contribute to residual symptoms during euthymic periods and may be more severe in early onset disorders.

P3.075
Aging and cortical excitability (TMS)

Bécu, M. (Grenoble)[1], Beynel, L. (Grenoble)[1], Chauvin, A. (Grenoble)[1], Guyader, N. (Grenoble)[2], Harquel, S. (Grenoble)[1,3], Marendaz, C. (Grenoble)[1]
[1]Laboratoire de Psychologie et de NeuroCognition, UMR CNRS 5105, Grenoble, France, [2]GIPSA-lab, Grenoble, France, [3]UMS "IRMaGe" CNRS UMS 017, Grenoble, France

Cortical excitability (CE) refers to the neural response to magnetic stimulation. CE is generally assessed on the motor cortex using transcranial magnetic stimulation (TMS). Paired-pulse TMS separated by various inter-stimuli interval (ISI) allow an indirect measure of neurotransmission: an ISI of 2ms gives rise to intracortical inhibition (ICI), while an ISI of 15ms leads to intracortical facilitation (ICF), involving respectively GABA and glutamate transmission. CE has been largely studied in mood disorder: there is growing evidence of an inhibition deficit in major depressive disorder (MDD). This deficit tends to be seen as a biomarker of MDD (Malsert et al., 2013). However, patients suffering from MDD are middle-aged or old age and it might be that inhibitory deficits were due to aging too. If some studies in healthy elderly people concluded to an inhibition deficit as well (Peinneman et al., 2001), due to small samples sizes and discrepancies among protocols, parameters and measures, it is in fact difficult to conclude about CE in aging. The present experiment was aiming to study whether cortical excitability varied with aging.
Method: Motor cortex of 20 young healthy subjects (< 40 years old) *vs.* 20 elderly healthy subjects (>60) was stimulated and motor evoked potentials (MEP) were collected on the contralateral first dorsal interosseous muscle.
Procedure:
- Motor Threshold: single pulses TMS to find the hotspot giving the best MEPs amplitudes and 50% motor response > 50µV.
- Baseline: 10 single pulses at 120% of Resting Motor Threshold.
- ICI/ICF: 10 paired pulses with ISI 2ms, 10 paired pulses with ISI 15ms, and 10 single pulse stimulations of Baseline (randomized).
A slight cognitive task was added to the protocol to control subjects' mental state.
Results: Experiment is in progress. Data obtained up to now show a significant reduction of ICI in elderly subjects.
Discussion: Several lines of evidence suggest that inhibition deficits are closely related to pathophysiology of MDD. However it might be that similar deficits are related to physiology of aging as well. As patients suffering from MDD are often rather aged, future researches on biomarkers of MDD will have to take age into account, as it could be a confounding factor.

P3.076

Identification of a new causal gene for autosomal dominant focal epilepsies

Baulac, S. (Paris)[1], Ishida, S. (Paris)[2], Picard, F. (Genève)[3], Rudolf, G. (Strasbourg)[4], Thomas, P. (Nice)[5], Genton, P. (Marseille)[6], Marescaux, C. (Strasbourg)[4], Miles, R. (Paris)[2], Baulac, M. (Paris)[2], Hirsch, E. (Strasbourg)[4], Leguern, E. (Paris)[2]
[1]ICM, Hopital de la Pitié-Salpetrière, Paris, France, [2]ICM, Paris, France, [3]Hôpitaux Universitaires de Genève, Genève, Switzerland, [4]Strasbourg University Hospital, Strasbourg, France, [5]Hopital Pasteur, Nice, France, [6]Centre Saint Paul, Marseille, France

Purpose: The main familial focal epilepsies are autosomal-dominant nocturnal frontal lobe epilepsy (ADNFLE), familial mesial temporal lobe epilepsy (FMTLE), familial lateral TLE (FLTLE), and familial focal epilepsy with variable foci (FFEVF). We aimed to identify a new gene in a family with FMTLE.
Method: We performed linkage analysis using a high-density genome-wide scan with 10,000 SNPs followed by exome sequencing in a large family with FMTLE. A cohort of 15 families with focal epilepsies was subsequently analyzed.
Results: A frameshift mutation in a new gene was identified in the FMTLE family. This ancient eukaryotic gene encodes a protein of unknown function.
Subsequent screening of 15 additional families revealed four nonsense mutations and one missense mutation in 5 families. All mutations were shown to fully segregate within the families. We demonstrated that one nonsense mutation specifically leads to mRNA degradation by nonsense mediated decay system (NMD). The three additional nonsense mutations were predicted to be degraded by the NMD. Mutations were found in families with different phenotypes: ADNFLE, FMLTE and FFEVF.
Conclusion: We report a new gene with frequent loss-of-function mutations (37%) within a broad spectrum of familial focal epilepsies. The implication of this gene will open new avenues for research.

P3.077

Regional expression and subcellular localization of the intracellular Na^+/H^+ exchanger NHE-7 in the rodent brain

Léna, I. (Nice)[1], Poet, M. (Nice)[1], Counillon, L. (Nice)[1]
[1]Université de Nice-Sophia Antipolis, FRE 3472 CNRS, Faculté des Sciences, Nice, France

To date, nine isoforms of Na^+/H^+ exchangers (NHEs) have been identified and mutations in NHE-6, 7 and 9 genes have been associated to brain disorders such as mental retardation, autism or attention-deficit/hyperactivity disorder. In this work we examined the neuroanatomical distribution of the Na^+/H^+ exchanger 7 isoform (NHE-7), at the protein level, in the rodent brain during adulthood and postnatal development. The highest level of NHE-7 in the adult mouse brain was found in the Purkinje cells of the cerebellum. Intense immunolabeling was also observed in the pyramidal cells of the hippocampus and motor neurons of the brainstem. Immunostaining was less prominent in the cortex, the amygdala, the olfactory bulb or the caudate-putamen. During post-natal development, NHE-7 could be detected at its adult locations quickly after birth. Furthermore, we studied the subcellular localization of NHE-7 using markers of the endosomal system. Taken together our results provide new clues to the physiological role of this intracellular exchanger in the brain and to associated brain disorders.

P3.078

Leucettine L41, a DYRK1A inhibitor, prevents memory impairments and neurotoxicity induced by icv administration of aggregated $A\beta_{25-35}$ in mice

Naert, G. (Montpellier)[1], Ferre, V. (Montpellier)[2], Meunier, J. (Montpellier)[2], Keller, E. (Montpellier)[2], Givalois, L. (Montpellier)[2], Carreaux, F. (Rennes)[3], Bazureau, J.-P. (Rennes)[3], Meijer, L. (Roscoff)[4]
[1]UMII INSERM U 710, Montpellier, France, [2]U 710, Montpellier, France, [3]UMR CNRS 6226, Rennes, France, [4]ManRos Therapeutics, Roscoff, France

Alzheimer's disease (AD) is characterized by the accumulation of amyloid-beta (Aβ) and the hyperphosphorylation of the Tau protein. More and more evidence highlights kinases as a possible link between Aβ and Tau in AD, and 'dual specificity, tyrosine phosphorylation regulated kinase 1A' (DYRK1A) has emerged as an important kinase in AD development. Therefore our objective was to

assess the neuroprotective potential of a DYRK1A inhibitor, the Leucettine L41, in the $A\beta_{25-35}$ mouse model. We co-injected intracerebroventricularly Swiss mice male with aggregated $A\beta_{25-35}$ peptide and L41. The mice were submitted to behavioral tests to address spatial and non-spatial, short- and long-term memories. The oxidative stress, apoptotic markers, kinases involved in Tau phosphorylation and synaptic integrity were analyzed by Western blotting and ELISA in the hippocampus. L41, at the dose of 4 µg, restored the mnesic deficits observed in the Y-maze, passive avoidance and water-maze tests in the $A\beta_{25-35}$ mouse model. In the hippocampus, L41 prevented the $A\beta_{25-35}$-induced oxidative stress, as revealed by measures of lipid peroxidation and ROS accumulation, and abolished $A\beta_{25-35}$-induced expression of proapoptotic markers. In addition, L41 prevented the decreased activation of Akt and enhanced activation of GSK3-β induced by $A\beta_{25-35}$, resulting in a decreased phosphorylation of Tau. Finally, L41 restored $A\beta_{25-35}$-reduced levels of synaptic markers. Leucettine L41, a DYRK1A inhibitor, prevent memory impairments and neurotoxicity induced by $A\beta_{25-35}$ in the mouse hippocampus. This first *in vivo* data highlights DYRK1A as a major kinase involved in Aβ pathology and DYRK1A inhibitors as potential therapeutics in AD.

P3.079

Nicotinic inhibition of a subpopulation of ventral tegmental area dopamine neurons

Valverde, S. (Paris)[1], Eddine, R. (Paris)[1], Tolu, S. (Paris)[1], Marti, F. (Paris)[1], Morel, C. (Paris)[1], Faure, P. (Paris)[1]
[1]*Université Pierre et Marie Curie, Paris, France*

Midbrain dopamine (DA) neurons are importantly involved in motivation and reward processing, and increase of dopamine release is thought to be central in the initiation of drug addiction. On the other side, inhibition of DA neurons has been linked to the processing of aversive stimuli. Our lab reports that the addictive drug nicotine can induce inhibition of neuronal firing in a subset of VTA DA neurons. This previously uncharacterized DA subpopulation indicates a new role of dopamine release contrasting with the classical views of reinforcement and motivation. This raises new questions about the role of the VTA in the initiation of drug addiction.

P3.080

Proactive inhibitory control dysfunctions may induce similar hypo- and hyper-reactivity symptoms in schizophrenia and Parkinson's disease

Boulinguez, P. (Bron)[1], Favre, E. (Bron)[1], Demily, C. (Lyon)[1,2,3], Franck, N. (Lyon)[1,3], Broussolle, E. (Lyon)[1,4], Thobois, S. (Lyon)[1,4], Ballanger, B. (Bron)[1]
[1]*Université Claude Bernard Lyon 1, Centre de Neuroscience Cognitive - UMR 5229 CNRS, Bron, France,* [2]*Centre Hospitalier St Jean de Dieu, Lyon, France,* [3]*Centre Hospitalier Le Vinatier, Lyon, France,* [4]*Hôpital Neurologique Pierre Wertheimer, Lyon, France*

Recent studies conducted on healthy human subjects showed that, when a situation is unpredictable, preventing automatic responses to potentially inappropriate stimuli is mainly achieved by proactive inhibitory control of movement initiation. Proactive top-down control would allow switching from controlled inhibition of response (anticipated suppression of the neuronal processes underlying movement initiation) to automatic processing of forthcoming sensorimotor information (unlocked state), depending on the expectations of upcoming events. The integrity of this function would be necessary to ensure the ability to prevent prepotent responses to potentially inappropriate stimuli, but also the ability to initiate responses to appropriate stimuli.
Here, we report the outcomes of behavioural experiments specifically designed to test proactive inhibitory control in 37 schizophrenic (SCZ), 25 parkisonian (PD) patients, and 43 controls. The results

broadly show that troubles in implementing proactive inhibition result in impulsive behavior, while troubles in releasing proactive inhibition induce movement initiation deficits.
SCZ patients were differently impaired in their ability to control proactive inhibition, 46% of the patients falling into the hypo-reactive category and 22% into the hyper-reactive category. No correlation was found between these dysfunctions and the clinical symptoms assessed by the main diagnostic tools in psychiatry.
In PD patients, the impairment of proactive inhibitory control resulted in a slowing down of movement initiation, accounting for akinesia. STN-DBS either restored this deficit or induced opposite impulsive behavior, highlighting the role the STN may play as an interface between executive and motor systems in cortico-basal ganglia loops.
Taken together, these results highlight the current need to refine diagnostic tests by providing objective, quantitative assessment of specific neurocognitive processes, with possible aetiological relevance.

P3.081

Regulation of endoplasmic reticulum function by spatacsin and spastizin, two proteins associated with hereditary spastic paraplegias

Darios, F. (Paris)[1], Brunet de Courssou, J.-B. (Paris)[1], Fuentealba, J. (Paris)[1], Esteves, T. (Paris)[1], Brice, A. (Paris)[1], Stevanin, G. (Paris)[1]
[1]*Institut du Cerveau et de la Moelle Épinière - Cricm - Neb, Paris, France*

The hereditary spastic paraplegias (HSP) are rare genetic neurological disorders mainly characterized by progressive spasticity. The symptoms are caused by axonal degeneration of the cortical motor neurons, especially in their terminal portions. More than 30 causative genes have been identified so far. The putative roles of the encoded proteins suggest that mitochondrial function, protein folding and intracellular trafficking are implicated in the dying back of pyramidal tract axons in these disorders.
A common form of autosomal recessive HSP associates spastic paraplegia with thin *corpus callosum.* Most families (~60-80%) are linked to SPG11, while ~11% of the families are linked to SPG15. *SPG11* and *SPG15* encode spatacsin and spastizin, respectively, two proteins of unknown function. In most cases, the pathology is due to loss of protein function. Both genetic entities are clinically very similar, both genes have similar patterns of expression, and spatacsin and spastizin are present in a same protein complex. Together these data suggest that spatacsin and spastizin very likely play a role in a same cellular pathway. Subcellular localization experiments showed that spatacsin is mainly colocalized with endoplasmic reticulum while spastizin is partly colocalized with this organelle as well as with microtubules. This observation led us to investigate whether the complex formed by spatacsin and spastizin could play a role in regulating the morphology or the dynamics of endoplasmic reticulum. This analysis was performed by live cell imaging using fluorescent proteins addressed to endoplasmic reticulum. The role of spatacsin and spastizin was monitored using RNA interference in cultured cell lines and by examining fibroblasts derived from patients. Preliminary data indicate that the complex formed by spatacsin and spastizin is important for the extension of endoplasmic reticulum network along the microtubules.

P3.082

Loss of dopaminergic nigrostriatal neurons accounts for the motivational and affective deficits in Parkinson's disease

Drui, G. (Grenoble)[1], Carnicella, S. (Grenoble)[1], Carcenac, C. (Grenoble)[1], Favier, M. (Grenoble)[1], Bertrand, A. (Grenoble)[1], Boulet, S. (Grenoble)[1], Savasta, M. (Grenoble)[1]

[1]*U 836 - Equipe 10 - Grenoble Institut des Neurosciences - Université Joseph Fourier, Grenoble, France*

Parkinson's Disease (PD) involves the degeneration of dopaminergic (DA) neurons in the substantia nigra pars compacta (SNc) that is thought to cause the classical motor symptoms of this disease. However, motivational and affective impairments are also often observed in PD patients. These are usually attributed to a psychological reaction to the general motor impairment and to a loss of some of the neurons within the ventral tegmental area (VTA). We induced selective lesions of the VTA and SNc DA neurons that did not provoke motor deficits, and showed that bilateral dopamine loss within the SNc, but not within the VTA, induces motivational deficits and affective impairments that mimicked the symptoms of PD patients. Importantly, these deficits were reversed by DA pharmacological treatments commonly used in PD. Thus, motivational and affective deficits are a core impairment of PD, as they stem from the loss of the major group of neurons that degenerates in this disease (DA SNc neurons) and are independent of motor deficits.

P3.083

BRG1 interacts with glucocorticoid receptor in dopaminoceptive neurons to modulate anxiety-like and abused drug-like responses

Baranowski, C.S. (Paris)[1], Karaki, S. (Paris)[1], Mailly, P. (Paris)[2], Boudier, T. (Paris)[2], Barik, J. (Paris)[1], Tronche, F. (Paris)[1]
[1]*Univ. Pierre et Marie Curie, INSERM UMRS 952 - CNRS UMR 7224 Génétique Moléculaire, neurophysiologie et comportement, Paris, France,* [2]*IFR 83 Biologie Intégrative, Paris, France*

Addiction is a chronic relapsing disorder underpinned by persisting neuroadaptations involving gene transcription. Alteration of the chromatin architecture by chromatin remodeling complexes, such as the SWI/SNF complex, is an essential step in transcriptional regulation of many genes. This multi-subunit complex contains either BRM (Brahma) or BRG1 (Brahma-related gene 1) as central ATPase subunit, which uses the energy of ATP to disrupt nucleosome DNA contacts, move nucleosomes along DNA and remove or exchange nucleosomes. It has been shown, in cell culture, that SWI/SNF subunits mediate critical interactions with nuclear receptors such as the glucocorticoid receptor (GR), and that a large proportion of GR-induced or -repressed genes requires the function of BRG1-containing remodeling complex. An essential role of GR in dopaminoceptive neurons has been shown in the modulation of cocaine. Using co-immunoprecipitation, we detected an interaction between GR and BRG1, and between GR and BRM in the striatum. To understand the role of BRM and BRG1 proteins, we developed animals with a constitutive BRM inactivation and/or with a conditional deletion of BRG1 in dopaminoceptive neurons, (*brm*$^{-/-}$*; Brg1*^{D1Cre} mice). We have studied cocaine-induced behavioral sensitization and demonstrated a different role of BRG1 and BRM. To get further insights into the role of chromatin remodelers in behavioral responses to cocaine we are currently investigating cocaine-induced conditioned place preference. Anxiety-like behaviors are also affected by the absence of these proteins. Furthermore, to decipher the interaction pattern of BRG1 and GR within the nucleus and to study consequences of the deletion of either one of these proteins, we performed immunofluorescence combined with confocal microscopy, using BRM$^{-/-}$BRG1^{D1Cre}, BRM$^{-/-}$, BRG1^{D1Cre}, and GR^{D1Cre} mice. In conclusion, we provide *in vivo* evidence that BRG1 in dopaminoceptive neurons is required for the modulation of behaviors that may be relevant for psychiatric disorders.

P3.084

Specific reversal of loss of motivated behaviors by dopamine D3 receptors stimulation: therapeutic implication for Parkinson´s disease-related neuropsychiatric symptoms

Favier, M. (Grenoble)[1], Carnicella, S. (Grenoble)[1], Drui, G. (Grenoble)[1], Boulet, S. (Grenoble)[1], Carcenac, C. (Grenoble)[1], Duran, T. (Grenoble)[1], Savasta, M. (Grenoble)[1]
[1]*Grenoble Institut des Neurosciences, Grenoble, France*

Beyond the classical triad of motor symptoms observed in Parkinson's disease (PD), behavioral and cognitive disturbances are also commonly observed, including severe motivational and affective impairments. We previously found that a partial and bilateral dopaminergic lesion of the substantia nigra compacta (SNc) in rats, the major group of neurons that degenerates in PD, recapitulates PD-related neuropsychiatric symptoms, including specific loss of motivation in instrumental procedures and anxiety- and depressive-like behaviors. Interestingly, these deficits were reversed by DA pharmacological treatments commonly used in PD. By using, selective or preferential agonists of D1 (SKF-38393), D2 (Sumanirole) and D3 (PD-128907) DA receptors, we aim at determine whether a specific receptor subtype would be involved in this reversal effect. We showed that, while affective-related impairments were relatively responsive to all agonists, only the preferential D3 agonist fully reversed the motivational deficits induced by the DA SNc lesion. These data therefore provide a new insight in the pathophysiology of motivational and affective dysfunction in PD and put forward the DA D3 receptor as a potential key target for the treatment of these neuropsychiatric symptoms.

P3.085

Chronic neglect and disconnection of white matter pathways: a longitudinal study

Lunven, M. (Boissise-Le-Roi)[1,2,3], Thiebaut de Schotten, M. (London)[4,5], Duret, C. (Boissise-Le-Roi)[1], Bourlon, C. (Boissise-Le-Roi)[1], Migliaccio, R. (Paris)[3], Rode, G. (Bron)[2,6], Bartolomeo, P. (Paris)[3,7]
[1]Clinique Les Trois Soleils, Boissise-Le-Roi, France, [2]Inserm 1028 CNRS UMR S 292 Equipe ImpAct, Bron, France, [3]Inserm UMR S 975, Hôpital Pitié Salpétrière, Paris, France, [4]Natbrainlab, King's College, London, United Kingdom, [5]CNRS UMR 7225, hôpital Pitié Salpétrière, Paris, France, [6]Hôpital Henry Gabrielle, Saint-Genis-Laval, France, [7]Department of Psychology, Catholic University, Milan, Italy

Unilateral spatial neglect frequently occurs after right-hemisphere stroke. It represents a major problem in the domain of public health, since it prevents patients from orienting or responding to left-sided stimuli. The exact anatomical location of lesions underlying the manifestation of this syndrome is currently debated (Bartolomeo, 2012). In the present study, we used a longitudinal approach in order to identify the lesional predictors of chronic neglect in long-range white matter bundles.
We present a longitudinal study of 37 patients with right-hemisphere damage, tested at the acute/subacute phase and at more than 1 year after the stroke. 27 patients presented signs of spatial neglect in the acute/subacute phase. Each patient underwent a radiological assessment including a DTI sequence, (50 directions; bvalue of 1000 mm2/s). Voxelwise statistical analysis of the fractional anisotropy (FA) data was carried out using TBSS (Tract-Based Spatial Statistics, Smith, 2006).
The longitudinal follow-up revealed that only 10 patients (27%) recovered from neglect at retest. In acute/subacute neglect, a lower FA was found in the way of the right Superior Longitudinal fasciculus (SLF II and III). In the chronic phase, TBSS analysis showed the implication of the posterior portion of the corpus callosum (splenium) and of the forceps major. The comparison between patients with persistent neglect and patients without neglect in the chronic phase also indicated a significant decrease of FA in the posterior segment of the arcuate fasciculus. Damage to SLF III was found in chronic neglect patients when compared to non-neglect patients.
Our results confirm a key role of fronto-parietal disconnection in the emergence and chronic persistence of neglect (Thiebaut de Schotten and al, in press). Moreover, we demonstrated an implication of interhemispheric disconnection (splenium and forceps major) in chronic neglect. These findings support the hypothesis that interhemispheric disconnection may deprive the right fronto-parietal pathway of visual inputs, amputing the brain reconstruction of the left hemi-space (Tomaiuolo and al, 2010), and that chronic neglect at least in part results from the activity of an isolated left hemisphere (Bartolomeo and al, 2007).

P3.086

beta-CTF-correlated burst of hippocampal TNF-alpha occurs at a very early, pre-plaque stage in the TgCRND8 mouse model of Alzheimer's disease

Krantic, S. (Paris)[1], Cavanagh, C. (Montreal)[2], Colby-Milley, J. (Montreal)[2], Bouvier, D. (Montreal)[2], Farso, M. (Montreal)[2], Chabot, J.-G. (Montreal)[2], Quirion, R. (Montreal)[2]

[1]Centre de Recherche des Cordeliers, UMRS 872, Equipe 19, Paris, France, [2]Douglas Mental Health University Institute (DMHUI), Dept of Psychiatry, McGill University, Montreal, Canada

Tumor Necrosis Factor-alpha (TNFα) regulates neuronal excitability. We investigated whether alterations in the level of TNFα occur at a time point that precedes the reported seizure-associated hyperexcitability of hippocampal networks in pre-plaque models of Alzheimer's disease (AD). Western blot and ELISA experiments indicated a significant increase in hippocampal TNFα expression in 1-month-old TgCRND8 mice that correlated with levels of the β-C-terminal fragment (βCTF) of amyloid-β (Aβ) protein precursor (AβPP). Such increase thus occurs at least 1 month before plaque burden (Chishti MA et al., 2001), neuroinflammation (Dudal S et al., 2004) and increased hippocampal susceptibility to seizures (Del Vecchio RA et al, 2004), as previously reported in this AD model. CD11b labeling and analysis by confocal microscopy indicated changes in microglial morphology towards an activated state. Taken together, these data suggest that microglia may be a putative source of the observed TNFα increase during this pre-symptomatic stage of AD-like pathology. Future experiments are needed to explain whether an initial βCTF-correlated increase in TNFα may function as a compensatory mechanism for Aβ-mediated impairments in synaptic transmission.

P3.087

Dopamine control of the globus pallidus and its impact on the subthalamic nucleus and the pars reticulate of substantia nigra

Mamad, O. (Rabat)[1,2], Delaville, C. (Bordeaux)[2], Benjelloun, W. (Rabat)[1], Benazzouz, A. (Bordeaux)[2]
[1]Université Mohamed 5 Faculté des Sciences Rabat, Rabat, Morocco, [2]Institut des Maladies Neurodégénératives, Equipe Monoamines, SCP et Parkinson, Bordeaux, France

In addition to GABA and glutamate innervations, globus pallidus (GP) neurons receive dopamine inputs from the *substantia nigra pars compacta* (SNc). However, the functional role of this pathway is not determined. Therefore we aimed to investigate the role of dopamine in the GP and its impact on the modulation of pallido-subthalamic (GP-STN) and pallido-nigral (GP-SNr) pathways. To this end, extracellular recordings were carried out in male Sprague Dawley rats under urethane anesthesia in GP, STN and SNr following the intrapallidal injections of dopamine and quinpirole (D2 dopamine agonist). We show that local injection of dopamine or quipirole increased the firing rate of majority of GP neurons (65%), decreased the firing rate of 5% (14%) neurons and was without any effect for 30% (21%) of GP cells. In parallel, injection of quinpirole in GP decreased the firing rate of majority of STN neurons (63%), with an increase in 29% and without any effect for 8% of STN cells. Accordingly, in SNr quinpirole decreased the firing rate of majority of neurons (82%), increased this rate in only 9% and was without any effect for 9% of SNr cells. In contrast to the firing rate, dopamine as well as quinpirole injection into the GP did not change the firing pattern of GP, STN and SNr neurons. Our results show that D2 dopamine receptors located into the GP play a key role in the modulation of the GABAergic pallido-subthalamic and pallido-nigral pathways. Furthermore, our data challenge assumptions about the important role of extrastriatal dopamine in the modulation of basal ganglia function.

P3.088

Repeated electroconvulsive seizures induce long-term restoration of STOP/MAP6 null mice behavioural disorders

Gory-Fauré, S. (Grenoble)[1], Dall' Igna, G. (Grenoble)[1,2], Chauliac, N. (Grenoble)[1,2], Jonckheere, J. (Grenoble)[1], Deloulme, J.-C. (Grenoble)[1], Andrieux, A. (Grenoble)[1], Holtzmann, J. (Grenoble)[1,2], Suaud-Chagny, M.-F. (Bron)[3]
[1]Grenoble Institut des Neurosciences CR Inserm U 836, Equipe 1, Physiopathologie du Cytosquelette CEA-iRTSV-GPC, Université Joseph Fourier - Site Santé La Tronche, Grenoble, France, [2]Service de Psychiatrie, CHU de Grenoble, Grenoble, France, [3]EA 4615 - SIPAD Stimulation in Psychiatric and Addictive Disorders, UCB Lyon 1, Centre Hospitalier Le Vinatier Bat. 520 - BP 300 39- 95 bd Pinel, Bron, France

The exact mechanisms underlying the actions of electroconvulsivotherapy (ECT) are not yet understood. The current hypotheses come from results obtained with ECS (the animal counter part of ECT). However, results are often inconclusive mainly because data are based on unchallenged animals. As a consequence, the biological effects driven by ECS are rarely time correlated with behavioural improvements. Thus we aim to develop a translational approach in STOP null mice to decipher the biological effects underlying ECT. STOP null mice exhibit some behavioural and biological features, and pharmacological response relevant to some aspect of the major depressive disorder.
The first objective was to study the effects of ECS in STOP null mice compared to WT mice or Sham-treated animals (handled the same way but no current delivered) at the behavioural level. Under anaesthesia, bilateral ECS-treatment was delivered via ear clips 10 times over 2 weeks (1session/day, 5days/week) using an ECS Unit. Stimulation parameters were initially set on the basis of a preliminary study, in order to trigger generalized tonic-clonic seizures lasting for 20-30 s. Mice were tested at different time points after the end of treatment in forced swim test and in conspecific interaction test
In the forced swim test, active ECS-treated STOP null mice exhibited less immobility and more climbing as compared to Sham-treated mice. Whereas STOP null mice have been shown to exhibit severe social withdrawal, after ECS treatment they were no more different from WT animals. The maintenance of the positive action on behavioural defects depended on the parameter tested; the positive effect was of longer duration in conspecific interaction test than in forced swim test.
Altogether results obtained after ECS in STOP null mice recapitulate some features observed after ECT in humans such as behavioural restoration with different kinetics depending of the parameter tested and relapse after treatment. We believe that these results validate STOP null mice as a pertinent model to investigate molecular and cellular changes triggered by ECS treatment with the big advantage of the use of an animal model where extended characterisation is available.

P3.089

Manganese-induced Parkinsonian-like motor and non-motor deficits associated with changes in basal ganglia neuronal activity in the rat

Bouabid, S. (Rabat)[1,2], Delaville, C. (Bordeaux)[2,3], De Deurwaerdère, P. (Bordeaux)[2,3], Lakhdar-Ghazal, N. (Rabat)[1], Benazzouz, A. (Bordeaux)[2,3]
[1]Univ. Mohamed V-Agdal, Faculté des Sciences, Equipe Rythmes Biologiques, Neurosciences et Environnement, Rabat, Morocco, [2]Univ. Bordeaux, Institut des Maladies Neurodégénératives, UMR 5293, Bordeaux, France, [3]CNRS, Institut des Maladies Neurodégénératives, UMR 5293, Bordeaux, France

Manganese (Mn) neurotoxicity is associated with motor and cognitive disturbances known as Manganism. However, the mechanisms underlying these deficits remain unknown. The present study aimed to investigate the effects of Mn on:
1) exploratory and locomotor activities, using an open field actimeter, and motor coordination using the rotarod test,
2) anxiety behavior using the elevated plus maze,
3) anhedonia and “depressive-like” behaviors using sucrose preference and forced swim tests,
4) globus pallidus (GP) and subthalamic nucleus (STN) neuronal activity using extracellular recordings and
5) tissue level of monoamines in the striatum and frontal cortex.
Male Sprague-Dawley rats were daily treated with MnCl2 (10 mg/kg, i.p.) for 5 weeks. We show that Mn progressively reduced exploratory and locomotor activities as well as the time spent on the rotarod. In addition to these motor deficits, Mn induced anxiety and “depressive-like” behaviors. Electrophysiological results show that, while majority of GP and STN neurons discharged regularly in controls, Mn increased the number of GP and STN neurons discharging irregularly and/or with bursts, in addition to a decrease in the firing rate. Interestingly, biochemical results show that Mn significantly decreased tissue levels of norepinephrine and serotonin in the prefrontal cortex with increased level of dopamine and DOPAC in the striatum. Our data provide new evidence supporting the relationship

between Mn intoxication and the alteration of monoaminergic neurotransmission, which may be at the origin of changes in basal ganglia neuronal activity and the manifestation of motor and non-motor deficits similar to those observed in parkinsonism.

P3.090

Implication of the sigma-1 chaperone protein in adult neurogenesis: contribution to neuroprotection?

Dekeuwer, A. (Montpellier)[1], Falco, A. (Montpellier)[2], Maurice, T. (Montpellier)[2], Chevallier, N. (Montpellier)[2]
[1]INSERM U 710/EPHE/UM2, CC 105 Bat 24, Montpellier, France, [2]INSERM U 710/EPHE/UM2, Montpellier, France

Objectives: Adult neurogenesis is characterized by proliferation, survival and differentiation of progenitor cells in the brain. This phenomenon is enhanced in enriched environment, but altered in pathological conditions, including major depression and Alzheimer´s disease (AD). The sigma-1 receptor (S1R) is a ligand-operated chaperone protein. S1R agonists were previously shown to enhance neurogenesis and allow effective neuroprotection in AD animal models. Here we assessed whether the S1R is involved in adult neurogenesis in relation to its neuroprotective activity.
Methods: WT mice were compared to S1R KO in normal or enriched environment (NE vs. EE). First, anxiety and memory capacities were analyzed using open-field, Y-maze and water-maze responses. Second, survival, proliferation and differentiation of progenitor cells in the hippocampus were visualized by BrdU incorporation combined with NeuN immunostaining.
Results: Behavioral phenotyping of S1R KO previously showed that in NE males are more anxious and females present memory decrement as compared to WT. EE increased exploratory behavior in all mice and alleviated the anxious response in KO males. It failed to improve learning deficits in females. In NE proliferation and survival were lower in KO vs. WT animals. EE resulted in increased proliferation in both WT and KO, making the difference between WT and KO fainter.
Conclusions: We report that, in NE, lack of S1R impacts proliferation and survival rates of progenitor cells in the adult hippocampus. Decreased proliferation in the KO is partially compensated by EE. EE improves anxiety in KO males but fails to impact learning deficits in KO females.

P3.091

The psychiatric comorbidity in adolescents with chronic daily headache

Gjeci, S. (Tirana)[1], Rakacolli, M. (Tirana)[1], Tomorri, S. (Tirana)[2], Dobi, F. (Tirana)[2], Como, A. (Tirana)[3], Alikaj, V. (Tirana)[3]
[1]Université de Médecine Department des Neurosciences, University Service of Neurology, Tirana, Albania, [2]Université de Médecine Department des Neurosciences, University Service of Pediatry, Tirana, Albania, [3]Université de Médecine Department des Neurosciences, University Service of Psychiatry, Tirana, Albania

Aim: To investigate the prevalence and correlates of comorbid psychiatric disorders in adolescents with chronic daily headache (CDH).
Methods: We recruided 61 adolescents (age 14 - 19 y.o) with CDH, identified from Headache Specialist in Service of Neurology, UHC “Mother Teresa”. CDH subtypes were classified according to the most updated criteria in the International Classification of Headache Disorders, 2nd edition. A psychiatric interview was performed for each person by the psychiatrist for adolescents to assess depressive and anxiety disorders (Beck Inventory for Depression and Hamilton Scale for Anxiety/ Assessment for anxiety disorders).

Results: 58 subjects (18 M and 40 F) finished the psychiatric interview. 24 Subjects (47 %) had > 1 assessed psychiatric comorbidities with major depression (21 %) and panic disorder (19 %) as the most common.Presence of migraine was associated with psychiatric comorbidities (odds ratio (OR) = 3, 5, p = 0, 002).The association with the psychiatric disorders were stronger for migraine with aura than for migraine without aura.In contrast, CDH subtypes, headache frequencies or medication overuse were not correlated.
Conclusion: The community- based study showed high comorbidity of psychiatric disorders in adolescents with CDH. The presence of migraine attacks, especially migraine with aura, was the major predictor for these associations.
Keywords: Headache, psychiatric disorders.

P3.092

Optogenetic stimulation of orbitofrontal cortex alleviates compulsive behavior in *Sapap3* mutant mouse

Burguiere, E. (Paris)[1,2], Monteiro, P. (Cambridge)[2], Feng, G. (Cambridge)[2], Graybiel, A. (Cambridge)[2], Work accomplished in Ann Graybiel laboratory, current affiliation: Team Behavior, Emotion and Basal Ganglia (Dir. Luc Mallet)
[1]UPMC, Inserm UMR_S 975, CNRS UMR 7225, Pitié-Salpêtrière Hospital, Paris, France, [2]McGovern Institute for Brain Research and Department of Brain and Cognitive Sciences, Massachusetts Institute of Technology, Cambridge, United States

Obsessive-compulsive disorder (OCD) and related conditions are characterized by the expression of repetitive behaviors that can be triggered by external stimuli or obsessive thoughts. Here, we developed an optogenetic therapeutic approach to block repetitive, compulsive behavior in a mouse model in which deletion of the synaptic scaffolding gene, *Sapap3,* results in excessive grooming. We conditioned tone-evoked grooming by pairing a neutral stimulus (8 kHz tone) with water dripping onto the mouse's forehead, which provoked unconditioned grooming responses. *Sapap3* mutant mice and wildtype littermates both became conditioned, but whereas wildtypes tended to adapt their conditioned responses by inhibiting early responses to the tone and by grooming only at the time of water drop, the *Sapap3* mutants did not exhibit response inhibition and kept grooming compulsively at the tone. This deficit in response inhibition has been proposed to be related to dysfunctional orbitofrontal-striatal pathway in OCD patients. Similarly, *Sapap3* mutants are characterized by a deficient cortico-striatal neurotransmission. Thus, to compensate this dysfunctional connectivity we performed optogenetic stimulation of orbitofrontal-striatal pathway. When applied during performance of the task, we could selectively block the compulsive grooming responses of the *Sapap3* mutants. Extended out-of-task stimulation of orbitofrontal-striatal pathway also significantly decreased the spontaneous over-grooming behavior of the mutants. These findings demonstrate that selective optogenetic stimulation of the orbitofrontal-striatal pathway can alleviate compulsive behavior in the *Sapap3* model of OCD and suggest that this pathway might be critical for behavioral inhibition.

P3.093

Organelle and cellular abnormalities associated with hippocampal heterotopia in neonatal doublecortin knockout mice

Francis, F. (Paris)[1,2], Khalaf-Nazzal, R. (Paris)[1,2], Bruel-Jungerman, E. (Paris)[1,2], Rio, J.-P. (Paris)[1,2], Bureau, J. (Paris)[1,2], Sumia, I. (Paris)[1,2], Roumegous, A. (Paris)[1,2], Martin, E. (Paris)[3,4], Olaso, R. (Evry)[5], Irinopoulou, T. (Paris)[1,2], Parras, C. (Paris)[3,4], Cifuentes-Diaz, C. (Paris)[1,2]
[1]Institut du Fer à Moulin, Paris, France, [2]INSERM UMRS 839, UPMC, Paris, France, [3]UPMC-Paris 6, Centre de Recherche de l'Institut du Cerveau et de la Moelle Epinière, Paris, France, [4]INSERM UMRS 975, CNRS UMR 7225, Paris, France, [5]Centre National de Génotypage, CEA/DSV/IG, Evry, France

Heterotopic or aberrantly positioned cortical neurons are associated with epilepsy and intellectual disability. Various mouse models exist with forms of heterotopia, but the composition and state of cells developing in heterotopic bands has been little studied. *Dcx* knockout (KO) mice show hippocampal CA3 pyramidal cell lamination abnormalities, appearing from the age of E17.5, associated with spontaneous epilepsy. The *Dcx* KO CA3 region is divided into two layers, thus resembling heterotopia. Here, we further characterized the CA3 pyramidal cell abnormalities in these mice. Electron microscopy confirmed that the *Dcx* KO CA3 pyramidal cells layers at postnatal day (P) 0 are distinct and separated by an intermediate layer devoid of neuronal somata. Morphology, organization and cytoplasm content are altered in *Dcx* KO CA3 pyramidal neurons, compared to wild type (WT) cells. Less regular nuclei, with mitochondrial and Golgi apparatus differences were observed. Each *Dcx* KO CA3 layer at P0 was found to contain pyramidal neurons but also other closely apposed cells, displaying different morphologies. Immature radial glial (Pax6, Sox2) and astrocytic markers (GFAP) showed no specific labeling in either the WT or KO pyramidal cell layers at this age. Quantitative PCR and immunohistochemistry for Olig1 however, revealed increased numbers of oligodendrocyte precursor cells in close proximity to *Dcx* KO pyramidal cells. *In situ* hybridization experiments showed somatostatin (Sst)-positive interneurons more frequently interspersed with KO pyramidal cells. Immunohistochemistry experiments showed that caspase-3 dependent cell death was also increased in the hippocampus of *Dcx* KO mice at P2. These data hence provide a detailed characterization of the *Dcx* KO hippocampus in early postnatal stages and reveal ultrastructural abnormalities and cellular heterogeneity which may contribute to abnormal neuronal function and the development of hyperexcitability.

P3.094
Study of RNA splicing by RNAseq in Parkinson´s disease

Nkiliza, A. (Lille)[1], Hopes, L. (Lille)[2], Demay, C. (Lille)[3], Mutez, E. (Lille)[2], Leprêtre, F. (Lille)[3], Bleuse, S. (Lille)[4], Destée, A. (Lille)[2], Figeac, M. (Lille)[3], Chartier-Harlin, M.-C. (Lille)[1]
[1]Univ. Lille, INSERM U 837 - Centre JPArc - Equipe 6 (IRCL), Lille, France, [2]Univ. Lille / CHRU Lille, INSERM U 837 - Centre JPArc - Equipe 6 (IRCL), Lille, France, [3]Univ. Lille, Functional Genomic Platform, Lille, France, [4]CHRU Lille, Lille, France

Parkinson's disease (PD) is one of the most common movement disorder. It is characterized by the dopaminergic neurons degeneration in *Substantia nigra pars compacta*. This neurodegenerative process begins years before the clinical diagnosis of PD can be made and this diagnosis stays difficult in early stages of disease. Identification of biomarkers would thus be beneficial for reducing risk of misdiagnosis, but also for proposing therapeutic intervention earlier. A molecular change easy to measure and detectable in blood such as splicing variants might represent a reliable source of biomarkers. Indeed, several studies suggest that RNA splicing is disturbed in neurodegenerative disease like PD. We found that whole gene expression arrays analyses of familial and sporadic PD, sex and age-matched controls peripheral blood mononuclear cell sustained this hypothesis.
We thus decided to study RNA splicing by another transcriptome approach, called RNA sequencing (RNAseq). We analyzed a first group of 6 subjects (3 familial PD and 3 controls) by using Bioscope software to identify exon-exon junction showing the existence of 342 genes with putative new splicing events. 20.76% of these genes are involved in RNA processing and other in relevant biological processes to PD such as apoptosis or cell cycle. The validation of some of these new splicing events by polymerase chain reaction in a second group of sporadic PD patients and controls confirmed the existence of new splicing variants with a difference in signal intensity between subgroups underlining the putative quantitative variations. Moreover, the analysis of the first group by Cufflinks package confirmed the expression deregulation of the genes we tested. These data are consistent with the hypothesis that RNA splicing deregulation in patients with Parkinson can trigger the formation of new transcripts and/or expression deregulation of known transcripts.
Further studies in a larger cohort should enable the identification of more new RNA splicing events and/or expression variations of genes usable as biomarkers. Finally these analyses will be a way to characterize new transcripts of genes involved in this disorder.

P3.095

Expression pattern of PD-related genes and pathways is altered in the intestine of oral rotenone treated mice

Vanbesien-Mailliot, C. (Lille)[1,2,3], Pan-Montojo, F. (Dresden)[4,5], Stievenard, A. (Lille)[1,2], Bourgois, P. (Lille)[1,2], Comptdaer, T. (Lille)[1,2], Denning, Y. (Dresden)[4], Destée, A. (Lille)[1,2,6], Funk, R. (Dresden)[4,7], Chartier-Harlin, M.-C. (Lille)[1,2]

[1]Univ. Lille Nord de France, Lille, France, [2]UDSL (Lille 2), INSERM U 837 - Centre JPArc - Equipe 6, Lille, France, [3]USTL (Lille 1), Neurosciences, Villeneuve d'Ascq, France, [4]Institute for Anatomy, TU Dresden, Dresden, Germany, [5]University Hospital Carl-Gustav Carus, Neurology Dpt, Dresden, Germany, [6]CHRU Lille, Neurology Dpt, Lille, France, [7]Center for Regenerative Therapies Dresden (CRTD), Dresden, Germany

Parkinson's disease (PD) is a common neurodegenerative disorder of multifactorial etiology involving genetic and environmental factors. PD classical motor symptoms appear after a long silent period during which individuals progressively develop minor symptoms while molecular and cellular modifications concomitantly occur and spread within the brain. Non-motor symptoms such as gastro-intestinal dysfunctions also occur in early PD stages and might even precede them by several years. In idiopathic PD patients, pathological studies suggest that PD pathology progresses from the enteric nervous system (ENS) and the olfactory bulb into the brain following a spatiotemporal pattern. Recent researches showed that the local effect of rotenone on the ENS can mimic this pathology progression in mice. In order to identify possible early biomarkers for PD, we designed a study to investigate the effect of 5 mg/kg of oral administered rotenone on gene expression in the ileum. Intestine samples were collected after 2 and 4 months of 5 days/week intragastric rotenone exposure. Total RNA was extracted and hybridized on Agilent SurePrint G3 mouse microarrays and differentially regulated genes were identified using R software and the Bioconductor package Limma. Deregulated metabolic pathways were further investigated using Ingenuity Pathway Analysis© software. Indeed, our results show that exposure to rotenone induces alterations in gene expression profiles of the gastro-intestinal tract and we observe the highest deregulation after 2 months of rotenone gavage. Interestingly, the expression of several PD-related genes such as *snca*, *th* or *lrrk-2* is modulated by chronic exposure to rotenone. A deeper analysis of metabolic pathways in the ileum shows that, at this time-point, alterations affect the nervous system, cell-to-cell signalling, gastro-intestinal disorders and cancer-related pathways, while pathways related to cellular movements and inflammatory processes are clearly involved after 4 months of exposure to rotenone. These results indicate that pathway analyses are powerful tools to identify interesting candidates as potential early PD biomarkers and establish the basis for further investigations to delineate altered parameters in the early steps of the pathological process.

P3.096

Assays for anti-potassium channel complex autoantibodies in patients with limbic encephalitis

Moutot, N. (Marseille)[1], De San Feliciano, M. (Marseille)[1], Maulet, Y. (Marseille)[1], Da-Nzi Mboumba, L. (Marseille)[1], El Far, O. (Marseille)[1], Seagar, M. (Marseille)[1]

[1]Inserm UMR 1072, UNIS - Unité de Neurobiologie des canaux Ioniques et de la Synapse, Marseille, France

Autoimmune synaptic encephalopathies are disorders in which patients develop autoantibodies against synaptic proteins. Limbic encephalitis (LE) is one of these CNS diseases, characterized by psychiatric symptoms, memory loss and seizures. Sera from LE patients were previously found to contain autoantibodies that immunoprecipitate voltage-gated potassium channels (VGKC). However,

these antibodies appear to be principally directed against a distinct protein that associates with VGKC: Leucine-rich Glioma Inactivated-1 (LGI1). LGI1 is a glycoprotein secreted by neurons and interacts with the trans-synaptic scaffolding molecules ADAM22 and 23. Interestingly mutations of the human LGI1 gene underlie autosomal dominant lateral temporal lobe epilepsy and LGI1 -/- mice display frequent spontaneous seizures (Chabrol et al 2010). A large proportion of LE patients have antibodies that recognize LGI1 and immunoprecipitate Kv1 channels labeled with ^{125}I-dendrotoxin from detergent extracts of rat brain (Lai et al 2010, Irani et al 2010).
Diagnosis of autoimmune LE is crucial, as it is readily treatable by immunosuppression. As anti-Kv antibodies have been identified in patients with a larger spectrum of disorders, a novel serological assay to detect antibodies to LGI1 should be useful. Using recombinant LGI1, our laboratory has recently developed an ELISA for anti-LGI1 autoantibodies detection from patients sera. This assay adds to our routine serological assays for the detection of antibodies that immunoprecipitate ^{125}I-dendrotoxin labeled VGKC. The ELISA was validated with our collection of autoimmune sera and will be a valuable tool to optimize diagnostics for autoimmune LE with LGI1 autoantibodies.
Chabrol et al (2010) Brain 133: 2749-2762
Irani SR et al (2010) Brain 133: 2734-2748
Lai M et al. (2010) Lancet Neurol 9: 776-785

P3.097

A possible mechanism leading glutamate transporters dysfunction to seizures generation in developing rat brain

Aniksztejn, L. (Marseille)[1], Molinari, F. (Marseille)[2], Cattani, A. (Marseille)[2], Mdzomba, B. (Marseille)[2], Milh, M. (Marseille)[3]
[1]Faculté des Sciences de Luminy, INMED, INSERM U 901, Marseille, France, [2]INMED, INSERM U 901, Marseille, France, [3]Hôpital de la Timone, Service de neuropediatrie, Marseille, France

Cell surface glutamate transporters help to maintain glutamate homeostasis in the brain through the binding and transport of glutamate from extracellular to intracellular space. Neuronal and glial glutamate transporters are expressed in both human and rat brain during the fetal period suggesting that they contribute to proper development of the brain. In keeping with this, we have shown that their inhibition with Dl-TBOA generated seizures in freely moving rat pups. Interestingly the electrographic pattern was reminiscent to those encountered in certain form of early epileptic encephalopathy (Othahara syndrom; early myoclonic encephalopathy) since the activity was characterized by partial seizures and by an interictal activity called “suppression burst” and was not produced with other convulsive agents. These observations raised the possibility that dysfunction of glutamate transporters could represent a potential cause of these very severe desease. The fact that similarly to in vivo, NMDA dependent recurrent pattern of paroxysmal burst was produced by dl-TBOA in developing cortical slices (hippocampus, neocortex), enable us to study the cellular mechanism responsible of these pathological activity. We have shown that their initiation resulted from the combination of $[glutamate]_o$ elevation that tonically activated groups I/II metabotropic glutamate receptors (mGluRs), and the spillover of synaptically released glutamate that phasically activated extrasynaptic NMDARs. Tonic activation of mGluRs, through the cross talk with extrasynaptic NMDAR and the blockade of the slow after-hyperpolarisation current, leads the network to a hyper-excitable state. Both effects enable NMDAR, phasically activated, to generate and maintain a suprathreshold depolarization associated with a prolonged firing of action potentials. This would increase synaptic interactions between populations of cells and promote the genesis of paroxysmal burst in cortical networks. We propose that any process that decreases the efficiency of EAAT, including an alteration of glial glutamate catabolism or hypoxia would be sensed by mGluR and, through the same mechanism described here, shift cortical network to a hyperexcitable state thereby facilitating seizures generation.

P3.098

Impact of cancer therapies on cognitive functions, metabolism and neuro-vascular cell plasticity

Dubois, M. (Mont-Saint-Aignan)[1], Sabot, C. (Mont-Saint-Aignan)[2], Lecointre, C. (Mont-Saint-Aignan)[1], Le Joncour, V. (Mont-Saint-Aignan)[1], Chevalier, A. (Mont-Saint-Aignan)[2], Dautrey, S. (Mont-Saint-Aignan)[2], Gandolfo, P. (Mont-Saint-Aignan)[1], Joly, F. (Caen)[3], Romieu, A. (Mont-Saint-Aignan)[2], Hilber, P. (Mont-Saint-Aignan)[4], Renard, P.-Y. (Mont-Saint-Aignan)[2], Castel, H. (Mont-Saint-Aignan)[1]
[1]Inserm U 982, Laboratory of Neuronal and Neuroendocrine Communication and Differentiation (DC2N), Astrocyte and Vascular Niche, Institute of Research and Biomedical Innovation (IRIB), Normandy University PRES, University of Rouen, Mont-Saint-Aignan, France, [2]Laboratory COBRA UMR 6014 & FR 3038, IRCOF, Mont-Saint-Aignan, France, [3]Centre François Baclesse et Centre Hospitalier Universitaire, Caen, France, [4]EA 4700, Laboratory of Psychology and Neurosciences of Cognition and Affectivity (PSY-NCA), University of Rouen, Normandy University PRES, Mont-Saint-Aignan, France

Evidence is emerging that, in addition to “cancer” itself, chemotherapy and targeted therapy can induce brain damage leading to adverse effects such as fatigue and deficits of memory, attention and cognitive flexibility. The understanding of the mechanisms involved in these cognitive dysfunctions is crucial for prevention, taking care of patient quality of life and evaluation of new potential therapeutics and neuroprotectants.
Our project concerns the evaluation of direct long-term impact of cancer treatments on brain activities. Our first results revealed that young and aged non-bearing cancer mice treated with chemotherapy displayed selective executive functions impairments associated with decreased hippocampal cell proliferation. Co-administration of glucose protected mice against chemotherapy-induced cognitive dysfunctions and neurogenesis inhibition. *In vitro*, when glucose was co-administered, the observed quiescent state of neural stem cells cultured as neurospheres likely induced protection from neurotoxic effects of chemotherapy. Moreover, we showed that targeted therapy did not provoke alteration of cognitive performances evaluated in hippocampal and prefrontal cortex-dependent tasks. However, selective modifications of cerebral activity in brain areas involved in feeding behavior and sleep/wake cycle regulation, would suggest that hypothalamus represents a key target of this therapy. These alterations could likely contribute to symptoms such as weight loss and fatigue that have previously been reported in patients receiving targeted therapies. In order to follow pharmacokinetic in brain and ability to cross the blood brain barrier, we initiate the first synthesis of a fluorescently-labelled derivative of campthotecin (CPT-FI). Our results indicate that camptothecin-FI was captured by tumor cells and inhibited glioma growth and cell cycle. In nude mice xenografted with NIH-3T3 cells, CPT-FI was detected into the tumor core after 4-days intratumoral administration.
Thus, the development of the know-how in the design of anti-cancer molecules for brain imaging will allow understanding how systemic administration of cancer treatments might impact cerebral activity through specific brain entries and lead to non desired side effects.

P3.099

Urotensin II receptor endogenous ligands as new candidates involved in high grade glioma tumor growth and neo-angiogenesis

Le Joncour, V. (Mont Saint Aignan)[1], Lecointre, C. (Mont Saint Aignan)[1], Jarry, M. (Mont Saint Aignan)[1], Joubert, J.-E. (Mont Saint Aignan)[1], Shouft, M.-T. (Mont Saint Aignan)[1], Morin, F. (Mont Saint Aignan)[1], Modzelewski, R. (Rouen)[2], Véra, P. (Rouen)[2], Bohn, P. (Rouen)[2], Castel, H. (Mont Saint Aignan)[1], Gandolfo, P. (Mont Saint Aignan)[1]
[1]Inserm U 982, Laboratory of Neuronal and Neuroendocrine Communication and Differentiation (DC2N), Astrocyte and Vascular Niche, Mont Saint Aignan, France, [2]EA 4108, Laboratoire d'Informatique, de Traitement de l'Information et des Systèmes (LITIS), Rouen, France

Glioblastoma represent the most common primary brain tumors in adults. They are mainly characterized by an intense neo-angiogenesis, tumoral invasiveness of the normal brain tissue and a

high degree of recurrence. Vasoactive neuropeptides have been identified as potential regulators of recruitment and entrapping of diverse pro-angiogenic cells, promoting in situ neo-angiogenesis. Considered as the most potent vasoactive peptides, urotensin II (UII) and its paralog urotensin II-related peptide (URP) are involved in the control of endothelial cell proliferation and migration. Recent data obtained in our team indicate that UII, URP and their receptor UT are systematically expressed in human brain tumors samples, and that UII exerts potent chemotactic activities on several glioblastoma cell lines.
The aim of this study was to investigate *in vivo* the role of the urotensinergic system in glioma tumorogenesis. We first identified the pro-tumoral cell types chemoattracted by urotensinegic peptides by using engrafted plugs of a synthetic extracellular matrix containing human UII (hUII), mouse UII (mUII), shorter peptides $hUII_{4-11}$ or URP. We observed that hUII/mUII but not shorter peptides or URP, stimulated invasion of macrophages, endothelial cells and vascular smooth muscle cells. We next investigated the growth of human glioblastoma xenografted in Nude mice during intratumoral treatments with hUII or URP and/or two different UT antagonists. Administration of hUII accelerated glioma tumor development and promoted neo-angiogenesis. Interestingly, exposure to URP or the peptidic UT antagonist urantide reduced tumor growth, inhibited angiogenesis and significantly prolonged mice survival. Through the development and validation of a technetium tracer (99mTC-RGD) targeting αvβ3 integrins expressed by the endothelial wall of newly formed vessels and on the plasma membranes of some tumors, we confirmed by SPECT imaging, the inhibition of the neo-vascularization of urantide-treated tumors. Altogether, these data suggest that hUII favors pro-angiogenic cell type recruitment. Thus, specific ligands for UT including antagonists may constitute an original strategy to block both tumor cell invasion and neo-angiogenesis in glioma development.

P3.100

HSV-derived viral vector encoding a dominant-negative RAGE reduces astroglial response to intermittent hypoxia-induced injury

Angelo, M.F. (Buenos Aires)[1], Aguirre, A.I. (Buenos Aires)[2,3], Lukin, J. (Buenos Aires)[4], Melendez, M.E. (Lyon)[5,6], Villarreal, A. (Buenos Aires)[1], Epstein, A. (Lyon)[6,7], Jerusalinsky, D.A. (Buenos Aires)[2,3], Ramos, A.J. (Buenos Aires)[4]
[1]Instituto de Biología Celular y Neurociencias Prof. E. De Robertis, Laboratorio de Neuropatología Molecular, Buenos Aires, Argentina, [2]Instituto de Biología Celular y Neurociencias Prof. E. De Robertis, Laboratorio de Neuroplasticidad y Neurotoxinas, Buenos Aires, Argentina, [3]LIA-Devenir, Buenos Aires, Argentina, [4]instituto de Biología Celular y Neurociencias Prof. E. De Robertis, Laboratorio de Neuropatología Molecular, Buenos Aires, Argentina, [5]Centre de Génétique et de Physiologie Moléculaire et Cellulaire, Lyon, France, [6]LIA-Devenir, Lyon, France, [7]Centre de Génétique et de Physiologie Moléculaire et Cellulaire, Lyon, Argentina

Glial cells play a crucial role in CNS injury since neurodegenerative processes are associated with neuroinflammation, involving microglial cells and subsequent activation of astrocytes. Glial activation involves changes in cell phenotype and gene expression that might trigger neuronal death. Using Intermittent Hypoxia (IH) to simulate Sleep Apnea as a model of CNS injury, we have previously demonstrated early reactive gliosis and neuronal alterations that ranged from decreased dendrite length and abnormal NeuN staining in hippocampus and brain cortex. We also demonstrated that IH induces the overexpression of the Receptor for Advanced Glycation End Products (RAGE) and its ligand S100B as well as the activation of the downstream NF-kB signaling. Mixed culture (glia and neurons) exposed to IH also showed astrocyte stellation (the *in vitro* version of reactive gliosis), reduction in neurite length and NF-kB activation.
To modulate inflammation we developed HSV-derived amplicon to overexpress RAGE (RAGE-FL) or its dominant negative (RAGE-Δcyt). *In vitro*, astrocytic stellation was prevented by the overexpression of RAGE-Δcyt. A similar effect was observed with RAGE-blocking antibodies but not with unrelated control IgG. Indeed, RAGE blockage has also reduced neuronal degeneration *in vivo* and *in vitro* after IH. In *naïve* (non-hypoxic) animals, overexpression of the amplicon-delivered RAGE-FL, but not RAGE-Δcyt, resulted in alterations of neuronal morphology similar to those observed in hypoxic animals. In IH exposed animals RAGE-Δcyt, but not RAGE-FL overexpression prevented abnormal NeuN distribution which is the first step in the events leading to neuronal degeneration.

These results suggest that using viral vectors to block RAGE depending pathways action may represent a new promising tool to diminish inflammation and reactive gliosis in the injured brain.
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P3.101

Deletion of the major myelin protein, PLP, lead not only to motor but also cognitive defects

Begou, M. (Paris)[1,2], Depiets, B. (Paris)[1,2], Giraudet, F. (Clermont-Ferrand)[2,3], Avan, P. (Clermont-Ferrand)[2,3], Bechon, C. (Paris)[1,2], Gueret-Gonthier, C. (Paris)[1,2], Vaurs-Barriere, C. (Clermont-Ferrand)[4], Boespflug-Tanguy, O. (Paris)[1,5]
[1]INSERM 676, Hôpital Robert Debré, Université Paris-Diderot, Paris, France, [2]Laboratoire de biophysique, Faculté de médecine, Université d'Auvergne, Clermont-Ferrand, France, [3]INSERM 1107 NEURO-DOL, Clermont-Ferrand, France, [4]INSERM 1103 - CNRS 6293, GReD, Clermont-Ferrand, France, [5]CHU Robert Debré, Reference Center for 'Leukodystrophies', Paris, France

Deletion or truncating mutation in the proteolipoprotein 1 gene (*PLP1*) cause a mild form of X-linked dysmyelinating disorders, the type 2 spastic paraplegia (SPG2). While motor signs with ataxia and spasticity are the classical manifestations, cognitive disturbances have been reported but are not well characterized. Mice with deletion of the PLP gene (Plp null mice) have been developped to model the human disease.
To further investigate the cognitive and behavioral abnormalities related to PLP null expression, we initiated a longitudinal study in Plp null mice on motor and cognitive behaviors. Auditory brainstem responses were measured to assess nerve conduction within the early auditory system and constitute an index of axonal function and myelination *in vivo*. We then performed a gene expression assay and an immunohistological analysis on Plp null brain to identify cellular substrates of the identified behavioral abnormalities.
We report that Plp null mice exhibit motor dysfunction which progressively detoriate with age. These mice also exhibit working memory impairement and anxiety disturbance. The gene expression assay reveals (1) important modifications of mRNA involved in the neurogenesis in 15-days-old Plp null mice; (2) modifications in some neurotransmitter systems. The immunohistological study shows a decrease in GFAP and calbindin expression in defined hippocampus areas.
We have identified a link between PLP deletion and the presence of cognitive defects in human that are recapitulated in our mouse model. Furthermore we showed that axonal degeneration and progressive demyelination could be associated with the development of such cognitive behaviors. Given the growing interest in determining the white matter implication in cognitive function, our data unveil a new model for further understanding this implication.

P3.102

Alteration of serotoninergic system is associated with respiratory alterations in rodent models of SUDEP

Kouchi, H.Y. (Lyon)[1], Georges, B. (Lyon)[1], Dieuset, G. (Rennes)[2], Bonnet, C. (Lyon)[1], Ogier, M. (Brétigny-sur-Orge)[3], Belmeguenai, A. (Lyon)[1], Martin, B. (Rennes)[2], Ryvlin, P. (Lyon)[1], Morales, A. (Lyon)[1], Bezin, L. (Lyon)[1]
[1]CRNL - Centre de Recherche en Neurosciences de Lyon-Equipe TIGER, Inserm U 1028 & CNRS UMR 5292, Université Claude Bernard Lyon 1, Lyon, France, [2]Laboratoire Traitement du Signal et de l'Image (LTSI)-UMR 1099, Rennes, France, [3]IRBA Institut de Recherche Biomédiale des Armées, Brétigny-sur-Orge, France

Fatal respiratory arrest has been proposed as a cause of Sudden Unexpected Death in Epileptic Patients (SUDEP), and may result from altered brainstem serotonin (5-HT) system, that remain to be better characterized in pertinent animal models. On one hand, DBA/2 mice can develop fatal respiratory arrest (fRAr) following audiogenic-induced seizures, and, on the other hand, epileptic rats following pilocarpine-induced *status epilepticus* (Pilo-SE) can develop respiratory alterations (RAl). Since DBA/2 mice are not "epileptic" mice, exhibiting spontaneous recurrent seizures (SRS), and epileptic rats are not subjected to fRAr after SRS, we hypothesized that alterations of 5-HT system that might be the most relevant to SUDEP should be shared by both animal models. We thus compared brainstem alterations in transcript levels of 5-HT system proteins between:
1) DBA/2 mice exhibiting fRAr or not (RAr);
2) epileptic rats with RAl (RAl+) or not (RAl-), at both onset of RAl+ (5 weeks after Pilo-SE) and 13 weeks after Pilo-SE.
We found that transcripts of tryptophan hydroxylase 2 (Tph2), 5-HT transporter (SERT) and 5-HT2C receptor are the most abundantly expressed in mouse and rat brainstem. By contrast, 5-HT1A and 5-HT2A are expressed at moderate levels. FRAr mice and RAl+ rats at 5 weeks after Pilo-SE had greater levels of all transcripts measured than that of RAr mice and RAl- rats, respectively. Only 5-HT1A receptor was not altered in RAl+ rats compared to RAl- rats. At 13 weeks after Pilo-SE, Tph2, SERT and 5-HT2A receptor transcript levels in RAl+ rats recovered those of RAl- rats. By contrast, transcript level of 5-HT2C receptor was maintained at a greater value in RAl+ rats compared to RAl- rats. In conclusion, our study suggests that:
1) higher expression of 5-HT1A receptor in DBA/2 mouse brainstem might be a vulnerability factor to fatal respiratory arrest induced by seizures since its expression remained constant in epileptic rats with respiratory alterations known to be resistant to fatal respiratory arrest;
2) maintained respiratory alterations in epileptic rats are associated with high expression of 5-HT2C receptor, that might be interesting to target pharmacologically due to its important role in the central regulation of respiratory function.

P3.103

Clinical and polysomnographic characteristics in 117 children with narcolepsy

Inocente, C.O. (Lyon)[1,2], Lavault, S. (Paris)[3,4], Dauvilliers, Y. (Montpellier)[5,6], Lecendreux, M. (Paris)[3,7], Reimão, R. (São Paulo)[2], Lin, J.-S. (Lyon)[8], Arnulf, I. (Paris)[3,4], Franco, P. (Lyon)[3,8,9]
[1]Integrative Physiology of Brain Arousal System, CRNL, INSERM U1028, CNRS UMR 5292, University Lyon 1, Lyon, France, [2]Sleep Medicine Advanced Research Group, Division of Clinical Neurology, Clinical Hospital, University of São Paulo School of Medicine, São Paulo, Brazil, [3]National Reference Centre for Orphan Diseases, Narcolepsy, Idiopathic Hypersomnia and Kleine-Levin Syndrome (CNR Narcolepsie-Hypersomnie), Paris, France, [4]AP-HP, Groupe Hospitalier Pitié-Salpêtrière, Unité des Pathologies du Sommeil & Université Pierre et Marie Curie - Paris 6, Centre de Recherche de l'Institut du Cerveau et de la Moelle épinière, UMR-S 975, CNRS UMR 7225 & Inserm, U 975, Paris, France, [5]National Reference Centre for Orphan Diseases, Narcolepsy, Idiopathic Hypersomnia and Kleine-Levin Syndrome (CNR Narcolepsie-Hypersomnie), Montpellier, France, [6]Inserm U 888, Service de Neurologie B, Hôpital Gui-de-Chauliac, CHU de Montpellier, Montpellier, France, [7]Centre pédiatrique des pathologies du sommeil, Hôpital Robert Debré, Paris, France, [8]Integrative Physiology of Brain Arousal System, CRNL, INSERM U 1028, CNRS UMR 5292, University Lyon1, Lyon, France, [9]Pediatric Sleep Unit, Hôpital Femme Mère Enfant, University Lyon1, Lyon, France

To report the clinical and polysomnographic characteristics in a narcoleptic children and adolescents evaluated in the National French Multicentric Research program on narcolepsy (NarcoBANK) from 2008-2011. The children received the diagnosis of narcolepsy after a complete evaluation including questionnaires (Epworth, Children´s Depression Inventory (CDI), Quality of Life (QOL), a PSG followed by a multiple sleep latency test (MSLT), HLA typing and Hcrt-1 level in CSF in some cases. Wilcoxon non-matched pairs signed-ranks test and Spearman test were used. The cohort included 117 children (65 boys) with a mean age of 11.6± 3.1 years at diagnosis (41.8% < 10 years). The first symptom at onset was excessive daytime sleepiness (9.25±3.8 years). Other symptoms were

cataplexy (80%), hypnagogic hallucinations (40%), sleep paralysis (24.7%), insomnia (16.2%), parasomnia (74.3%) and night eating (12.8%). 91.3% were DQB1*0602 positive. The hypocretin-1 level (n=31) was 31±46 pg/mL. Mean BMI was 23.2±5.2 kg/m2, 59.8% were obese, Z-score was 2.92±2.63. Eight out of 31 girls (26%) had precocious puberty (62.5% obese). Thirty patients (11.1%) received H1N1 vaccination prior the onset of the symptoms, 6.8% had a familial narcolepsy. 21% had signs of depression on the CDI score. There was a positive correlation between CDI and Epworth (r=0.40, p=.001) but not Z-score. 36.7% of the children had school difficulties, 23.6% repeated a year and 26.5% had absenteeism. The QOL for children, adolescents, parents were correlated to CDI score (r=-0.48 p=.008, r=-0.78 p< .001, r=-0.57 p< .001) but not to Epworth or Z-score. The mean total sleep time was 471.4±85 min, sleep efficiency 81.8±16.1%, % N3 27.6±11.2, %REM 19.8±7, AHI 2.2±3.6/h, arousal index 7.6±7.5/h. The sleep latency was 16.8±24.6 min and the REM latency on PSG was at 88.5±86.2 min. The sleep latency on MSLT was 5.5±4.6 min with 3.3±1.3 SOREM. No correlation was found between sleep onset latency on MSLT and sleep efficiency, apnea-hypopnea index, respiratory related arousals, CDI and QOL. In this large cohort study, narcolepsy in children was associated with obesity and precocious puberty in respectively 60% and 25% of the patients. This disease had a severe impact on mood and school performances.

P3.104

Examining transcranial direct-current stimulation (tDCS) as a treatment for hallucinations in schizophrenia

Suaud Chagny, M.F. (Bron)[1], Brunelin, J. (Bron)[1], Mondino, M. (Bron)[1], Gassab, L. (Monastir)[2], Haesebaert, F. (Bron)[1], Gaha, L. (Monastir)[2], Saoud, M. (Bron)[1], Mechri, A. (Monastir)[2], Poulet, E. (Bron)[1]
[1]UCBL/Hopital du Vinatier, EA 4615, Bron, France, [2]Centre Hospitalo-Universitaire Fattouma Bourguiba, Monastir, Tunisia

Some 25%-30% of patients with schizophrenia have auditory verbal hallucinations that are refractory to antipsychotic drugs. Neuroimaging studies have implicated hyperactivity in the left temporoparietal cortex during auditory hallucinations, and hypoactivity in the prefrontal cortex in schizophrenia, including negative symptoms.
Here we hypothesise that transcranial direct current stimulation (tDCS) by acting antagonistically on these two distinct brain areas involved in the pathophysiology of schizophrenia could reduce the severity of refractory symptoms. tDCS is a method of non-invasive brain stimulation which uses weak electric currents applied on the scalp to modulate activity of underlying brain tissue. With tDCS, the cortical neuronal excitability is increased in the vicinity of the anode and is reduced near the cathode.
Thirty patients with schizophrenia and medication-refractory auditory verbal hallucinations were randomly allocated to receive 20 minutes of active 2-mA tDCS or sham stimulation twice a day on 5 consecutive weekdays. The anode was placed over the left dorsolateral prefrontal cortex and the cathode over the left temporo-parietal cortex.
Auditory verbal hallucinations were robustly reduced by tDCS relative to sham stimulation, with a mean diminution of 31%. The beneficial effect on hallucinations lasted for up to 3 months. Moreover, compared with sham treatment, active tDCS induced a significant effect on negative dimension. Positive and depressive dimensions showed medium effect sizes, while no effect was observed on the dimensions of disorganization and grandiosity/excitement.
Our results suggest that tDCS, an easy-to-use, low-cost tool with few side effects, could constitute a new tool in the treatment of refractory symptoms of schizophrenia. Further studies with larger samples and additional evaluations, such as functional evaluations and imaging, are needed to confirm these promising results.

P3.105

Serotonin regulates hippocampal synaptic plasticity and object memory in mice

Fernandez, S. (Paris)[1], Gruart, A. (Seville)[2], Delgado Garcia, J.M. (Seville)[2], Gaspar, P. (Paris)[1]
[1]Institut du Fer a Moulin, INSERM U 839, Paris, France, [2]Pablo Olavide University, Seville, Spain

Low levels of serotonin (5-HT) have been associated with the learning and memory deficits seen in Alzheimer's disease, autism and major depression. Studies performed in healthy volunteers have shown that 5-HT depletion disrupts consolidation of new information, specifically in tasks involving delayed recall and/or recognition of visually-presented words, spoken words, pictures or abstract figures. Despite this evidence, the mechanism by which 5-HT regulates learning and memory function is not clear.
We used a genetic model of 5-HT depletion, Pet1 knock-out mice (Pet1 KO), to understand the role that 5-HT serves in modulating learning and memory function. In Pet1 KO mice, the levels of 5-HT are highly decreased, specifically associative cortical areas and the hippocampal formation are completely depleted of 5-HT axons. Our results showed that Pet1 KO mice are capable of acquiring associative fear conditioning, reward reinforced associative learning and motor learning; however, they do not recall familiarization with objects. The novel object recognition test was used to measure hippocampal-dependent declarative memory. The animals are trained to familiarize with two identical objects, and then subjected to a test session where one object is replaced by a novel one. This observation strongly linked object recognition deficit with 5-HT depletion; however does not specify the underlying mechanism. Previously it was shown that the consolidation of object memory involves the enhancement of synaptic strength across the CA3-CA1 hippocampal connexion; a phenomenon called as long-term potentiation (LTP). To test the hypothesis that LTP deficit is responsible for the memory impairment observed in Pet1KO mice, we performed in vivo recordings of field potentials in the CA1 region of the hippocampus of mice while performing the object memory task. We found that in Pet1 KO, experience-dependent LTP is exaggerated compared to wild-type mice, possibly explaining the aberration seen in forming correct maps of the object presented. These results established for the first time a direct link between central 5-HT depletion, memory impairment and synaptic plasticity deficits.

P3.106

Ataxia with cerebellar lesions in mice expressing chimeric PrP-Dpl protein

Lemaire Vieille, C. (Grenoble)[1], Bailly, Y. (Strasbourg)[2], Brocard, J. (Grenoble)[3], Erlich, P. (Grenoble)[1], Loeuillet, C. (Grenoble)[1], Haeberlé, A.-M. (Strasbourg)[2], Rak, C. (Grenoble)[1], Demais, V. (Strasbourg)[2], Dumestre Pérard, C. (Grenoble)[1], Gagnon, J. (Grenoble)[1], Cesbron, J.-Y. (Grenoble)[1]
[1]Laboratoire adaptation et pathogénie des microorganismes CNRS-UMR 5163 UJF, Grenoble, France, [2]Institut des Neurosciences Cellulaires et Intégratives CNRS UPR 3212, Cytologie et cytopathologie neuronales, Strasbourg, France, [3]Grenoble - Institut des Neurosciences CR Inserm U 836, Grenoble, France

Prion diseases represent a group of singular transmissible and fatal neurodegenerative disorders characterized by the transition of the cellular prion protein (PrP^C) into an aberrant protein conformer, PrP^{Sc}. This form of PrP is the hallmark of prion infection. However, many experiments have shown that PrP^{Sc} may not be the proximate cause of neuronal dysfunction and neurodegeneration. Toxicity and infectivity in prion diseases are distinct properties of PrP that do not necessary coincide. Mutations within the central region of PrP have been shown to be associated with severe neurotoxic activity similar to that observed with Dpl, a PrP like protein. To further investigate this neurotoxic effect, we generated lines of transgenic (Tg) mice expressing three different chimeric PrP-Dpl proteins. Chi1 (aa 1-57 of Dpl replaced by aa 1-125 of PrP) and Chi2 (aa 1-66 of Dpl replaced by aa 1-134 of PrP) abrogated the pathogenicity of Dpl indicating that the presence of a N-terminal domain of PrP (23-134) reduced the toxicity of Dpl, as reported. However when the aa 1-24 of Dpl were replaced by aa 1-124 of PrP, Chi3 Tg mice, that express the chimeric protein at a very low level, start developing ataxia at the age of 5-7 wk. This phenotype was not counteracted by a single copy of full length-PrP^c but rather by its overexpression, indicating the strong toxicity of the chimeric protein Chi3. Chi3 Tg mice exhibit

severe cerebellar atrophy with a significant loss of granule cells. We concluded that the first 33 aa of Dpl, that are not present in Chi1 and Chi2 constructs, confers toxicity to the protein. We tested this possibility by using the 25-57 Dpl peptide in primary culture of mouse embryo cortical neurons, and found a significant neurotoxic effect. This finding identifies a protein domain that plays a role in mediating Dpl-related toxicity. This data could provide insights into understanding the molecular mechanisms involved in the pathogenesis of prion disease and more globally of neurodegenerative disease.

P3.107

Reward- and effort-related neuronal activity in the subthalamic nucleus of Parkinson's disease patients

Eusebio, A. (Marseille)[1,2], Duclos, Y. (Marseille)[3], Regis, J. (Marseille)[1], Azulay, J.-P. (Marseille)[1], Zenon, A. (Bruxelles)[4]
[1]CHU La Timone, Marseille, France, [2]Institut de Neuroscience de la Timone (INT), Marseille, France, [3]Institut de Neuroscience de la Timone, Marseille, France, [4]Universite Catholique de Louvain, Bruxelles, Belgium

In Parkinson Disease (PD), there is evidence suggesting an inadequate evaluation of the cost-benefit ratio of actions ultimately leading to a suboptimal allocation of resources. In addition there is evidence indicating that the Subthalamic Nucleus (STN) is involved in motivated behaviours and in reward-related processes.
Starting from these considerations, we tried to determine the extent to which movement vigor and the variables that are critical to its computation - in particular reward - can be identified in the neural activity of basal ganglia. We also evaluated how the dopamine level affects the representation of these variables and how these changes relate to the behavioral deficits in PD.
To address these issues, we recorded local field potentials (LFP) in the STN of 5 PD patients having benefited from the implantation of deep brain stimulation (DBS) electrodes. During these recordings, the patients were asked to perform a task requiring to squeeze a dynamometer with variable force and with a promise of a variable virtual monetary reward. In each trial, the patients were allowed to choose to either perform or skip the effort proposed as a function of effort intensity and reward amount. These two pieces of information were provided by visual cues at the beginning of each trial. Each patient was tested twice, ON and OFF dopamine replacement therapy.
Preliminary results indicate that robust responses to the different visual cues are present in STN. In some cases, these responses, mostly confined to low frequency ranges (< 10Hz), are proportional to the reward and effort intensity. We also found activities in higher frequency ranges (>55 Hz) during the effort exertion and these activities were most affected by the dopamine replacement therapy.
In conclusion, our preliminary data indicate that signals relevant to motivation control and effort-based decision making can be identified in STN. It remains to be determined whether these signals are predictive of actual motivational adjustments or cost-benefit decisions.

P3.108

MPTP treatment in the cat reproduces some aspects of the precocious sleep/wake cycle disturbances and late locomotor symptoms of human Parkinson´s disease

Akaoka, H. (Lyon)[1], Cendres-Bozzi, C. (Lyon)[2], Buda, C. (Lyon)[2], Sastre, J.-P. (Lyon)[2], Lin, J.-S. (Lyon)[2]

[1]CRNL, INSERM U 1028 - CNRS UMR 5292, Université Lyon1, Integragrative Physiology of Brain Arousal Systems Group, Lyon, France, [2]CRNL, INSERM U 1028 - CNRS UMR 5292, Université Lyon1, Lyon, France

Sleep disorders, e.g., excessive daytime sleepiness or narcoleptic attack, are frequently reported in human Parkinson's disease (PD). These perturbations constitute major predicaments in everyday life, and these disorders may result from dopaminergic (DA) deficiency, nocturnal motor disability or deleterious effect of anti-PD drugs. In cats treated with MPTP, a neurotoxin causing selective DA neuronal loss, we have studied the functional link between the sleep-wake cycle disturbances induced by MPTP, motor activity and DA cell loss.
MPTP-treatment (5mg/kg/day x 5, i.p.) caused, during the acute period (10 days), a severe hypersomnia in slow wave sleep (SWS, up to 80% of recorded time) and a suppression of paradoxical sleep (PS). These animals exhibited an almost complete inactivity (as assessed by Actiwatch) and sluggish initiation of movements if any. SWS hypersomnia, but not PS suppression, was improved by administration of low doses of direct or indirect DA agonists (L-dopa, ropinirole). These agonists largely restored the behavioural activity, with almost normal locomotor activity. During the chronic period (3rd-4thweek post-treatment), MPTP-treated animals have apparently achieved a complete motor recuperation, while the following sleep/arousal perturbations were observed: an overall PS increase (30-50%), prolonged PS episode duration and narcolepsy-like episodes. DA agonists failed to affect the locomotor activity during such chronic period. Substantial DA cell and fiber loss was verified by ex-vivo immunohistochemistry of tyrosine-hydroxylase, the marker of catecholaminergic neurons, in the striatum and substantia nigra.
Thus, MPTP-treatment produces in cats the major signs of motor and sleep-wake disorders of human PD, establishing MPTP treated cats as a useful model for PD. In particular, our results underscore the importance of studying from the beginning of MPTP treatment, as the acute period was the only period during which a conspicuous locomotor effect was observed for DA agonists, thereby reproducing the major clinical feature observed in the human. Conversely, the chronic period is most suited to study the sleep disturbances observed during preclinical phase of PD, when the main clinical motor symptoms are efficiently prevented to appear.

P3.109

Maladaptive response to stress: a high resolution structural 7T MRI study in rats

Bourgin, J. (Paris)[1,2,3], Mériaux, S. (Gif sur Yvette)[4], Boumezbeur, F. (Gif sur Yvette)[4], Djemaï, B. (Gif sur Yvette)[4], Poupon, C. (Gif sur Yvette)[4], Bottlaender, M. (Gif sur Yvette)[4], Jay, T. (Paris)[1,3], Cachia, A. (Paris)[1,3,5]
[1]INSERM U 894 Centre de Psychiatrie & Neurosciences, Paris, France, [2]Centre Hospitalier Sainte-Anne, Service Hospitalo-Universitaire, Paris, France, [3]Université Paris Descartes, Sorbonne Paris Cité, Paris, France, [4]Neurospin, I2BM, CEA, Gif sur Yvette, France, [5]CNRS U 6232, Laboratoire de Psychologie du Développement et de l'Education de l'Enfant, Paris, France

Affective disorders result from gene-environment interactions with stress as a major risk factor. Studies in humans have shown that chronic stress and related glucocorticoid elevation can damage the brain. While most people are resilient to such effects, in some subjects, chronic stress triggers affective disorders. Such maladaptive response to stress has been associated with susceptibility to the development of affective disorders.
The aim of this brain imaging study in rats was to investigate the anatomical brain changes associated to stress in two rat strains with normal (Sprague Dawley; SD, N=12) and pathological (Fisher 344; F, N=15) response to stress. We compared two rat strains at baseline and after 15 days of chronic stress exposure (repeated inescapable stress on an elevated and unsteady platform for 30 minutes). At baseline or immediately after the stress procedure, rats were anaesthetized, killed, their brains were fixed with paraformaldehyde and placed in Fluorinert. Blood samples were collected from the tail 10 min after anesthesia. Rat brain anatomy was quantitatively assessed using ex vivo Magnetic Resonance Imaging (MRI) and parametric T2* maps acquired on a Bruker PharmaScan 7T with a 3D MGE sequence (acquisition time = 34 h), yielding a high isotropic spatial resolution of 80x80x80 µm.

Brain regions *a priori* vulnerable to stress - dorsal and ventral hippocampus, medial and lateral prefrontal cortex and amygdala - were manually delineated from MRI slices with Brainvisa software. As expected, we found a differential stress effect between the two strains with a corticosterone plasma level increase in F compared to SD rats after chronic stress exposure. A significant lower baseline volume was found in F (N=7) compared to SD (N=6) in the dorsal hippocampus. After chronic stress, a significant volume reduction in F (N=8) compared to SD (N=6) was found in the dorsal and ventral hippocampus.
Our results suggest that the hippocampus is a marker of stress vulnerability and its volume derived from structural MRI may be used as a marker of maladaptive response to stress. Such findings show the potency of structural MRI in preclinical research with translational benefit to and from the clinic.

P3.110

Can inactivation of the subthalamic nucleus help treating alcoholism?

Pelloux, Y. (Marseille)[1], Baunez, C. (Marseille)[1]
[1]*Institut de Neuroscience de la Timone (INT), Marseille, France*

The present study aimed to assess the effect of inactivating the subthalamic nucleus (STN) on diagnostic criteria for alcoholism i.e.
1) loss of control over alcohol intake,
2) alcohol use at the expense of other rewards and
3) despite adverse consequences (punishment).
We developed a model evidencing after the loss of control over alcohol intake, a preference for the substance over non addictive reward in some vulnerable animals. Preliminary data showed that the lesion of the STN tended to prevent the loss of control over alcohol intake. Ongoing experiments are assessing the ability of STN lesion
1) to restore the control over alcohol intake of rats having previously lost it and
2) to reallocate alcohol related behaviors towards alternative natural rewards. After having controlled that STN lesion leaves the memory for aversive event unaffected,
3) further studies are also currently assessing its ability to reduce alcohol use in face of punishment.
These results should validate the pertinence of targeting the STN against alcoholism.

P3.111

Expression of intellectual disability genes, circadian rhythm and Ophn1

Dorseuil, O. (Paris)[1], Renaud, J. (Paris)[1], Dumont, F. (Paris)[1], Jacques, S. (Paris)[1], Challet, E. (Strasbourg)[2], Chelly, J. (Paris)[1], Billuart, P. (Paris)[1]
[1]*INSERM U 1016, Institut Cochin, Paris, France,* [2]*CNRS UPR 3212, Institut des Neurosciences Cellulaires et Intégratives, Strasbourg, France*

Children with intellectual disability (ID) or autism spectrum disorders (ASD), tend to have modified sleep cycles. As synaptopathy is considered as the main hypothesis for pathophysiology of ID, it suggests that synaptic activity participates to circadian clock regulation. Reciprocally, circadian rhythm governs many aspects of behavior and physiology, including sleep/wake phases. Interestingly, several synaptic proteins exhibit circadian expression, suggesting a circadian regulation of synaptic activity by the master clock. Here we have investigated in rodent, the apparent interplay between circadian clock and synapse activity in hippocampus of wild type animals and also studied, in a mouse model of ID, the functional consequences of synaptic plasticity alterations on circadian activity. Transcriptomic analysis of gene expression in wild type mouse hippocampus, over a 24h period, allowed us to define, in this brain structure associated with learning defects, a list of “circadian genes”. Pathway analysis on

gene functions show that not only clock genes follow a circadian expression, but also the ubiquitination and glutamate receptor signalings. Surprisingly, ID/ASD and synaptic genes do not appear enriched in our circadian gene list, thus not supporting a circadian dysregulation in hippocampus as a major pathogenic mechanism of ID/ASD. We recently reported that Oligophrenin1 (Ophn1), an X-linked ID gene product regulating synaptic activity through glutamate receptor internalization, interacts with Nr1d1, a transcriptional repressor of the master clock. To search for circadian dysregulations in Ophn1-KO mice, we compared gene expression patterns of WT and KO in the hippocampus over a 24h period. We report here, slight dysregulations of numerous circadian-regulated genes, including some master clock genes, as well as genes of the protein ubiquitination pathway, glucocorticoid receptor signaling and regulation of transcription. Promoter analysis of the dysregulated circadian genes shows enrichment in Nr1d1-target genes, thus highlighting the role of "Ophn1-synaptic activity-Nr1d1" link in pathophysiology of the disease. Altogether this work showed minor transcriptional modifications during circadian rhythm in agreement with no modification in circadian behavior.

P3.112

Ketamine anesthesia following long-lasting *status epilepticus* in juvenile rats counteracts brain inflammation response, epileptogenesis and cognitive impairment

Bezin, L. (Villeurbanne)[1], Belmeguenai, A. (Villeurbanne)[1], Georges, B. (Villeurbanne)[1], Bonnet, C. (Villeurbanne)[1], Dournaud, P. (Paris)[2], Auvin, S. (Paris)[2], Ryvlin, P. (Villeurbanne)[1], Bodennec, J. (Villeurbanne)[1]
[1]Université Lyon 1, Centre de Recherche en Neurosciences de Lyon, CNRS UMR 5292 - Inserm U 1028, Villeurbanne, France, [2]INSERM U 676, Paris, France

Brain inflammation following severe *status epilepticus* (SE) is suggested to play a crucial role in epileptogenesis. In juvenile rats, SE triggered by 3 mEq/kg lithium and 25 mg/kg pilocarpine is usually stopped, 30 min after its onset, by 10 mg/kg diazepam. In reality, diazepam at that time is crucial to increase survival rate, but rats are still seizing and sedation is only achieved when a second diazepam administration is given at 5 mg/kg 2h after the onset of SE. In this model, rats exhibit a severe inflammatory response (induction of IL-1β, TNF-α, IL-6, MCP-1, MIP-1α, MIP-1β genes) in several forebrain areas. In this study, we tested whether ketamine administration, instead of diazepam, could improve the outcome of rats following SE. At the dose of 100 mg/kg given 30 min after the onset of SE, supplemented with the same dose 40 min later to maintain anesthesia, we observed a total blockade of SE. In comparison with diazepam treatment, ketamine-induced anesthesia dramatically reduced brain inflammation and counteracted epileptogenesis, anxiety-like behavior and cognitive impairment (spatial learning). Because this protection could be attributed to the reduction of SE duration *per se*, we empirically determined a ketamine administration protocol (so-called Ket4X) making it possible to increase survival rate and maintain SE for a period of 2h, as in the protocol using diazepam. Ketamine was thus administered twice (at 10 mg/kg 30 min and 70 min after the onset of SE), followed by two injections of ketamine at 100 mg/kg and 50 mg/kg 2h and 3h after the onset of SE, respectively. We found that Ket4X protocol: i) dramatically reduced (≈ -75%) the induction of pro-inflammatory genes in the different brain areas; ii) prevented epileptogenesis and the development of anxiety-like behavior and cognitive impairments (spatial learning and hippocampal LTP). In addition, SE managed by Ket4X protocol did not compromise the development of brain plasticity induced by environmental enrichment. We conclude that more than duration of severe SE, profound reduction in brain activity after severe SE is crucial for the long-term outcome, and that good resolution of the inflammatory response may play a major role.

P3.113

Linking the brain to the environment: utilization of mobile technologies in MRI research

Rens, N. (Bordeaux)[1], Catheline, G. (Bordeaux)[2], Pelletier, A. (Bordeaux)[2], Allard, M. (Bordeaux)[3], Pérès, K. (Bordeaux)[4], Dartigues, J.-F. (Bordeaux)[5], Chanraud, S. (Bordeaux)[2], Swendsen, J. (Bordeaux)[6]
[1]Institut de Neurosciences Cognitives et Intégratives d'Aquitaine, Université Victor Segalen Bordeaux 2, Bordeaux, France, [2]EPHE, Bordeaux, France, [3]EPHE, CHU, CNRS, Bordeaux, France, [4]ISPED, Bordeaux, France, [5]ISPED, Bordeaux, Gabon, [6]EPHE, CNRS, Bordeaux, France

A major scientific challenge in treating Alzheimer's disease and other forms of dementia is the need to establish biomarkers for the earliest stages of cognitive decline, as well as to identify the lifestyles and

behaviors that may slow the dementia process. These important goals have been hindered by methodological limitations of traditional techniques, notably neuropsychological tests and neuroimaging, which are administered at one point in time and within a single environmental context. Mobile technologies such as smart phones offer the ability to provide repeated cognitive assessments that reduce the error associated with single-test data, and can readily identify daily life behaviors that promote cognitive health. This study included 55 participants (mean age=75.27, 45% female) from the AMI cohort of elderly individuals in the Bordeaux region. Participants completed a one-week period of ambulatory monitoring of semantic memory performance and daily life behaviors, MRI examinations, and a full neuropsychological battery. Brain diffusion biomarkers were found to be significantly associated with the frequency of various daily life behaviors. Fractional anisotropy of the left superior corona radiata correlated positively with participation in stimulating activities such as listening to music (t=2.807, p=0.007) and playing board games (t=2.203, p=0.032). In the corpus callosum, mean diffusivity was lower with greater participation in intellectually stimulating activities (t=-2.881, p=0.006) and recreation (t=-2.541, p=0.014). Such behaviors potentially have a protective effect as, in the same area, a reduction in fractional anisotropy corresponded to more frequent difficulties in the surroundings (t=-2.067, p=0.044). This investigation also provides evidence that specific activities of daily life prospectively improve key cognitive functions such as semantic memory. The findings demonstrate that mobile assessment techniques offer an important complement to neuropsychological data and provide a powerful tool for aiding the identification of anatomical correlates in the brain of early cognitive decline.

P3.114

Anxiety-like behavior of prenatally stressed rats is associated with a selective reduction of glutamate release in the ventral hippocampus

Morley-Fletcher, S. (Villeneuve d'Ascq)[1], Marrocco, J. (Villeneuve d'Ascq)[1], Mairesse, J. (Villeneuve d'Ascq)[1], Ngomba, R.T. (Pozzilli)[2], Van Camp, G. (Villeneuve d'Ascq)[1], Bouwalerh, H. (Villeneuve d'Ascq)[1], Silletti, V. (Villeneuve d'Ascq)[1], Summa, M. (Genoa)[3], Pittaluga, A. (Genoa)[3], Nicoletti, F. (Pozzilli)[2,4], Maccari, S. (Villeneuve d'Ascq)[1]
[1]Université Lille Nord de France, UMR 8576 UGSF, Equipe Neuroplasticité, Villeneuve d'Ascq, France, [2]IRCCS Neuromed, Pozzilli, Italy, [3]Dept. of Experimental Medicine, University of Genoa, Genoa, Italy, [4]Sapienza University, Dept. of Pharmacology, Rome, Italy

Abnormalities of synaptic transmission and plasticity in the hippocampus represent an integral part of the altered programming triggered by early life stress. Prenatally restraint stressed (PRS) rats develop long-lasting biochemical and behavioral changes, which are expression of an anxious/depressive-like phenotype. We report here that PRS rats showed a selective impairment of depolarization- or kainate-stimulated glutamate and [^{3}H] D-aspartate release in the ventral hippocampus, a region encoding memories related to stress and emotions. GABA release was unaffected in PRS rats. As a consequence of reduced glutamate release, PRS rats were also highly resistant to kainate-induced seizures. Abnormalities of glutamate release were associated with large reductions in the levels of synaptic vesicle-related proteins, such as VAMP (synaptobrevin), syntaxin-1, synaptophysin, synapsin Ia/b and IIa, munc-18, and Rab3A in the ventral hippocampus of PRS rats. Anxiety-like behavior in male PRS (and control) rats was inversely related to the extent of depolarization-evoked glutamate release in the ventral hippocampus. A causal relationship between anxiety-like behavior and reduction in glutamate release was demonstrated using a cocktail of the mGlu2/3 receptor antagonist, LY341495, and the $GABA_B$ receptor antagonist, CGP52432, which was shown to amplify depolarization-evoked [^{3}H] D-aspartate release in the ventral hippocampus. Bilateral microinfusion of CGP52432 *plus* LY341495 in the ventral hippocampus abolished anxiety-like behavior in PRS rats. These findings indicate that an impairment of glutamate release in the ventral hippocampus is a key component of the neuroplastic program induced by PRS, and that strategies aimed at enhancing glutamate release in the ventral hippocampus correct the “anxious phenotype” caused by early life stress.

P3.115

Transplantation of human stem cells from the umbilical cord to repair perinatal brain injury: a pre-clinical study

Dalous, J. (Paris)[1,2], Pansiot, J. (Paris)[1,2], Pham, H. (Paris)[1], Bruneau, L.C. (Paris)[1], Chatel, P. (Paris)[1], Nadaradja, C. (Paris)[1], David, G. (Paris)[1], D'Agostino, I. (Paris)[1], Vottier, G. (Paris)[1], Schwendimann, L. (Paris)[1], Vanneaux, V. (Paris)[3], Charriaut-Marlangue, C. (Paris)[1], Titomanlio, L. (Paris)[1], Gressens, P. (Paris)[1,2], Larghero, J. (Paris)[3], Baud, O. (Paris)[1,2,4]

[1]UMR 676, INSERM-Université Paris-Diderot, PRES Sorbonne Paris-Cité, Hôpital Robert Debré, Assistance Publique-Hôpitaux de Paris, Paris, France, [2]PremUp Foundation, Paris, France, [3]Assistance Publique-Hôpitaux de Paris, Hôpital Saint Louis, Unité de Thérapie Cellulaire et Centre d'Investigation Clinique en Biothérapies CIC-BT 501, Paris, France, [4]NICU, Université Paris Diderot, Hôpital Robert Debré, APHP, Paris, France

Cerebral palsy is a neurological disorder that affects the developing brain causing motor and cognitive deficits. There is no specific treatment for these perinatal brain lesions.
Cell therapy seems promising to repair brain damages. The human umbilical cord is a rich source of stem cells with many advantages (easy isolation, low immunogenicity, etc).
Our goal is to investigate the potential of human cord blood mononuclear cells (hUCB-MNCs) or mesenchymal stem cells from the Wharton's jelly (hUC-MSCs) to prevent or repair cerebral lesions in an animal model of neonatal excitotoxic brain injury.
This animal model consisted of an intracranial (i.c.) injection of ibotenate, in P5 Sprague-Dawley rats. Cells (from 10^6 to 10^7) were injected either intraperitoneally (i.p.), i.c. or intravenously (i.v.), soon or 24 hours after ibotenate injection. Cell fate and effects of transplantation on brain lesions were studied using molecular biology and histological methods.
Thus, we used various routes of administration, various cell amounts and various injection delays after injury.
Using the i.p. route, hUCB-MNCs could not enter the systemic circulation. However, the injection of 10^7 hUCB-MNCs entailed a deleterious increase in the lesion size exclusively in the WM that was associated with perilesional microgliosis and with the increase in serum concentrations of several cytokines.
On the contrary, the i.p. injection of 10^6 hUCB-MNCs was beneficial as it decreased the lesion size in the cortex.
hUCB-MNCs injected i.v. soon or 24 hours after the excitotoxic insult did not affect lesion size, serum cytokine concentrations, microglial activation, astroglial cell density, or cell proliferation at any concentration used.
First i.c. injections with hUC-MSCs into the lesion side entailed a decrease in astrogliosis in the perilesional WM.
As a conclusion, we showed that in most conditions tested, hUCB-MNCs could not integrate into the developing brain or promote subsequent repair. The intraperitoneal injection of high amounts of hUCB-MNCs aggravated WM damage and was associated with systemic inflammation, while small amounts were protective in the cortex (Dalous et al., 2013).
Preliminary results with hUC-MSCs are interesting and support their possible protective role.

P3.116

Environmental enrichment improves hyperdopaminergic-related deficits in DAT-KO mice

Grannec, G. (Paris)[1], Morice, E. (Paris)[1], Germain, J. (Paris)[1], Giros, B. (Paris)[1], Nosten-Bertrand, M. (Paris)[1]

[1]Université Pierre et Marie Curie, INSERM URMS 952, CNRS UMR 7224, Paris, France

Imbalance of dopaminergic (DA) neurotransmission is thought to contribute to a broad spectrum of symptoms observed in many psychiatric disorders, including schizophrenia, drug addition, and attention-deficit hyperactivity disorders.
The dopamine transporter (DAT) plays a critical role in calibrating the duration and intensity of DA neurotransmission. Mice lacking the *DAT* gene (*DAT*-KO) have been extensively used as a genetic model of persistent and functional hyperdopaminergia. We previously showed that the genetic background dramatically affects many of hyperdopaminergic-related physiological and behavioral endophenotypes in *DAT*-KO mice.
In the present study, we test the extent of phenotypic variation in *DAT*-KO mice depending on their environmental conditions. We report that exposure to environmental enrichment (EE) from weaning to adulthood modulates both quantitative and qualitative patterns of novelty-driven spontaneous hyperactivity and remarkably reduces the severity of stereotypic activities. In addition, we show that EE improves procedural learning and memory performances in various conditions.
Altogether these results will contribute to better understand how genetic-environment interaction contributes to the variability of DA-related endophenotypes of psychiatric.

P3.117

Dopaminergic modulation of the emotional conflict in Parkinson's disease

Fleury-Nissen, V. (Grenoble)[1], Cousin, E. (Grenoble)[2], Schmitt, E. (Grenoble)[3], Tropres, I. (Grenoble)[3], Krainik, A. (Grenoble)[3], Krack, P. (Grenoble)[1]
[1]Centre Hospitalier de Grenoble/Université Joseph Fourier/INSERM, Unité 836, Institut des Neurosciences de Grenoble, Grenoble, France, [2]Laboratoire de Psychologie et Neurocognition, CNRS UMR 5105, Grenoble, France, [3]Centre Hospitalier de Grenoble, Grenoble, France

Objective: To explore the dopaminergic modulation of emotional conflict processing in Parkinson´s disease (PD) using functional magnetic resonance imaging (fMRI).
Background: A distinctive personality type, characterized by introversion, inflexibility, low novelty seeking, depression and anxiety, has been suggested to be associated with PD. The neural mechanisms underlying the parkinsonian personality are still largely unknown. Some studies have indicated a relationship with decreased striatal and frontal dopaminergic tone. We hypothesized that the personality traits in PD relate to a tendency to be more vulnerable to emotional stimuli and that this process is dopamine-dependent.
Methods: Total of 24 participants: 12 PD patients (3♀ , 9♂ , mean age 59 years) without manifestations of depression or dementia and 12 healthy volunteers chosen to match the patient group for age, sex and education level, performed a modified Emotional Stroop (ES) task during fMRI. Each PD patient performed the ES task twice, once during maximum therapeutic effect of dopaminergic treatment (drug-on state) and secondly in a hypodopaminergic (drug-off) state. The dopaminergic modulation during emotional conflict (incongruent versus congruent contrast) as well as the phases of emotional conflict (monitoring and resolution) were studied.
Results: Whole-brain analysis in controls showed a consistent activation of the rostral anterior cingulate cortex (rACC) during the negative ES contrast. Region of interest analyses centered on the rACC showed a modulation of this area depending on the medication condition and on the conflict phase condition. The activation of the rACC was modulated by the medication condition with an attenuation in patients in their drug-off state, and was associated with the resolution of emotional conflict.
Conclusion: Our study demonstrates the existence of dopaminergic modulation in the resolution of emotional conflict and the role of the dopaminergic system in the interactions between decision making and emotional processing in PD patients, which partly explains their vulnerability to emotive stimuli such as psychosocial stressors.

P3.118

Brain functional connectivity and morphology changes in medication-overuse headache: evidence for addiction-related processes

DiScala, G. (Talence)[1], Chanraud, S. (Talence)[1,2], Dilharreguy, B. (Talence)[1], Schoenen, J. (Liège)[3], Allard, M. (Talence)[1,2], Radat, F. (Bordeaux)[4]
[1]Institut de Neurosciences Cognitives, et Intégratives d'Aquitaine (INCIA), CNRS UMR 5287, Université Bordeaux 1, Talence, France, [2]EPHE, Bordeaux, France, [3]Centre de Recherche des Céphalées, Université de Liège, Liège, Belgium, [4]Département de Neurosciences Cliniques, CHU Pellegrin, Bordeaux, France

Medication-overuse headache (MOH) derives from episodic migraine (EM) and becomes chronic with repetitive and excessive intake of pain killer drugs. The objective of the present study was to jointly evaluate voxel-based morphometry and whole-brain functional connectivity at rest in MOH patients, in comparison with EM patients and healthy controls (HC). Anatomical MRI and resting state functional MRI scans were obtained in MOH patients (n= 17 and 9 respectively), EM patients (n= 18 and 15 respectively), and HC (n= 17 and 17).
Using a seed-to-voxel approach, results revealed an altered functional connectivity at rest in MOH patients, comprising decreased connectivity between precuneus and other regions of the default-mode network (frontal and parietal cortices), and increased connectivity between precuneus and hippocampal/temporal areas. These functional alterations were not accompanied with significant morphological changes. In MOH patients, connectivity between precuneus and frontal areas was negatively correlated with migraine duration and positively correlated with self-evaluation of medication dependence. Interestingly, grey matter volumes of frontal regions, precuneus and hippocampus were also negatively related to migraine duration. Finally, functional connectivity within the default-mode network appeared to predict MOH's anxiety scores; and, grey matter volumes in the same network predicted MOH's depression scores.
Our data suggest that MOH is related to alterations within intrinsic functional networks rather than modifications at a structural level. They also support the view that addiction-related processes might play a prominent role in its development.

P3.119

Intraglomerular lateral inhibition promotes spike timing variability in principal neurons of the olfactory bulb

Desaintjan, D. (Paris)[1,2], Najac, M. (Paris)[1], Charpak, S. (Paris)[1]
[1]INSERM U 603, CNRS UMR 8154, Université Paris Descartes, Paris, France, [2]Institut des Neurosciences Cellulaires et Intégratives, Strasbourg, France

In the olfactory bulb, two layers of interneurons mediate inhibition. Granule cells make lateral inhibitory connections between spatially distributed glomeruli. They generate spike synchrony and fast oscillations in mitral and tufted cells, the principal neurons of the bulb, and play an important role in odor processing. In contrast, little is known about intraglomerular inhibition generated by periglomerular (PG) cells, a heterogeneous population of interneurons that surrounds each glomerulus. Recent data suggest that the feed-forward inhibition evoked by an olfactory nerve input in mitral and tufted cells is generated within the glomerulus (Najac et al., JNeurosci 2011, Shao et al., JNeurophysiol 2012). We investigated the circuit that mediates this inhibition as well as its impact on principal neurons using patch-clamp recording in olfactory bulb slices and in anesthetized animals in a transgenic mouse expressing EYFP in a subset of PG cells. These monoglomerular PG cells represent 20-30% of the whole PG cell population. Paired-recording demonstrate that they are activated by mitral, tufted and external tufted cells and in turn release GABA onto these output neurons. We also show that EYFP-positive PG cells mediate lateral inhibition between principal neurons connected to the same glomerulus and are activated during feed-forward inhibition evoked by

even the weakest olfactory nerve input. This pathway is also strongly activated during each respiratory cycle *in vivo*. Intraglomerular lateral inhibition reduces the output frequency and promotes spike timing variability in mitral and tufted cells, suggesting that intraglomerular inhibitory circuits play a key role in processing of olfactory information.

P3.120

Real-time decoding from population activity in both FEF hemispheres: how flexible is the decoding?

Astrand, E. (Bron)[1], Huguet, N. (Bron)[1], Wardak, C. (Bron)[1], Ben Hamed, S. (Bron)[1]
[1]*ISC - Centre de Neuroscience Cognitive, CNRS UMR 5229, Bron, France*

Current invasive Brain Machine Interfaces (iBMI) have up to now essentially focused on interpreting motor variables from the motor primary, the premotor or the parietal cortex. The field of cognitive iBMI (allowing a real-time access to internal mental representations), in spite of its potential high therapeutical impact, is barely trodden. A prerequisite for the success of cognitive iBMI is the capacity to access to a reliable estimation of cognitive variables, independently of the context. Indeed, in real life, we are confronted with a changing sensory environment and changing internal drives and goals. Here, we approach this issue by introducing
1) environmental visual changes in a stable cognitive context and
2) changes in the cognitive demands, in an otherwise stable sensory context, while decoding in real-time visual- and motor-related processes from prefrontal neuronal population multi-unit (MUA) and local field potential (LFP) responses.
The real-time decoding accuracy is obtained from simultaneous invasive recordings in both FEF bilaterally (using two 24-contact linear multi-electrodes, one in each FEF), in two monkeys, while they are engaged in three behavioral tasks. Task 1 is a memorized saccade task designed to dissociate between visual- and motor-related processing. The monkeys need to memorize the position of a visual stimulus that is flashed for 100ms and make saccade to its position after a variable delay period. Task 2 is the same as task 1 except that a random noise background is present all throughout the trials. Task 3 is a simple detection task. The monkeys have to fixate a central point and detect a flashed stimulus by releasing a bar. Tasks 1 and 2 have the same cognitive demands but different sensory environments. Tasks 1 and 3 have the same sensory environment at the moment of the visual presentation but different cognitive demands.
We show that while the average real-time accuracy at decoding the visual-related information in each independent task is above 90%, the generalization of the decoding from a given task to another, incurs a loss of 5 to 10% in average real-time accuracy. Accuracy increases back to around 90% when the initial training of the decoder is performed on population responses collected on all three tasks.

P3.121

Cortical mechanisms for shifting and holding visuospatial attention: a magnetoencephalography (MEG) study

Huguet, N. (Bron)[1], Ibos, G. (Bron)[1], Ben Hamed, S. (Bron)[1]
[1]*Centre de Neuroscience Cognitive, CNRS UMR 5229, Bron, France*

Visuo-spatial attention is a critical cognitive function that allows the detailed analysis of the most relevant information in the visual field. It is well established that multiple frontal and parietal regions in the human cortex are involved in the voluntary control of spatial attention, top-down biasing visual processing in occipital visual areas. In particular, imaging studies suggest that shifting attention from

one peripheral location to another is associated with transient increases in neural activity in the superior parietal lobule (SPL) and frontal eye fields (FEF). In contrast, precentral sulcus (PreCS) and posterior parietal cortex (PPC) have been described as more active when attention is maintained in a given spatial location. However, little is known about the timing and sequence of activation within this network.
We used magneto-encephalographic recordings while subjects performed a cued-target detection task embedded in a rapid serial visual search presentation. The cue instructed subjects either to shift their attention or to hold it at a given spatial position. We show that shifting and holding attention recruit only partially overlapping networks. Specifically, shift cues produce a sustained activation of VLPFC, promptly followed by a fronto- (IFG) parieto- (TPJ, SPL and IPS) frontal (IFG and FEF) activation sequence. An enhanced endpoint contralateral occipital response is observed around 180ms following shift cue onset. In contrast, stay cues produce a transient VLPFC activation followed by a direct fronto- (MFG) parietal (TPJ, IPS, precuneus, cuneus and SPL) activation sequence. An enhanced endpoint contralateral occipital response is observed around 150ms following stay cue onset. This 30ms offset in occipital enhancement following shift and stay cues nicely match our behavioral estimates of attentional shift time cost (Ibos et al., 2009).
Overall, we thus propose that holding attention in a given spatial location involves a direct interaction between the frontal and parietal attention-related areas while shifting attention from one hemisphere to the other requires a more complex interplay between frontal and parietal areas, incurring at least 50ms time cost.

P3.122

Eye movements control by the cerebellar nuclei: polysynaptic pathways from fastigial, interpositus posterior and dentate nuclei to lateral rectus motoneurons in primates

Ugolini, G. (Gif sur Yvette)[1], Prevosto, V. (Durham)[1,2], Graf, W. (Washington)[3]
[1]CNRS, Neurobiologie & Développement, UPR 3294 CNRS, Gif sur Yvette, France, [2]Center for Cog. Neuro., Duke University, Durham, United States, [3]Dept. Physiol. & Biophysics, Howard University, Washington, United States

We used retrograde transneuronal transfer of rabies virus (RV) in macaque monkeys (n=4), to identify polysynaptic pathways to lateral rectus (LR) motoneurons (MNs). We found that they are the target of cerebellar output channels from the fastigial (F) di- and trisynaptically, and from caudal dentate (D) and rostral ventrolateral interpositus posterior (IP) nuclei trisynaptically. RV was injected into the left LR muscle and visualized immunohistochemically 2.5, 3 and 3.5 days (d.) later. Transfer from LR MNs visualized connected second-order neurons at 2.5 d. (Ugolini et al., 2006), third-order at 3 d. (e.g., in superior colliculus, SC, Grantyn et al., 2002) and fourth-order at 3.5 d. Disynaptic cerebellar inputs to LR MNs, revealed at 3 d., are derived from the F (caudal 2/3) bilaterally (ipsi 154/contra 97 neurons). Trisynaptic inputs (3.5 d.) originate bilaterally from the ventrolateral IP rostrally (ipsi 54/contra 93), caudal D (lateral half) (ipsi 104/contra 133) and F (ipsi 420/contra 482, including also the rostral F). Disynaptic inputs to LR MNs from the caudal F (involved in saccades, smooth pursuit and convergence) is explained by its projections to second-order oculomotor neurons in paramedian pontine, dorsal paragigantocellular and central mesencephalic reticular formation (saccade modulation), supraoculomotor area (SOA, near-response) and medial vestibular nucleus (Noda et al. 1990). Trisynaptic F inputs are consistent with caudal F projections to third-order neurons in these and other cell groups, including raphe interpositus and SC. Inputs from the rostral F, which contributes to the control of combined eye-head gaze shifts, is likely mediated by projections to third-order populations in the vestibular nuclei, that have access to both eye and head motor control pathways. Caudal D inputs can be explained by its SC projections and likely target saccade-related burst neurons (labeled at 3 d.). The same caudal D portion also targets disynaptically frontal and parietal eye fields (Prevosto et al., 2010). Labeling of the rostral ventrolateral IP (involved in divergence) is likely mediated by projections to SOA (May et al., 1992) and from SOA to LR MNs (Ugolini et al., 2006), explaining LR MNs activation during divergence.
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P3.123

New insights into the generators of the auditory frequency following response: a SEEG study with speech sounds

Bellier, L. (Bron)[1], Thai-Van, H. (Lyon)[2], Caclin, A. (Bron)[1]
[1]Centre de Recherche en Neurosciences de Lyon, Équipe DyCog, Bron, France, [2]Hospices Civils de Lyon, HEH, Service Audiologie, CRNL, Équipe PACS, Lyon, France

Speech Auditory Brainstem Responses (*Speech ABR*s) are scalp-recorded evoked potentials reflecting the first steps of speech sounds processing in the auditory system. Due to their high fidelity to the stimulus, they represent a very promising diagnostic tool for hearing impairment assessment and also a powerful way to better understand the first steps of speech processing in the central nervous system. However, little is known about their cerebral generators.
Two main components of *Speech ABR*s are classically described: the *onset response*, evoked by transient speech sounds (e.g., stop consonant /b/) and similar to the *click ABR*'s wave V; and the *Frequency Following Response* (*FFR*), evoked by periodic signals (e.g., vowel /a/) and similar to that evoked by harmonic tones. This second component replicates the fundamental frequency of the vowel: since the *FFR* contains high frequencies (170 or 200 Hz in our case), it is thought to be strictly of sub-cortical origin.
In previous research, we simultaneously recorded sub-cortical and cortical evoked responses to /ba/ speech syllables using a 32-channel EEG system. Topographical analysis revealed the existence of different generators of the *onset response* and the *FFR* components. In addition, an unsettling similarity was found between cortical ERPs (Evoked-Related Potentials) P50-N1-P2 and brainstem *FFR* scalp current density maps. This raised the following question: Could there be a cortical component of the *FFR* - supposed to be sub-cortically elicited?
We thus recorded intracranial cortical activity in response to the same /ba/ speech stimuli, in epileptic patients who were pre-surgically implanted for epileptogenic focus localization purpose. We indeed observed high frequency activity within the auditory cortex, concomitant to *FFR* recordings. This striking result questions a substantial amount of *Speech ABR*s and *FFR* studies, especially in expert (e.g., musicians) or deficient (e.g., dyslexics) individuals, that interpreted signal change in the *FFR* as the reflection of brainstem plasticity.

P3.124

Role of the basal ganglia and cerebellum in the functional phenotype of *Dab1^{scm}* (*scrambler*) mutant mice

Strazielle, C. (Vandoeuvre-les-Nancy)[1], Jacquelin, C. (Vandoeuvre-les-Nancy)[1], Lalonde, R. (Mont-Saint-Aignan)[2]
[1]Université de Lorraine, Faculté de Médecine, Vandoeuvre-les-Nancy, France, [2]Université de Normandie, Faculté des Sciences, Mont-Saint-Aignan, France

In the mutations affecting the reelin signaling pathway, disruption of *disabled-1* gene characterizes a natural recessive *scrambler* mutation (*Dab1^{scm}*). The resulting ataxic mice display disorganized lamination of the cerebral and cerebellar cortex but also malpositioning of mesencephalic dopaminergic neurons.
To investigate the possible impact of specific brain regions on the functional phenotype of the *Dab1^{scm}* mutation, regional brain metabolism, as assessed by cytochrome oxidase (COX) activity, was measured in mutant mice in comparison with controls. Metabolic changes occurred mainly in mutant structures intimately connected with the cerebellum, in numerous structures of basal ganglia circuitries, in limbic regions, particularly hippocampus, as well as in visual and parietal sensory

cortices. Although behavioral results characterized a major cerebellar disorder in the *Dab1^{scm}* mutants, motor activity impairments in the open-field were associated with COX activity changes in the basal ganglia efferent structures such as the substantia nigra, pars reticulata. Hypometabolism in this structure was also associated with the anxiety changes observed in the elevated plus-maze and emergence test. Additional neurochemical studies were performed to investigate the basal ganglia defect. The dopaminergic innervation visualized by immunocytochemistry was lower in the mutant mesencephalic regions, more particularly in substantia nigra and retrorubral field. However, HPLC measurements of biogenic amines revealed differences between the two genotypes in the cerebellum but not in neostriatum or brainstem.
In addition of the impact of the cerebellum, these results indicate a crucial participation of the basal ganglia in the functional phenotype of ataxic *Dab1^{scm}* mutants.

P3.125

Studying the cellular basis of axial motor activity in the neonatal rat spinal cord

Beliez, L. (Bordeaux)[1], Barriere, G. (Bordeaux)[1], Bertrand, S. (Bordeaux)[1], Cazalets, J.-R. (Bordeaux)[1]
[1]*Université Bordeaux Segalen, CNRS UMR 5287 INCIA, Bordeaux, France*

The ability to move from place to another effectively requires interactions between various body parts and the central mechanisms involved in the functional coupling of these body segments remain so far largely unknown. If many studies have focused on the organization and physiology of hindlimb locomotor networks, few have addressed the motor control of the trunk. The aim of our study is to investigate the physiology of trunk motor networks in mammals.
We have used an isolated neonatal rat spinal cord preparation to investigate some features of the coupling between the lumbar locomotor network and the thoracic networks involved in the control of limb and axial movements, respectively. In this preparation, fictive locomotion can be pharmacologically triggered and extracellularly recorded from various ventral roots by application of NMA (a glutamate analogue) and serotonin (5HT) on restricted spinal segments.
In a first set of experiments, we confirmed that thoracic networks had intrinsic ability to generate rhythm. Then, by combining extracellular ventral roots recordings and intracellular recordings from thoracic axial motoneurons we have shown that during fictive locomotion, some of these cells exhibit coordinated rhythmic membrane potential oscillations, on the top of which they can fire action potentials suggesting a control from lumbar networks to thoracic motoneurons.
A pharmacological approach allowed us to investigate more precisely the synaptic connections and neurotransmitters involved in this coupling. We also study the strength of this coupling along different thoracic levels.
This is the first description at the cellular level of the neuronal trunk networks.

P3.126

Neuromodulation during ventral spinal cord development in the SOD1 G93A mouse model of amyotrophic lateral sclerosis

Milan, L. (Bordeaux)[1], Barrière, G. (Bordeaux)[1], De Deurwaerdère, P. (Bordeaux)[2], Cazalets, J.-R. (Bordeaux)[1], Bertrand, S.S. (Bordeaux)[1]
[1]*Université Bordeaux Segalen, INCIA, Bordeaux, France,* [2]*Université Bordeaux Segalen, IMN, Bordeaux, France*

As animals live in a constantly changing environment, the motor outputs generated by spinal neuronal networks in vertebrates should be permanently adjusted. This flexibility partly relies on the interplay

between the numerous neuromodulatory influences received by the motoneurons that link the central nervous system to the muscles. Amyotrophic lateral sclerosis (ALS) is a neurodegenerative disease characterized by the selective loss of motoneurons in the spinal cord. While ALS symptoms occur in adulthood, modifications of motoneuron excitability and morphology have been observed in early developmental stages in the copper/zinc superoxide dismutase (SOD1) mouse model of ALS. There is however no data available so far on possible alterations of neuromodulatory systems in SOD1 spinal motor networks during development. To address this question, we conducted a series of experiments using the *in vitro* spinal cord preparation from newborn mice (postnatal day 0-3). This *in vitro* preparation could express different motor activities when superfused with appropriated neuroactive compounds. It has been shown that the concomitant bath-application of an agonist of the NMDA receptors, NMA, with serotonin (5HT) induces a locomotor-like activity characterized by right and left and extensor-flexor alternating bursts of action potentials recorded from the lumbar ventral roots. In this study, we examined and compared the ability of biogenic amines (dopamine, noradrenaline and serotonine) and cholinergic agonists to produce motor activity and to modulate NMA-5HT-induced fictive locomotion in the isolated spinal cord preparation from neonatal $SOD1^{G93A}$ mice and age-matched littermate controls. To complement this study, the tissue content of biogenic amines and their metabolites was measured by quantitative HPLC. In addition to a significant contribution to the general understanding of the physiology and development of spinal motor networks, this study provides, to our knowledge, the first precise analysis of the neuromodulatory state in motor spinal networks during ALS development.

P3.127

Pain, fourteen days after spinal nerve ligature, is correlated to a decreased number of astrocytes and microglia in the thalamic ventroposterolateral nucleus

Blaszczyk, L. (Bordeaux)[1,2], Maître, M. (Bordeaux)[1,2], Lesté-Lasserre, T. (Bordeaux)[1,2], Oliet, S.H.R. (Bordeaux)[1,2], Fénelon, V.S. (Bordeaux)[1,2]
[1]INSERM U 862 Neurocentre Magendie, Bordeaux, France, [2]Université de Bordeaux, Bordeaux, France

Neuropathic pain is a chronic pain that develops after neural damage or dysfunction. At the spinal level, it is well established that reactive astrocytes and microglia (changes in morphology, number of cells and expression of specific markers) contribute to neuropathic pain. Therefore, the aim of this study was to determine whether astrocytes and microglia at the supra-spinal nociceptive transmission relays, the thalamic ventroposterolateral (VPL), ventroposteromedial (VPM), mediodorsal and intralaminary nuclei, display signs of reactivity and could therefore be involved in neuropathic pain mechanisms. To this end we used the unilateral L5/L6 spinal nerve ligature rat model (SNL).
By using von Frey filaments and a dynamic weight bearing apparatus, we found that mechanical allodynia and hyperalgesia as well as behavioural signs of ambulatory pain of the affected limb developed after 14 days post-surgery in SNL rats but not in sham (surgery without ligature) rats. Surface staining of GFAP (astrocytic intermediate filament), S100beta protein (astrocytic calcium binding protein) and iba-1 (microglial calcium binding protein) immunoreactivity (IR) was unchanged in all nociceptive thalamic nuclei in SNL and sham rats. By contrast, we found a decreased number of astrocytes (IR for S100beta) as well as a decreased number of microglia (IR for iba-1) in the contralateral VPL of SNL compared to sham rats. However, qRT-PCR analysis of transcripts of 28 structural and functional markers of reactive glia on lateral and central thalamic punches showed no difference between SNL and sham rats. Furthermore, by using qRT-PCR after performing laser microdissection, we found no change in transcript levels of 4 markers of reactive glia in VPL and VPM nuclei between SNL and sham rats.
Altogether our results demonstrate that after spinal nerve ligature, at a time when spinal astrocyte and microglia display robust morphological and functional signs of reactivity, these glia in nociceptive thalamic nuclei do not. Furthermore, we found a decreased number of astrocytes and microglia in the thalamic VPL of SNL rats. Therefore compensatory mechanisms may take place at the thalamic level in order to counterbalance the spinal sensitization that occurs in neuropathic pain model.

P3.128
Different retention decays after adaptation to in-depth distortion

Priot, A.-E. (Brétigny-sur-Orge)[1,2], Philippe, M. (Brétigny-sur-Orge)[1], Neveu, P. (Brétigny-sur-Orge)[1], Prablanc, C. (Bron)[2], Roumes, C. (Brétigny-sur-Orge)[1]
[1]Institut de Recherche Biomédicale des Armées (IRBA), Brétigny-sur-Orge, France, [2]Centre de Recherche en Neurosciences de Lyon (CRNL), Inserm U 1028 - CNRS UMR 5292, ImpAct, Bron, France

In a previous study (Priot et al., 2011), we found that visual adaptation to in-depth prismatic distortion actually involves i) a visual calibration process consistent with optimal reliability-based calibration (Ghahramani et al., 1997; van Beers et al., 2002) and ii) an eye muscle potentiation (Paap and Ebenholtz, 1977), corresponding to a change in sensed vergence effort following sustained vergence. The goal of the present study was to get into the dynamics of these processes after the end of the adaptation period. We measured visual aftereffects after an 8-min exposure to telestereoscopic viewing. We compared three target viewing conditions in an otherwise dark environment:
i) a red pinpoint of light (pinhole),
ii) a red cross which diameter was randomized in order to prevent size-cue, and
iii) a slide of a real-size one-euro coin.
Eye-muscle potentiation was assumed to develop equally in the three conditions, as target sequence and oculomotor tasks were exactly the same. The coin condition allowed some calibration of the mapping between the vergence/disparities signal and perceived distance. To investigate the time constants of these mechanisms, we tested subjects at three times after exposure, with a 1-hour interval of free normal binocular viewing and interacting. At the first post-test, an aftereffect was observed for all targets conditions. At the second post-test, an aftereffect was observed for both the cross and coin conditions. At the third post-test, only the coin viewing condition exhibited an aftereffect. These results suggest different retention processes after adaptation to in-depth distortion with their own decay time constants.

P3.129
Nesfatin-1 and rhythmic swallowing in rat

Felix, B. (Marseille)[1,2], Abysique, A. (Marseille)[1]
[1]AMU, EA 4674 PPSN, Marseille, France, [2]INRA, U 1189 Département AlimH, Saint-Genès-Champanelle, France

The caudal brainstem is the seat of motor networks for ingestive behaviour as it contains all the motoneurons that participate to swallowing, chewing, suckling, and licking. These ingestive patterns depend upon organized inputs from premotor neurons located in the dorsal vagal complex (DVC). The DVC contains the nucleus tractus solitarius (NTS) which is the site of integration of satiety signals, and the motoneurons that drive swallowing. Concerning the role of peptides and neurohormones in the regulation of feeding behaviour, the DVC contains abundant receptors and neuropeptide mediators relevant to food intake control.
In previous studies in the adult rat, we have demonstrated that the anorexigenic leptin and Brain-derived neurotrophic factor (BDNF) modify the activity of premotoneurons involved in swallowing. Nesfatin-1, a newly discovered potent anorexigenic factor, is also expressed in the brainstem. To investigate the involvement of Nesfatin-1 in the regulation of swallowing, we examined its effects on the reflex triggered by repetitive electrical stimulation of the superior laryngeal nerve (SLN) in anaesthetized rats. We found that a microinjection of Nesfatin-1 in the swallowing network induced complex responses. Although inhibitory effects were most often recorded, facilitatory effects were also

observed. Pretreatment by an ocytocinergic receptor antagonist potentiated the inhibitory effects, suggesting that Nesfatin-1 effects could be mediated via a modulation of the ocytocinergic signalling. To understand the mechanism of facilitatory effects of Nesfatin-1 on rhythmic swallowing, we want to determine whether there is an interaction between Nesfatin-1 and NPY. Indeed, we found that NPY was able to facilitate swallowing. Moreover, recent data obtained *in vitro* showed that Nesfatin-1 depolarized NPY neurons in the NTS.
Finally, Nesfatin-1 is able to modulate swallowing partly by inhibiting or facilitating the reflex. The inhibitory effects could be in part mediated by ocytocinergic signalling. This inhibition of swallowing could explain the anorexigenic effect of Nesfatin-1 in the DVC regulation of food intake in mammals.
Keywords: swallowing, Nesfatin-1, inhibition, facilitation, Ocytocin

P3.130
Privileged visual processing of near space in humans

Severac Cauquil, A. (Toulouse)[1], Paez, E. (Toulouse)[1], Rosito, M. (Toulouse)[1], Celebrini, S. (Toulouse)[1]
[1]*CerCo UMR 5549, Cnrs/Université Paul Sabatier, Toulouse, France*

Three-dimensional (3D) space can be divided into different zones according to our behaviour: near, peripersonal or reaching space involved in objects manipulation and far, extrapersonal space involved in visual search, recognition, posture and locomotion (Previc, 1998, Psych bull, 124(2) 123-64). Different brain structures process these different spaces: dorso parieto-occipital cortex for near space and ventro-occipital for far space (Weiss et al., 2000, Brain, 123, 2531-41). Our team showed recently that straight ahead direction, ecologically meaningful, was processed quicker (Durand et al., 2012, J of vision, 12(6):34, 1-13). Since it also is ecologically crucial for our security, the present work addresses the question whether the near space benefits from a privileged processing.
7 subjects (6 right-handed) were included in the study. In total dakness, they had to fixate a red point in the middle of a screen and strike the return key of the keyboard as soon as the stimulus, a 1-° gabor was flashed. 3 distances (30, 60, 90 cm) and 4 positions of the stimulus (6° above, below, at the right and at the left of the fixation point) were used. Each position of the stimulus was repeated 50 times for each of the 3 distances, 600 trials were therefore recorded during the 3 blocks, one block for each distance. The order of the blocks was randomized across subjects.
Subjects were significantly quicker for 30-cm and 60-cm presentations than for 90-cm (333 and 339 ms vs 349 ms; ANOVA, $p< 0.05$) and for lower-field and right presentation than upper and left (333 ms and 335 ms vs 350 ms and 344 ms; ANOVA, $p< 0.001$). Distance effect is even stronger for lower-field presentation (ANOVA, interaction distance*position, $p< 0.05$).
Our results suggest that near space does benefit from a privileged processing, making the detection quicker, probably in order to allow us to avoid or grasp objects in our reaching space. This could be achieved by a sharper sensitivity of cells of the visual cortex when the visual stimulus is closer, mediated by a higher spontaneous activity as shown by Trotter et al (1992, Science, 257, 5074, 1279-81). In the latter study, only vergence changed with distance, we then hypothesize that the vergence signal is responsible for this modulation.

P3.131
Excitability and integrative properties of neocortical neurons after global suppression of spontaneous electrical brain activity

Altwegg-Boussac, T. (Paris)[1], Chavez, M. (Paris)[2], Mahon, S. (Paris)[3], Charpier, S. (Paris)[4]

[1]UPMC, Paris, France, [2]CNRS, Paris, France, [3]INSERM, Paris, France, [4]UPMC University, CNRS UMR 7225, INSERM UMRS 952, Paris, France

In absence of any sensory stimuli or motor behaviours, the “resting” brain generates a continuous and endogenous electrical activity, which dynamically varies as a function of states of vigilance. In neocortical neurons, this activity takes the form of spontaneous membrane potential fluctuations, reflecting ongoing excitatory and inhibitory synaptic inputs. The impact of this internal activity on the functional properties of cortical neurons remains unclear. *In vitro* studies, using artificial synaptic “noise”, suggest either a facilitating effect on neuronal responsiveness to external stimuli or a decreased excitability due to an augmentation of membrane conductance.
Here, by the means of *in vivo* simultaneous electrocorticographic (ECoG) and intracellular recordings of pyramidal cells of the rat somatosensory cortex, we compared in the same neurons, in control condition and after complete suppression of synaptic activities, the membrane excitability and the input-output function. To reproduce the two main brain states, we generated in control condition a slow-wave sleep-like activity (pentobarbital anesthesia) or a desynchronized waking-like ECoG pattern (fentanyl sedation).
Waking-like activity, compared to sleep-like condition, was characterized by a more depolarized membrane potential, smaller synaptic fluctuations and lower values of input resistance. Global suppression of synaptic activities, by an elevated dose of anesthetic, led to an isoelectric ECoG and a complete disappearance of synaptic events in cortical neurons. Whatever the initial condition, abolition of brain activity resulted in a hyperpolarization of cortical neurons, a large increase in the current threshold for action potential generation but did not affect the membrane input resistance. Cortical neurons under fentanyl exhibited after induction of the isoelectric state a decreased neuronal gain. The obliteration of endogenous activity was also responsible for a remarkable temporal fidelity in the current-induced neuronal firing pattern. Altogether, these findings suggest that background brain activity, whatever the state of vigilance, makes cortical neurons highly excitable, facilitates their global responsiveness to external inputs and increases their output temporal variability.

P3.132

Role of the noradrenergic system in olfactory learning and adult neurogenesis

Didier, A. (Lyon)[1], Richard, M. (Lyon)[1], Vinera, J. (Lyon)[1], Moreno, M. (Lyon)[1], Sacquet, J. (Lyon)[1], Mandairon, N. (Lyon)[1]
[1]Centre de Recherche en Neurosciences de Lyon (CRNL), Inserm U 1028 - CNRS 5292 - UCBL1, Lyon, France

The main olfactory bulb is the first cortical relay involved in the transmission, processing and integration of olfactory information. It is also one of the brain regions receiving strong noradrenergic innervation from the *locus coeruleus*. Noradrenergic inputs play a critical role in several olfactory functions, such as odor discrimination, and various forms of olfactory memory. In addition, the olfactory bulb is one of the rare brain regions where neurogenesis persists during adulthood. Adult neurogenesis is differently required for specific forms of olfactory learning. Indeed, neurogenesis is necessary for the acquisition of perceptual learning (a form of learning where two undiscriminated odors become discriminated after a 10-day exposure to these two odors), but only for the long term retention of associative olfactory memory (the animal learns to associate a reward to the presence of a specific odor).
In this study, we aimed at identifying the cellular and molecular basis of the different requirement of neurogenesis in perceptual and associative olfactory learning. We first investigated the role of noradrenaline in these two forms of learning. We performed *in vivo* intrabulbar infusion of noradrenergic receptor antagonist to locally block the noradrenergic signaling pathway during perceptual and associative learning. We showed that bulbar noradrenaline is necessary for perceptual learning but not for associative olfactory memory.
We then investigated the mechanisms linking adult neurogenesis, olfactory learning and noradrenaline. Blocking the noradrenergic pathway induces alterations of newborn neuron survival during perceptual learning but not during associative learning. In addition, we showed that noradrenergic receptors are expressed in the olfactory bulb and that newborn neurons receive

noradrenergic synaptic contacts at crucial time points during their maturation and integration in the bulbar neuronal network.
Altogether our results support the hypothesis of a task-specific requirement of noradrenaline as a key regulator of adult neurogenesis during various forms of olfactory learning.

P3.133
Neurobehavioral evaluation of *Dab1^{scm}* (*scrambler*) mutant mice

Jacquelin, C. (Vandoeuvre-les-Nancy)[1], Strazielle, C. (Vandoeuvre-les-Nancy)[1], Lalonde, R. (Mont-Saint-Aignan)[2]
[1]Université de Lorraine, Faculté de Médecine, Vandoeuvre-les-Nancy, France, [2]Université de Normandie, Faculté des Sciences, Mont-Saint-Aignan, France

As disabled-1 protein (DAB1) acts downstream in the reelin signaling pathway modulating neuronal migration, glutamate neurotransmission, and cytoskeletal function, the *disabled-1* gene mutation (*scrambler* or *Dab1^{scm}* mutation) results in ataxic mice displaying dramatic neuroanatomical defects similar to those observed in the reeler gene (*Reln*) mutation. Characterized by cell ectopias and degeneration in cerebellum, hippocampus, and neocortex, the *Dab1^{scm}* mutant mice were compared to non-ataxic controls in various tests evaluating motor activity and coordination, spontaneous alternation, anxiety, and spatial learning in the Morris water maze.
As expected from their ataxia, the *Dab1^{scm}* mutants showed severe motor coordination impairments on stationary beam, coat-hanger, and rotorod tests although they were able to improve their performances on the vertical grid climbing test attesting motor learning capacity in a task adapted to their abilities.
Despite ataxia, the mutant mice displayed a higher level of motor activity in the open-field. They were also less anxious in the elevated plus-maze but with higher latencies in the emergence test. In the spontaneous alternation test, there were no group differences in alternation rates but controls alternated above chance levels, whereas the mutants did not. In the Morris water maze, *Dab1^{scm}* mutants showed deficits on both hidden and visible platform subtests.
The phenotypes of *Dab1^{scm}* mutants are similar to those of the *Reln$^{rl\text{-}orl}$* mutation.

P3.134
Aversive olfactory conditioning influences neurogenesis in the adult mouse olfactory bulb

Mandairon, N. (Lyon)[1], Kermen, F. (Lyon)[1], Sacquet, J. (Lyon)[1], Didier, A. (Lyon)[1]
[1]Centre de Recherche en Neuroscience de Lyon INSERM 1028 CNRS UMR 5292 Univ Lyon 1, Lyon, France

The olfactory information coming from the sensory neurons in the nasal cavity is processed in the olfactory bulb (OB). This structure undergoes a constant renewal of its interneurons during adult life, which function remains unknown.
Previous works showed that an associative olfactory conditioning, during which mice learned to associate a positive reward to an odorant, increases survival of newly formed neurons in the OB. However, it is unknown whether these changes in neurogenesis contribute to the encoding of the acquired positive significance of the odor.
To address this issue, we currently investigate the influence of an appetitive versus an aversive olfactory learning on bulbar neurogenesis. Adult mice learn during 5 days to associate an odor (+Limonene or -Carvone) to a sweet or a bitter bit of cereal. A week after conditioning , mice are tested for long term retention of the association and euthanized to assess survival and spatial

distribution of new neurons in the granule cell layer of the OB. Preliminary data suggest that both appetitive and aversive learning elicit an increase in neurogenesis in the OB. However, the spatial distribution of new neurons in the granule cell layer of the OB is different depending on the hedonic value acquired by the odor. Thus, this would suggest that neuronal bulbar turnover could contribute to the encoding of odor hedonic value.

P3.135

Antidepressant drugs relieve neuropathic allodynia by a peripheral beta2 adrenoceptor mediated anti-TNFalpha mechanism

Freund-Mercier, M.-J. (Strasbourg)[1], Bohren, Y. (Strasbourg)[1], Tessier, L.-H. (Strasbourg)[1], Petitjean, H. (Strasbourg)[1], Megat, S. (Strasbourg)[1], Kremer, M. (Strasbourg)[1], Daniel, D. (Strasbourg)[1], Hein, L. (Freiburg)[2], Schlichter, R. (Strasbourg)[1], Hugel, S. (Strasbourg)[1], Yalcin-Christmann, I. (Strasbourg)[1], Barrot, M. (Strasbourg)[1]
[1]UPR 3212 CNRS, Université de Strasbourg, Strasbourg, France, [2]University of Freiburg, Freiburg, Germany

Neuropathic pain is defined as a chronic pain resulting from a direct consequence of a lesion or disease affecting the somatosensory system. Antidepressant drugs (ADS) are clinically recommended as first-line drugs for chronic treatment of neuropathic pain. Noradrenalin recruited by ADs has been proposed to act through beta2-adrenoceptor (beta2-AR) to lead to their antiallodynic action. However, the precise downstream mechanism by which ADs relieve neuropathic pain remains poorly understood. To mimic human neuropathy resulting from a trauma of peripheral nerves, we used chronic sciatic nerve cuffing in mice. The cuff insertion induced a sprouting of noradrenergic fibers in lumbar dorsal root ganglia (DRG). We then investigated the possible source of endogenous noradrenaline which is used by antidepressant drugs to alleviate neuropathic allodynia. For this purpose, noradrenergic lesions were done with guanethidine and demonstrated that the antiallodynic effect of TCA is mediated through the peripheral nervous system. More particularly, ADs-recruited noradrenaline acts within DRG, on beta2-AR which is expressed by non neuronal cells. These findings reveal a novel anatomomolecular substrate for the antiallodynic action of antidepressants. Our results also showed that the stimulation of beta2-AR leads to decrease of mRNA overexpression of membrane-bound form of TNF-alpha production which is increased by neuropathic pain. This effect was confirmed at protein level. Our findings suggest that ADs act by a peripheral beta2-AR-dependent mechanism targeting TNF-alpha-containing non neuronal cells, which prevents maintenance of neuropathic pain and may offer novel opportunities for management of painful neuropathies.This work was supported by CNRS (contract UPR3212), Université de Strasbourg and Neurex.

P3.136

What are the effects of a lateral fluid percussion injury (LFPI) generated by 1.6-1.8 atm or 2.5-3.0 atm on different brain surrogate markers in rats?

Lieutaud, T. (Villeurbanne)[1], Balanca, B. (Lyon)[2], Bapteste, L. (Lyon)[3], Carrillon, R. (Lyon)[4], Maucler, C. (Lyon)[5], Georges, B. (Lyon)[6], Bonnet, C. (Lyon)[6], Marinesco, S. (Lyon)[7], Bezin, L. (Lyon)[6]
[1]Université Claude Bernard Lyon 1, Crnl, Cnrs Umr 5292, Inserm U 1028, Eequipe Tiger - Bât Dubois, Villeurbanne, France, [2]Centre de Recherche en Neuroscience de Lyon, Equipe WAKE, Lyon, France, [3]Hospices Civils de Lyon, Lyon, France, [4]Hospices Civils de Lyon, Hôpital Neurologique Pierre Wertheimer, Service d'anesthésie réanimation, Lyon, France, [5]Centre de Recherche en Neuroscience de Lyon, Equipe Wake, Lyon, France, [6]Centre de Recherche en Neuroscience de Lyon (CRNL), Equipe TIGER, Lyon, France, [7]Centre de Recherche en Neuroscience de Lyon (CRNL), Equipe WAKE, Lyon, France

Objectives: LFPI is known to generate brain lesions, evidenced by behavioral, histologic, and MRI alterations, accompanied by inflammatory processes. Very few studies have however explored how these alterations combine together. This study was aimed at characterizing how these alterations are developed over time when TBI is modeled by a 1.6-1.8 atm or 2.5-3.0 atm LFPI.
Methods: Sprague Dawley male rats (300-350g) were subjected to LFPI on the left parietal cortex, and then distributed, in a first set of experiments, into four groups: (1) to monitor spatial learning and memory and anxiety, (2) to determine tissue levels of transcripts encoding inflammatory genes using cRT-qPCR, (3) to monitor potential damage to the white matter in corpus callosum and thalamus using Diffusion Tensor Imaging (DTI), and (4) to perform immune-histological studies using antibodies directed against GFAP, NeuN, and CD11b at different times post-injury with computer-assisted counting procedures to evaluate cell density. A group of Sham animals served as reference. A group of animal subjected to a 2.5-3.0 atm LFPI sustained T2 and DTI MRI under isoflurane anesthesia.
Results: LFPI of 1.6-1.8 atm and 2.5-3.0 atm correspond respectively to a mild and moderate to severe brain injury, with 1% and 20% mortality rates respectively. In rats subjected to 1.6-1.8 atm, transient differences in anxiety-like behavior only were found between Sham and LFPI rats. Massive transient increase (50- to 150-fold increase) in inflammatory transcript levels were also measured in the ispilateral side whatever the brain region analyzed, basal levels being recovered by day 7. In both groups of rats subjected either to mild and severe LFPI, T2-MRI analysis revealed hematoma at the level of internal capsule increasing with the severity of the trauma. By contrast, slight differences were measured using DTI at 2.5-3.0 atm LFPI only. All rats subjected to LFPI exhibited, in the cortical zone, a reduced number of neurons.
Conclusion: A 1.6-1.8 atm LFPI can be considered as a model for mild trauma while 2.5-3.0 atm LFPI corresponds to a moderate to severe trauma considering the consequences on mortality rate, DTI and behavior. However, LFPI induced massive inflammatory responses independently of the severity.

P3.137

Neural activation induced by an odour mixture perceived elementally or configurally by the newborn rabbit

Schneider, N. (Dijon)[1], Datiche, F. (Dijon)[1], Royer, C. (Dijon)[1], Saidi, M. (Dijon)[1], Ferreira, G. (Bordeaux)[2], Thomas-Danguin, T. (Dijon)[1], Coureaud, G. (Dijon)[1]
[1]Centre des Sciences du Goût et de l'Alimentation, UMR 6265 CNRS, 1224 INRA, Université de Bourgogne, Dijon, France, [2]Laboratoire de Nutrition et Neurobiologie Intégrée, Bordeaux, France

Perception of odours plays a crucial role in mammals facilitating interindividual communication, food choice and detection of danger, even from early in life (e.g. mother-young communication). However, little is known about the brain processing and perception of odorants in mixtures, which is by far the more common situation in life. To better understand how the brain processes odorants in mixtures, we used the newborn rabbit as a model. Rabbit pups display a clear sucking behaviour in response to the mammary pheromone (MP, the single molecule 2MB2) carried in the milk of lactating rabbit females. The MP also promotes associative conditioning and very rapid acquisition of any novel odorant paired with it. Our previous results showed that after MP-induced conditioning to ethyl maltol (odorant A), pups do not respond to odorant B (ethyl isobutyrate) or to the AB mixture (70/30 ratio), but they respond to A and to the A´B´ mixture (32/68 ratio). These results suggest configural perception of AB and elemental perception of A´B´. Here, we investigated by c-Fos immunodetection the neural activation induced by AB or A´B´ in the olfactory bulbs and central regions of 4-day-old rabbit pups, 24h after a single MP-conditioning to odorant A. Our hypothesis was that activated regions, or level of activation of certain regions, should differ depending on the mode of perception of odorants A and B in the mixture. Regarding forebrain regions, preliminary results show a stronger activation in the anterior and posterior piriform cortex, tenia tecta, ventral hippocampus, basal and medial amygdala in pups exposed to A´B´ compared to pups exposed to AB. Within the anterior piriform cortex, the labelled cells are not homogeneously distributed, according to the stimulus. Analyses of the olfactory bulb are in progress. These first results indicate that the same odorants may induce different neural forebrain

activation of rabbit neonates, and consecutively different behavioural responsiveness, depending on whether the odorants are perceived as elements in the mixture, or involved in a mixture configuration.

P3.138

Functional organization of synaptic inputs on mouse cerebellar Golgi cells related to Zebrin band patterns

Valera, A.M. (Strasbourg)[1], Dupont, J.-L. (Strasbourg)[1], Cruveiller, H. (Strasbourg)[1], Binda, F. (Strasbourg)[1], Pawlowski, S.A. (Montreal)[2], Rothstein, J. (Baltimore)[3], Poulain, B. (Strasbourg)[1], Isope, P. (Strasbourg)[1]
[1]Institut des Neurosciences Cellulaires et Intégratives, CNRS UPR 3212, Dépt neurotransmission et sécrétion neuroendocrine, Strasbourg, France, [2]McGill University, Montreal, Canada, [3]John Hopkins University, Dept of Neurology, Baltimore, United States

Despite its apparent uniformity, the mammalian cerebellar cortex is highly compartmentalized into distinct functional and anatomical modules. The expression of zebrins, a family of biochemical markers that are confined to subsets of Purkinje cells, results in highly reproducible parasagittal bands of high and low immunoreactivity within the cerebellar cortex. Recent studies suggest that cerebellar afferences and patterns of zebrin bands reflect a common organizational scheme within the cerebellum, involving a modular treatment of the information. We postulate that this columnar information processing will lead to a specific regulation of functional synaptic organization. Among the cell types that may contribute to this processing, we found that the distribution of Golgi cell (GoCs) subtypes, defined by the expression of the GlyT2 transporter and neurogranin, varies with the Zebrin II/aldolase C expression. We then investigated how synapses onto GoCs are affected by the segregation of incoming information, focusing on Granule cell (GC) to GoCs connection. Using two transgenic strains of mice, one expressing GFP under the control of the Glyt2 promoter, allowing the localization of this subset of GoCs, and a second with GFP under the control of the EAAT4 promoter, allowing visualization of Zebrin II bands during the experiment, we studied the spatial organization of GC inputs onto GoCs in relation to Zebrin II bands. By combining whole cell patch clamp of GoCs and RuBi-glutamate uncaging, we have established a map of functional connectivity in the vermal region of the mouse cerebellum. Preliminary results showed that the probability of finding functional GC-GoCs connection is much higher when cells belong to the same zebrin II band, independently of the position of GoCs in the band, suggesting a modular organization of information processing in the cerebellar cortex.

P3.139

Paralbumin-expressing GABAergic neurons gate sensory perception in mouse barrel cortex

Sachidhanandam, S. (Lausanne)[1], Sreenivasan, V. (Lausanne)[1], Kyriakatos, A. (Lausanne)[1], Kremer, Y. (Lausanne)[1], Petersen, C.C.H. (Lausanne)[1]
[1]Ecole Polytechnique Federale de Lausanne, Brain Mind Institute, Lausanne, Switzerland

Sensory percepts are thought to be generated by the activity of neurons in the cerebral cortex. However, a causal description of the synaptic interactions driving the neural circuit computations underlying any learned, goal-directed sensorimotor transformation is currently lacking. Here, we explore the role of primary sensory cortex in the simplest possible perceptual task, stimulus detection. Mice were trained to detect single brief whisker stimuli and to report perceived stimuli by licking to obtain a reward. Optogenetic stimulation of the primary somatosensory barrel cortex readily substituted for whisker stimulation in both learning and execution of the detection task. The barrel cortex therefore plays a causal role in this behavior. Whole-cell membrane potential recordings from layer 2/3 barrel cortex neurons during task performance revealed that both synchronized, slow

oscillatory brain states and desynchronized brain states were compatible with high performance. Whisker deflection evoked an early reliable sensory response, indifferent to behavioral outcome, encoded through cell-specific reversal potentials and sparse reliable action potential firing in excitatory neurons. A secondary late depolarization in excitatory neurons was larger on hit trials compared to misses, likely driven by reduced action potential firing in parvalbumin-expressing GABAergic neurons on hit trials. Optogenetic inactivation of barrel cortex, furthermore, revealed a causal role for the late depolarization in the detection task. Our data reveal dynamic stimulus processing in sensory cortex during task performance, with an early sensory response reliably encoding the stimulus and later secondary activity contributing to driving the subjective percept as reported through licking.

P3.140

V1 neurons can distinguish between motion in the world and visual displacements due to eye movements: a microsaccade study

Troncoso, X. (Gif sur Yvette)[1,2], Najafian Jazi, A. (Phoenix)[2,3], Otero-Millan, J. (Phoenix)[2,4], Macknik, S. (Phoenix)[2], Martinez-Conde, S. (Phoenix)[2]
[1]CNRS-UNIC, Gif sur Yvette, France, [2]Barrow Neurological Institute, Phoenix, United States, [3]Arizona State University, Tempe, United States, [4]Univeridade de Vigo, Vigo, Spain

How do perceptual systems differentiate between self-motion and motion in the world? This problem has special importance in vision: we can easily distinguish between real world motion and a comparable displacement of the image over the retina due to an eye movement, despite equivalent retinal stimulation. How the brain performs this operation or which brain areas are involved remains unknown.
Here we recorded from single neurons in awake monkey area V1 and compared the responses triggered by microsaccades (small-magnitude saccades that occur while attempting to fixate) to the responses induced by stimulus motion mimicking microsaccades. Our experiments allowed us to determine,
1) whether V1 neurons can differentiate between internal and external motion, and
2) the contribution of retinal versus non-retinal sources to microsaccade-driven neuronal responses in V1.
Because microsaccades displace receptive fields (RF) over an image, the interplay between RF and visual stimulus might fully explain V1 responses to microsaccades. If so, real microsaccades should elicit the same responses as stimulus motion that mimics microsaccades. Alternatively, responses to real and simulated microsaccades might be different, indicating that responses to real microsaccades include inputs from both retinal and non-retinal sources (such as corollary discharge proprioceptive signals, and/ or global motion integration).
We found that neuronal responses to real microsaccades were generally biphasic: a quick increase over baseline was typically followed by a smaller and slower trough below baseline, whereas responses to simulated microsaccades included an excitatory peak but no trough. These findings suggest that excitatory responses to real microsaccades result from the displacement of the visual stimulus over the classical RF, with the subsequent inhibition reflecting non-retinal sources. The differential neural response to real versus simulated microsaccades further indicates that V1 neurons can distinguish between internally and externally generated motion (visual displacements due to eye movements versus actual motion in the world). These findings help to delineate the role of V1 in information processing, visual stability, and/or perceptual suppression during microsaccades.

P3.141

Characterization of olfactory abilities of Thy-Tau22 mice

Martel, G. (Paris)[1], Blum, D. (Lille)[2], Lepage, Y. (Lille)[2], Simon, A. (Paris)[1], Kalainathan, S. (Paris)[1], Geny, D. (Paris)[1], Buée, L. (Lille)[2], Viollet, C. (Paris)[1], Epelbaum, J. (Paris)[1]
[1]Centre de Psychiatrie et Neurosciences, INSERM, Université Paris Descartes, Paris, France, [2]UMRS 837 Université Lille2, Lille, France

Impaired olfaction is a landmark of the early stages of Alzheimer's disease (AD), sometimes detected before cognitive defects. Since
1) cognitive impairments and Tau pathology progression are correlated in both AD patients and experimental models and
2) Tau pathology is enriched in olfactory structures of definite AD cases (Attems et al, 2006), we took advantage of a relevant experimental model of AD (THY-Tau22; Schindowski et al. 2006) displaying

progressive development of Tau pathology, to study its impact on olfactory performances. Wild-type and transgenic (Tg) mice were studied at three time points: at the age of 4 months when hippocampal Tau pathology is starting and associated with slight cognitive impairments, at mid-course (7 months) and at 12 months when Tau pathology is maximal and cognition is markedly altered. Hyperphosphorylated Tau was detected early in the anterior olfactory nucleus and sharply accumulated from 7 to 12 months in the Tg mice but it only appeared at 12 months in the olfactory bulb. Spontaneous olfactory behavior and fine olfactory discrimination abilities were evaluated in 4-, 7- and 11-month mice. A strong olfactory deficit appeared with aging without major difference between WT and Tg mice. In addition, at each time-point, Tg mice learned the association between odors and reward faster than WT mice. Studies are in progress to characterize molecular and cellular changes underlying the behavioral differences. Funded by Inserm, ANR-10-MALZ-003-01 SOMADOLF, Fondation Recherche Plan Alzheimer and DIM Cerveau et Pensée.

P3.142

Maternal separation delays C fiber efficiency onset in spinal cord and alters descending inhibitory pain pathway in rats

Juif, P.-E. (Strasbourg)[1], Salio, C. (Turin)[2], Darbon, P. (Strasbourg)[1], Merighi, A. (Turin)[2], Lelievre, V. (Strasbourg)[1], Poisbeau, P. (Strasbourg)[1]
[1]CNRS & Université de Strasbourg, Molecular Determinants of Pain, Strasbourg, France, [2]University of Turin, Dipartimento di Morfofisiologia Veterinaria, Turin, Italy

Maternal separation (MS) is an experimental model of psychosocial stress that reflects human prematurity. Whereas this paradigm has been described to be associated with visceral pain hypersensitivity, drugs addiction and social isolation at adulthood, the consequences during the early developmental period have been poorly investigated.This period of time is particularly relevant since the development of nociceptive processing of rats is not yet achieved at birth in rodents. Indeed, until the second post natal week, C fiber inputs exhibit weak excitation, are relatively sparse and A fibers take in charge the nociceptive signaling. On the other hand, descending inhibitory pathways and, among them, diffuse noxious inhibitory controls (DNIC) appear and are fully efficient at 21 days post natal.
Here, we investigated the effect of MS on the development of spinal nociceptive circuits. It was performed using *in vivo* electrophysiological recordings of dorsal horn spinal cord neurons in anesthetized rats, behavioral test and *in vitro* technique (PCR) for specific maker measurement. We first observed that the functional establishment of C fibers was delayed and this was associated with an alteration in the expression of several growth factors. In addition and compared to control animals, DNIC was absent in MS rats after P21. This impairment is currently investigated in order to identify possible molecular substrates responsible for this lack of inhibitory control.

P3.143

TAFA4, a C-Low threshold mechano receptor derived chemokine-like protein, modulates the excitability and synaptic transmission in mice substancia gelatinosa neurons

LeFeuvre, Y. (Bordeaux)[1], Delfini, M.-C. (Marseille)[2], Mantilleri, A. (Marseille)[2], Landry, M. (Bordeaux)[1], Moqrich, A. (Marseille)[2]
[1]IINS-CNRS UMR 5297, Université de Bordeaux II, Bordeaux, France, [2]IBDML, CNRS UMR 6216, Aix-Marseille University, Marseille, France

C-LTMRs (C-Low Threshold Mechano Receptors) are unique among C-unmyelinated primary sensory neurons. These neurons convey two opposite aspects of touch sensation; a sensation of pleasantness and a sensation of injury-induced mechanical pain. How this dual function is achieved is still largely unknown. Recent work has identified a new secreted peptide (TAFA4, from the TAFA chemokine family) that is specifically expressed by this population of sensory neurons. Moreover, genetic models of TAFA4 deficient mice, demonstrated that TAFA4 exhibits anti-nociceptive properties in inflammatory pain models.
In the present work, we have investigated the effects of TAFA4 in the operation of spinal nociceptive networks. Using patch-clamp approaches on transverse spinal slices, we have compared the intrinsic and synaptic properties of dorsal horn spinal neurons in wild type and TAFA4 deficient mice. We show that,
i) TAFA4 deficient mice display enhanced spiking and rebound spiking activity compared to neurons from their wild type littermates. This difference in firing properties can be correlated with an increase in low-threshold T-Type current density and a decrease in slowly inactivating low threshold outward current in TAFA4-/- mice.
ii) in response to local stimulation of attached dorsal roots, TAFA4-/- mice display enhanced paired pulse facilitation compared to wild type animals.
We then investigated the effects of bath applied recombinant TAFA4 on lamina II neuron electrophysiological properties in TAFA4-/- animals. We show that,
i) in most lamina II neurons, 20nM human recombinant TAFA4 elicits a slowly inactivating outward current. This current is blocked by 4 aminopyridine but not by Tetra ethyl ammonium, suggesting A-Type current pharmacology. This effect was not observed when other TAFAs (TAFA2 and TAFA5) were applied on the preparations.
ii) TAFA4 induces a decrease in paired pulse facilitation of EPSCs evoked by stimulation of dorsal roots. Hence, bath superfusion of TAFA4 restores wild type properties in neurons of TAFA4-/- animals.
By providing first cellular insights in the mechanism of action of TAFA4, these data will greatly enhance our understanding of the role of C-LTMRs in modulating pain perception.

P3.144

Functional interaction between the cerebellum and the hippocampus during spatial navigation

Rochefort, C. (Paris)[1], Zhang, L. (Paris)[1], Lefort, J. (Paris)[1], Jarlier, F. (Paris)[1], Rondi-Reig, L. (Paris)[1]
[1]*CNRS-UPMC, Neurobiologie des Processus Adaptatifs, UMR 7102, ENMVI, Paris, France*

Spatial navigation is a complex function that requires the integration of heterogeneous information to build a coherent representation of the external world and drive optimal goal-directed behavior. A key neural substrate enabling such representation is the hippocampus, which contains pyramidal cells described as place cells. Each place cell fires for a restricted region of the environment. Both external (visual, olfactory, auditory, and somatosensory) cues and self-motion (vestibular, proprioceptive and optic flow signals) cues control place cell firing. This multimodal integration suggests that a large network of cortical and subcortical structures interacts with the hippocampus for navigation. Determining the functional architecture of such a network is thus essential to our understanding of how the spatial code is generated. We aim at deciphering the role of the cerebellum in this network. Cerebellum is classically described as a structure involved in motor function. However, using L7PKCI transgenic mice in which no motor deficit was detected despite a lack in cerebellar plasticity; we demonstrated that a deficit in cerebellar long-term depression (LTD) leads to a dysfunction of the hippocampal place cells as well as impaired goal-directed navigation abilities. This deficit was specifically observed when mice had to rely on self-motion information. Our findings thus revealed that during navigation, the cerebellum communicates with the hippocampus to shape the spatial code. Our current investigation aims at further investigating this functional interaction. For this purpose we performed *in vivo* hippocampal and cerebellar electrophysiological recordings during goal-directed navigation using either external or self-motion information. Both hippocampal place cell properties and synchronized activity between the cerebellum and the hippocampus are being analyzed in wild type and two lines of transgenic mice, the L7PKCI and the L7PP2B, that respectively lack LTD and LTP at

parallel fiber-Purkinje cell synapses. Using this multidisciplinary approach, we hope to understand how the cerebellum interacts with the hippocampus to optimize spatial navigation.

P3.145

The μ-opioid receptor agonist fentanyl induces the activation of a spinal NR2B NMDA receptors-P38 pathway leading to exaggerated post-operative pain in rat

Rivat, C. (Montpellier)[1], Harada, F. (Seattle)[2], Xu, M. (Seattle)[2], Cahana, A. (Seattle)[2], Richebé, P. (Seattle)[2]
[1]*Université de Montpellier 2, Institut des Neurosciences de Montpellier (INM) INSERM U 1051, Montpellier, France,* [2]*University of Washington, Anesthesiology and Pain Medicine, Seattle, United States*

The opioid analgesics may worsen pain especially after surgery. Besides the activation of the NMDA receptor, little is known about the contribution of central sensitization in the paradoxical effects of opioids. Here we investigated whether activation of the spinal MAPK kinase p38, known to be implicated in spinal sensitization initiated by chronic pain, is also triggered by the μ-opioid receptor agonist fentanyl, administered in rat with or without post-operative pain. Rats received 4 fentanyl (4x80 μg/kg, s.c.) or saline injections administered at 15 min intervals. After the second injection, some groups of animals underwent incision on the left hindpaw. Thermal and mechanical sensory hypersensitivities were evaluated after the surgery. In animals without incision, fentanyl induced analgesia followed by sensory hypersensitivity, as previously described. Fentanyl also enhanced plantar incision-induced pain hypersensitivity. On D1, increased activity of the dorsal horn neurons (DHN) in response to mechanical stimulation was recorded in incision and fentanyl groups as compared to saline animals. DHN activity was dramatically enhanced in fentanyl-incisioned animals. In rats with or without incision, an increased p-p38 immunoreactivity was observed in the spinal dorsal horn of fentanyl-treated animals as compared to saline or incision-treated animals, alone. The intrathecal injection of p38 inhibitor SB 203580 (10 μg) reduced the mechanical pain hypersensitivity observed in fentanyl-treated rats with or without incision, p-p38 immunoreactivity and DHN hyperexcitability measured in fentanyl-incisioned animals. Similar effects were observed with the NMDA receptor antagonist ketamine (10 mg/kg, s.c.) injected at the time of surgery. NMDA receptor activation was confirmed by the increased phosphorylation of the NR2B subunit observed in incision group and more extensively in fentanyl groups as compared to saline animals. These data indicate that the NR2B NMDA receptors-p38 pathway mediates fentanyl enhancement of surgery-induced pain hypersensitivity suggesting common cellular pathways between opioid and chronic injury-induced pain hypersensitivity.

P3.146

The human OPA1^{delTTAG} mutation induces premature age-related systemic neurodegeneration in mouse

Sarzi, E. (Montpellier)[1], Angebault, C. (Angers)[2], Seveno, M. (Montpellier)[1], Gueguen, N. (Angers)[2], Bielicki, G. (Saint-Genès Champanelle)[3], Boddaert, N. (Paris)[4], Rigau, V. (Montpellier)[5], Bonnefont, A.-L. (Montpellier)[6], Cazevieille, C. (Montpellier)[7], Renou, J.-P. (Saint-genès Champanelle)[8], Delettre, C. (Montpellier)[1], Brabet, P. (Montpellier)[1], Hamel, C. (Montpellier)[1], Reynier, P. (Angers)[2], Lenaers, G. (Montpellier)[1]
[1]*Institut des Neurosciences de Montpellier, Montpellier, France,* [2]*Inserm U 1083, Angers, France,* [3]*Inra, UR 370, Saint-Genès Champanelle, France,* [4]*Hôpital Necker Enfants-Malades, Paris, France,* [5]*Hopital Guy de Chauliac, Montpellier, France,* [6]*Inserm U 844, Montpellier, France,* [7]*Centre de*

Recherche en Imagerie Cellulaire, Montpellier, France, [8]Inra UR 370, Saint-genès Champanelle, France

Dominant Optic Atrophy (DOA) is a rare inherited optic nerve degeneration caused by mutations in the mitochondrial fusion gene OPA1. Recently, the clinical spectrum of DOA has been extended to frequent syndromic forms, exhibiting various degrees of neurological and muscle impairments frequently found in mitochondrial diseases. Although characterized by a specific loss of retinal ganglion cells, the DOA pathophysiology is still poorly understood. We generated an Opa1 mouse model carrying the recurrent Opa1^{delTTAG} mutation, found in 30% of all DOA patients. We show that this mouse displays multi-systemic poly-degenerative phenotype, with a presentation associating signs of visual failure, deafness, encephalomyopathy, peripheral neuropathy, ataxia and cardiomyopathy. Moreover, we found premature age-related axonal and myelin degenerations, increased autophagy and mitophagy, and mitochondrial supercomplex instability, preceding degeneration and cell death. Thus, these results support the concept that Opa1 protects against neuronal degeneration and open new perspectives for the exploration and the treatment of mitochondrial diseases.

P3.147

Vagus nerve stimulation and the cholinergic anti-inflammatory pathway: a potential new therapeutic approach in inflammatory bowel diseases

Bonaz, B. (Grenoble)[1,2,3], Sinniger, V. (Grenoble)[3], Picq, C. (La Tronche)[4], Pellissier, S. (Grenoble)[3], Clarençon, D. (La Tronche)[4]
[1]Inserm U 836, Grenoble Institut des Neurosciences, Grenoble, France, [2]CHU de Grenoble, Clinique Universitaire d'Hépato-gastroentérologie, 38, Grenoble, France, [3]Inserm U 836, Grenoble Institut des Neurosciences, 38, Grenoble, France, [4]IRBA Institut de Recherche Biomédiale des Armées, 38, La Tronche, France

Background: Brain and gut communicate through the autonomic nervous system (ANS), represented by the sympathetic and parasympathetic nervous systems. The vagus nerve (VN), a major component of the ANS, plays a key role in the neuroendocrine-immune axis to maintain homeostasia through its afferents (via a modulation of the hypothalamic pituitary adrenal axis; HPA) and through its efferents (via a modulation of the cholinergic anti-inflammatory pathway; CAP) (**1**). High frequency VN stimulation (VNS) of afferents is used for the treatment of drug-resistant epilepsy and depression in Humans. Low frequency VNS (5Hz) of efferents can activate the CAP and decrease the secretion of TNFα, a proinflammatory cytokine. Our group explores how this anti-inflammatory effect of VNS could be used in inflammatory bowel diseases (IBD) represented by ulcerative colitis and Crohn's disease. *Team results*: The ANS has been shown to be imbalanced in irritable bowel syndrome and IBD (**2**). Consequently, VNS could restore the ANS equilibrium in these disorders. A chronic 5Hz VNS performed in a rat model of colitis was able to reduce body weight loss, a classical inflammatory parameter in IBD patients, and decrease TNFα and myeloperoxydase (a marker of leukocytes infiltration) in the colonic mucosa (**3**). We performed the first fMRI study of acute VNS in rodents showing that even low-frequency VNS at 5Hz, known to theoretically activate vagal efferents, was also able to have central effects through afferents (**4**). Low-frequency VNS could thus both have a peripheral (CAP) and a central effect (vago-vagal positive loop; HPA axis activation; modification of the central ANS), modulating the sympatovagal balance and thus inflammation (**5**). **Conclusion:** These data argue for an anti-inflammatory role of a chronic VNS and provide potential therapeutic applications for IBD patients. We are currently running a pilot study of VNS in patients with moderate to severe Crohn's disease (ClinicalTrials.gov Id: NCT01569503).

1.Bonaz B and Bernstein CN **Gastroenterology** 2013.144:36-49**.**
2.Pellissier S, *et al* **Psychoneuroendocrinology** 2010.35:653-62.
3.Meregnani J *et al* **Auton Neurosci** 2011.24;160:82-9.
4.Reyt S, Picq C *et al* **Neuroimage** 2010.1;521456-64.
5.Bonaz B *et al* **Neurogastroenterol Motil** 2013.25:208-21.

P3.148
Decrease of correlations at stimulus onset

Deneux, T. (Marseille)[1], Masson, G.S. (Marseille)[1], Vanzetta, I. (Marseille)[1]
[1]*Institut de Neurosciences de la Timone (INT), CNRS UMR 7289 - Aix Marseille Univ., Campus de la Timone, Marseille, France*

In absence of external sensory input, sensory brain areas display a spontaneous activity whose structure is still incompletely characerized and whose role is still debated. In particular, the question exists how the structure of this spontaneous activity relates to that observed during the processing of sensory inputs. In order to address this issue, we compared the activity pattern observed in absence of stimulation with those observed during the response to visual stimuli of different contrast. Our experiments were performed using optical imaging of voltage-sensitive dye in the V4 visual area of an awake macaque monkey, over a region of several millimeters. Our results show that during the stationary period of a sustained stimulation the correlations between different cortical locations are significantly lower than the correlation levels observed during the spontaneous activity. Surprisingly, this was true even for contrast values that were too weak to evoke any detectable activity in the raw responses. Our results could be explained by the presence of a mechanism that attenuates the highly correlated aspects of spontaneous neuronal activity patterns (global fluctuations and cortical waves) when a sensory input arrives, in a way that depends non-linearly on the amplitude of the locally evoked response.

P3.149
Spike estimation from calcium optical recording

Lakner, T. (Marseille)[1], Deneux, T. (Marseille)[1], Kaszas, A. (Marseille)[2], Vanzetta, I. (Marseille)[1]
[1]*Institut de Neurosciences de la Timone (INT), CNRS UMR 7289 - Aix Marseille Univ., Campus de la Timone, Marseille, France,* [2]*Institut de Neurosciences des Systèmes, Inserm UMR 1106 - Aix Marseille Université, Marseille, France*

Since optical methods allow to record simultaneously and at high resolution from a large population of neurons, their use in neuroscience research has remarkably increased over the last few decades. Examples of recent technical breakthroughs in the field of imaging but also in the development of optical probes are 2-photon microscopy, cell-permeant dyes, opto-genetics and virus transfection. Despite this progress, to-date spiking activity cannot yet be probed directly by optic methods. Rather, spikes are generally inferred from recordings of the changes in fluorescence intensity of intracellular calcium indicators. Despite the large fractional changes in calcium fluorescence transients (in the order of 10%) induced by an action potential, inferring the correct number and timing of individual spikes in a given recording is difficult because of the long decay time of the fluorescent transients (~1s). As a result, calcium transient evoked by successive spikes overlap largely, and the backward problem of estimating spike trains from calcium fluorescence recordings becomes computationally intractable (the computation times increases exponentially with the number of time instants). Previous approaches proposed to address this problem have relied on approximations or on heuristics valid only for very specific dataset, and yet were computationally expensive.
We have developed a novel algorithm based on a novel filtering technique, that solves this problem without making any approximation. Moreover, it has a computational efficiency that allows real-time estimation. Here, we report the results of tests we performed on a number of simulated and real data with different characteristics of spiking activity and noise.

P3.150
Contribution of the cerebellum to sensory-motor thalamocortical activity during whisking

Popa, D. (Paris)[1], Spolidoro, M. (Paris)[1], Proville, R. (Paris)[1], Guyon, N. (Paris)[1], Belliveau, L. (Paris)[1], Lena, C. (Paris)[1]
[1]Ecole Normale Supérieure - Neuroscience Section, IBENS, CNRS UMR 8197, INSERM U 1024, Cerebellum Group, Paris, France

The cerebellum is a structure essential for the control of movement, but its contribution to motor cortex dynamics remains largely unknown. We examined the influence of the cerebellum on cortical activity in rats during haptic exploration. To this end, we recorded the field and cellular activity in the motor cortex, sensory cortex and motor thalamus before and after inactivation of the cerebellum. Our results show that the inactivation of the cerebellum decreases the firing rate of cells in the thalamus and in the motor cortex of the vibrissae during episodes of free whisking in the air. We also found a decreased spectral coherence between the motor cortex and the sensory cortex in the gamma band during whisking. Our results show that the cerebellum is a major determinant of the sensory-motor cortical interactions during motor activation.

P3.151
Astroglial networks promote efficient hippocampal bursts

Chever, O. (Paris)[1], Dossi, E. (Paris)[1], Rouach, N. (Paris)[1]
[1]Collège de France, CNRS UMR 7241, INSERM U 1050, CIRB, Paris, France

Astrocytes play crucial roles in brain physiology by dynamic interactions with neurons. They form plastic and extensive networks mediated by gap junction channels. It has recently been shown that astroglial networks limit neuronal network activity. However, it is currently unclear how astroglial networks influence neuronal excitability and population activity. To investigate how astroglial assembly regulates neuronal network activity, we performed electrophysiological recordings in hippocampal slices (CA3 and CA1 areas) from mice with disconnected astrocytes, in which both astroglial gap junction forming proteins, connexin 30 (Cx30) and connexin 43 (Cx43), are knocked out (GFAP-Cre $Cx30^{-/-}Cx43^{fl/fl}$, dKO). Synchronized excitatory discharges were recorded in an acute pharmacological model of epileptic-like activity. In this model, spontaneous bursting discharges are characterized by a sharp and large amplitude shift in field potential, generated by a major depolarization and firing of all putative neurons. Pyramidal cells depolarized up to 40 mV during seconds and such depolarization is followed by a long-lasting undershoot of the membrane potential. We found that the frequency of bursting activity in dKO hippocampal slices is drastically increased. However, the duration of neuronal depolarizing bursts is severely reduced. Furthermore, pyramidal neurons are more depolarized due to an increase of synaptic bombardment. This suggests that increased synaptic background promoting resting membrane potential depolarization facilitates triggering of neuronal bursts, but compromises the strength of synchronized events. To investigate local dynamics of bursts generation, we performed multielectrodes arrays recordings in the CA3 area of the hippocampus. In slices from dKO mice, bursting activity in the CA3 region is generated within a more restricted area, indicating that the number of neurons to be recruited is highly decreased. Altogether, these results indicate that gap junction-mediated astroglial networks strengthen the coordination of neurons during synchronized events.

P3.152
Cholinergic neurons in the macaque sensory cord: characterization of a newly-identified population and a potential substrate for pain therapy

Cordero-Erausquin, M. (Strasbourg)[1], Pawlowski, S. (Strasbourg)[1,2], Gaillard, S. (Strasbourg)[1], Ghorayeb, I. (Bordeaux)[3], Ribeiro-da-Silva, A. (Montreal)[2], Schlichter, R. (Strasbourg)[1,4]
[1]Institut des Neurosciences Cellulaires et Intégratives, Strasbourg, France, [2]McGill University, Departments of Pharmacology & Therapeutics and Anatomy & Cell Biology, Montreal, Canada, [3]Université de Bordeaux 2, Institute of Neurodegenerative Disease, CNRS-UMR 5293, Bordeaux, France, [4]Université de Strasbourg, Strasbourg, France

Endogenous acetylcholine (ACh) is a well-known modulator of nociceptive transmission in the spinal cord of rodents. It arises mainly from a sparse population of cholinergic interneurons located in the dorsal horn of the spinal cord. This population was thought to be absent from the spinal cord of monkey, what might suggest that spinal ACh would not be a relevant clinical target for pain therapy. In humans, however, pain responses can be modulated by spinal ACh, as evidenced by the increasingly used analgesic procedure (for postoperative and labor patients) consisting in the epidural injection of the acetylcholinesterase inhibitor neostigmine. The source and target of this ACh remains yet to be elucidated. In this study, we used an immunolabeling for choline acetyltransferase to demonstrate for the first time the presence of a plexus of cholinergic fibers in laminae II-III of the dorsal horn of the macaque monkey. Moreover, we show the presence of numerous cholinergic cell bodies within the same laminae and compared their density and morphological properties to those previously described in rodents. An electron microscopy analysis demonstrates that cholinergic boutons are presynaptic to dorsal horn neurons as well as to the terminals of sensory primary afferents, suggesting that they are likely to modulate incoming somatosensory information. Our data suggests that this newly-identified dorsal horn cholinergic system in monkeys is the source of the ACh involved in the analgesic effects of epidural neostigmine, and could be more specifically targeted for novel therapeutic strategies for pain management in humans.

P3.153
Cerebellum and vocal learning in songbird

Pidoux, L. (Paris)[1], Levenes, C. (Paris)[1], Leblois, A. (Paris)[1]
[1]*CNRS, Universite Paris Descartes, UMR 8119, Paris, France*

Human speech is a complex sensorimotor skill and vocal learning is one of the most striking cognitive abilities of the brain. As many other complex motor skills, vocal learning involves the basal ganglia (BG)-thalamo-cortical and the cerebello-thalamo-cortical network in humans. While the BG and cerebellar sub-cortical loops have been shown to interact at least through two different pathways in mammals, the role of their interaction during sensorimotor learning, and in particular during vocal learning, remains undetermined.
Songbirds are one of the few accessible animal models for vocal learning, as they have a specialized portion of their BG-thalamo-cortical circuitry dedicated to song learning. Additionally, a cerebellar projection to the thalamic region adjacent to the song-related thalamic nucleus receiving BG input suggests that BG and the cerebellum may interact during song learning. However, very little is known about song-related circuits in the cerebellum, or about a putative cerebellar function in song learning. We are studying the interactions between BG and cerebellar sub-cortico-cortical loops involved in avian song learning.
In order to determine to what extent the cerebellum is involved in song learning and dissect the cerebellar circuits interacting with thalamic and cortical song-related nuclei, we performed two sets of experiments. On one hand, we explored putative anatomical pathways linking the song control areas in the forebrain to the cerebellum. In particular, we looked for a pathway linking high auditory areas to the relay nuclei of the Pons. On the other hand, we investigated the physiological mechanisms underlying the integration and transfer of cerebellar signals in the BG using electrophysiological recordings of evoked BG activity following electrical stimulation in the cerebellum. Stimulation of the deep cerebellar nuclei evoked fast excitatory responses in BG neurons located in the song-related BG nucleus Area X.

P3.154
A neuro-computational account of economic choices in the human brain

Redoute, J. (Bron)[1,2], Domenech, P. (Bron)[1,3], Dreher, J.-C. (Bron)[1]
[1]*Centre de Neuroscience Cognitive, CNRS UMR 5229, Bron, France,* [2]*CERMEP – Imagerie du Vivant, Lyon, France,* [3]*Ecole Normale Superieure, Département d'Études Cognitives, Paris, France*

Economic decision-making is the selection of actions based on the subjective value one's attribute to its prospective outcome. Such decisions entail at least two stages:
(1) Ordering all available options on a common scale of values (valuation), and
(2) Selecting the option associated with the most valuable outcome (selection).
Although converging evidence link the ventromedial prefrontal cortex (vmPFC) to valuation, we still know very little on the algorithm implementing value-based selection in the human brain. Diffusion models integrating the difference between options values (decision value) predict behavior during economic decisions. Here, we combined model-based fMRI and pattern analyses to tear apart valuation and selection, and map their key computations onto the brain. Fourteen men performed an fMRI paradigm requiring to choose between two gambles, one yielding fruit juice (0.5ml) and the other an erotic picture. Choice probability was a sigmoid function of decision value. A diffusion model correctly captured the relationship between RTs, choice and subjective values, as a set of four parameters: preference, gain, decision threshold and non-decision time. We found that activity in vmPFC correlated with decision value and with model's gain, but not with RTs or the slope of the diffusion process, showing that this brain region encoded the input of the diffusion process. Connectivity increased between vmPFC and a parieto-prefrontal network during choice formation. Activity correlated with the slope of the diffusion process in the right dorsolateral prefrontal cortex (DLPFC), but not in the inferior parietal sulcus (IPS), where activity strongly correlated with RTs. Finally, we decoded the temporal course of information on chosen option from these brain regions. During deliberation, information on the chosen option was constant in vmPFC, while it increased in DLPFC and IPS. DLPFC engagement in selection preceded that of IPS. Taken together, our results demonstrate that economic decision-making is implemented in the human brain as two distinct stages; vmPFC valuation signals drive a diffusion process implementing selection in a parieto-prefrontal network. Our results further suggest that DLPFC integrates decision values, while IPS reads out decision outcome.

P3.155

Functional and anatomical characterization of the two glutamate-gated chloride channel subunits in the honeybee brain (*Apis mellifera*)

Démares, F. (Toulouse)[1], Crattelet, C. (Toulouse)[1], Loeuillet, A. (Toulouse)[1], Bettiol, C. (Toulouse)[1], Raymond, V. (Angers)[2], Armengaud, C. (Toulouse)[1]
[1]Université Paul Sabatier - Toulouse III, Centre de Recherches sur la Cognition Animale, Toulouse, France, [2]Université d'Angers, INRA, Récepteurs et Canaux Ioniques Membranaires, Angers, France

Glutamate acts as an excitatory or inhibitory neurotransmitter, according to species and tissues, and based solely on its receptor. In insects, different studies indicate that glutamatergic transmission is crucial for olfactory learning. A part of this transmission is mediated through glutamate-gated chloride channels (GluCls), specific to invertebrate species. Jones and Sattelle (2006) stated the existence of two spliced-variants, coming from a single gene *amel_glucl*, resulting in two different subunits, Amel_GluCl A and Amel_GluCl B. The aim of this work was to characterize and localize these GluCl subunits in honeybee's brain and analyze their roles on olfactory learning and memory processes. RT-PCR indicated that Amel_GluCl had two alternatives for exon 3; both variants were expressed in honeybee brain. Using antibodies against each variant, immunohistochemical experiments revealed that Amel_GluCl A variant was mainly expressed in neuropiles, and the other variant, Amel_GluCl B mainly somatic (Démares et al 2013). The proteins were differentially localized in the central nervous system of honeybee, thus suggesting a different functional involvement. This had been further analyzed by behavioral experiments coupled with RNA-interference technique.
Based on Amel_GluClα sequences to design specific small-interfering RNA (siRNA), we repress each subunit's expression. Consequences of these siRNA-mediated knockdowns were analyzed at protein level and behavioral level. Injections of siRNA were done through ocellar tract. Biochemical and behavioral effects of siRNAs were both transient. siRNA against each variant was injected alone 24h before a five-trials olfactory conditioning of the proboscis extension reflex (PER). Both siRNAs induced a decrease in conditioned odor retrieval 1h after learning; moreover, siRNA against somatic variant

induced a faster generalization to new odors. Overall, these results indicate that glutamatergic transmission mediated by GluCl channels is necessary for the olfactory memory retrieval, and may have a role in salience of conditioned stimuli compared to new odors.

P3.156

The dorsal paragigantocellular reticular nucleus facilitates paradoxical (REM) sleep genesis by mean of the inhibition of adrenergic and noradrenergic neurons

Clement, O. (Lyon)[1,2], Libourel, P.-A. (Lyon)[1,2], Arthaud, S. (Lyon)[1,2], Fort, P. (Lyon)[1,2], Luppi, P.-H. (Lyon)[1,2]
[1]CRNL, INSERM U 1028, CNRS UMR 5292, Lyon, France, [2]Université Claude Bernard Lyon 1, Lyon, France

The dorsal paragigantocellular reticular nucleus (DPGi) contains GABAergic neurons specifically active during paradoxical sleep (PS-on neurons). Previous data indicate that these neurons are responsible for the cessation of activity during PS of the wake-active noradrenergic neurons of the locus coeruleus (LC). To unravel the role of DPGi GABAergic PS-on neurons in PS onset and maintenance, we pharmacologically inactivated the DPGi by local microinjection of muscimol, a $GABA_A$ agonist, or clonidine, an alpha-2 adrenergic receptor agonist. Slow wave sleep (SWS) and PS were abolished during 3 and 5 h after muscimol injection in the DPGi, respectively. Clonidine treatment specifically induced a decrease in PS quantities and a delay in PS appearance. Combining c-FOS immunostaining with that of markers for the different populations of wake-active neurons, we found out that after muscimol treatment more than 75 % of the noradrenergic and adrenergic neurons, 60 % of the hypocretinergic, 51 % of the histaminergic, 44-27 % of the cholinergic neurons of the basal forebrain and less than 15 % of the dopaminergic and the ponto-mesencephalic cholinergic and serotoninergic neurons were activated.
These results show that DPGi PS-on neurons participate in PS control probably by means of silencing noradrenergic and adrenergic systems. Surprisingly, they also suggest that DPGi neurons might also dampen the activity of the noradrenergic and adrenergic systems also during SWS and waking.

P3.157

High accuracy decoding of dynamical motion from a large retinal population

Marre, O. (Paris)[1], Tkacik, G. (Klosterneuburg)[2], Simmons, K. (Philadelphia)[3], Mora, T. (Paris)[4], Berry, M. (Princeton)[5]
[1]Institut de la Vision, UPMC, INSERM, CNRS, Paris, France, [2]IST Austria, Klosterneuburg, Austria, [3]University of Pennsylvania, Philadelphia, United States, [4]Ecole Normale Superieure, Laboratoire de Physique Statistique, Paris, France, [5]Princeton University, Molecular Biology Dpt, Princeton, United States

We recorded a large population of ganglion cells in a dense patch of salamander and guinea pig retinas while displaying a bar moving randomly. We show that the bar´s position can be reconstructed from retinal activity with a precision in the hyperacuity regime using a linear decoder acting on 100+ cells. Bayesian decoders based on receptive field models did not perform as well, in part because those models did a poor job predicting ganglion cell firing rates. Instead of a simple population vector code, ganglion cells employed a distributed code to represent the object´s trajectory, with both ON and OFF cells contributing significantly and cells responding to motion in their surround. Population redundancy was high, but could be predicted from the information conveyed by individual cells. This

"uniform" redundancy allowed for diverse collections of ganglion cells to represent high-accuracy motion information in a form easily read out by downstream neural circuits.

P3.158
Cortical dynamics during wakefulness in the mouse

Fernandez, L.M.J. (Lausanne)[1], Comte, J.-C. (Lyon)[2], Le Merre, P. (Lyon)[3], Clairis, N. (Lyon)[3], Salin, P. (Lyon)[4], Crochet, S. (Lyon)[3]
[1]Université de Lausanne, Département des Neurosciences Fondamentales, Lausanne, Switzerland, [2]INSERM U 1028, CNRS UMR 5292, Université Lyon 1, Neuroscience Research Center, 2-Photon Microscopy Imaging Team, Lyon, France, [3]INSERM U 1028, CNRS UMR 5292, Université Lyon 1, Neuroscience Research Center, Physiology of Brain Arousal System Team, Lyon, France, [4]INSERM U 1028, CNRS UMR 5292, Université Lyon 1, Neuroscience Research Center, Physiopathology of the Neuronal Network Responsible for the Sleep-Wake Cycle Team, Lyon, France

The electroencephalographic activity in the brain during wakefulness has been typically described as being rapid, desynchronized and of low amplitude. However, some recent studies in rodents, based on local recordings in the cortex, support evidence for a more complex panel of activities depending on the behaviour. For instance, a state change in primary somatosensory cortex (S1) from quiet (QW) to active wakefulness (AW) has been observed in mice, while the animal is scanning the environment with its whiskers. During QW, the cortical activity exhibits slow, large-amplitude and locally synchronized fluctuations, which are strongly suppressed during AW, whereas fast, low-amplitude fluctuations persist. In the present study, we investigated whether the state change related to the behaviour during QW and AW is restricted to S1, or, instead, extends to other cortical areas.
For this purpose, we have chronically implanted mice with high impedance fine tungsten electrodes, allowing recording of Local Field Potentials (LFPs) in several areas. In addition to S1, we recorded LFPs in the secondary somatosensory (S2) and the primary motor areas (M1), in the primary visual (V1) and auditory (A1) areas, and in the parietal association area (PtA), the medial prefrontal cortex (mPFC) and the dorsal hippocampus (dCA1). After a period of habituation, mice were recorded in head-restrained condition during wakefulness, and behaviour was assessed from electromyogram (EMG) and fronto-parietal electroencephalography. Based on EMG activity, we have classified periods of wakefulness as AW or QW. Our results show that not only we were able to observe a state change related to muscular activity in S1, but also in sensorimotor areas (S2 and M1). The state change was observed to a less prominent extent in other sensory modalities (A1 and V1), and associative cortices (mPFC and PtA). Spectral and correlation analysis of LFPs revealed specific networks of cortical activities during wakefulness and various state changes during AW and QW, leading to the conclusion that cortical dynamics are highly heterogeneous during wakefulness in relationship with the behavior.

P3.159
Does autoantibody impairs astrocyte function? Neuromyelitis optica as a study model

Cavagna, S. (Lyon)[1], Nicolle, A. (Lyon)[1], Watrin, C. (Lyon)[1], Touret, M. (Lyon)[1], Ruiz, A. (Lyon)[1], Benetollo, C. (Lyon)[2], Peyron, C. (Lyon)[3], Marignier, R. (Lyon)[1], Giraudon, P. (Lyon)[1]
[1]Centre de Recherche en Neuroscience de Lyon (CRNL), Neurooncology Neuroinflammation Team, Lyon, France, [2]Centre de Recherche en Neuroscience de Lyon (CRNL), Plateforme Genetique Fonctionnelle et Optogénétique, Lyon, France, [3]Centre de Recherche en Neuroscience de Lyon (CRNL), Equipe Sleep, Lyon, France

Devic's neuromyelitis optica (NMO) is a rare autoimmune disease of the central nervous system (CNS) mainly directed against the spinal cord and the optic nerve. NMO is mediated by a specific serum autoantibody directed against the astrocyte water channel, aquaporin-4 (AQP4) mainly expressed as the blood brain barrier (BBB). It is now demonstrated that AQP4-Ab antibody (Ab) has a pathogenic role through complement-dependent astrocyte toxicity. However, autoantibodies can also trigger cell dysfunction. These later mechanisms probably took part, in some extend, in NMO. Indeed, some early NMO lesions are characterized by complete loss of AQP4 on preserved astrocyte. Modulation of AQP4 membrane expression had been also demonstrated in several study models. To explore the effect of the autoantibody by itself apart from complement activation, we propose a novel and unique NMO animal model based on a chronic infusion (7 days) of purified antibody from NMO patients and from healthy donors as controls, directly into the lateral brain ventricle of living rats. In parallel, we investigated the effect of a prolonged exposure of AQP4-Abs on astrocyte in primary culture. First, we showed that infused Ab was present in the rat blood and CNS structures (brain, optic nerve, the spinal cord), confirming the validity of our model. Infiltration of immune cells (T and B lymphocytes, macrophages) was also detected in the parenchyma of AQP4-Ab infused rats. Importantly, we also detected alterations similar to those reported for NMO patients: AQP4 loss, myelin loss, GFAP and NF disorganisation. In addition, connexin-43 and the inward rectifying potassium channel Kir 4.1 were modified. Westernbloting performed on punched tissues and analysis of AQP4-Ab-treated primary astrocytes (protein trafficking and level expression) confirmed these alterations. Finally, we showed that AQP4-Ab infusion led to a modulation of the blood-brain barrier, with a loss/disorganisation of Claudine 5 and accumulation of rat Ig around blood vessels within the CNS . In conclusion, we propose a new and original animal model for autoantibody mediated disease of CNS based on the effect of the Ig by itself and confirm the modulator effect of auto-Ab directed against membrane proteins.

P3.160

The chemokine MCP1 mediates inflammation-induced anorexia through its action on hypothalamic MCH neurons

Le Thuc, O. (Valbonne)[1], Blondeau, N. (Valbonne)[1], Rostène, W. (Paris)[2], Guyon, A. (Valbonne)[1], Cazareth, J. (Valbonne)[1], Heurteaux, C. (Valbonne)[1], Nahon, J.-L. (Valbonne)[1], Rovère, C. (Valbonne)[1]
[1]*IPMC - CNRS/UNS UMR 7275, Valbonne, France,* [2]*Institut de la Vision, UMRS 968-Université Pierre et Marie Curie, Paris, France*

Cytokines and chemokines are secreted by the immune system in response to injuries and/or infections and play an important role in anorexia. Lipopolysaccharide (LPS)-injection induces both pro-inflammatory pathways and weight loss and is used as a model to investigate the central regulation of appetite behavior and body weight by neuroinflammation. The lateral hypothalamic area (LHA) shelters two neuronal populations producing the two major orexigenic peptides: Melanin Concentrating Hormone (MCH) and hypocretin/orexin (ORX). In the present study, we showed by quantitative PCR and immunoassays that central injections of LPS induced a drastic expression of the Monocyte Chemoattractant Protein (MCP-1) mRNA and derived protein in LHA, while MCH and ORX mRNA and proteins were down-regulated. The time frame for this modification suggested that MCP1 overexpression could participate to the LPS-induced down-regulation of both MCH and ORX. Indeed, our results showed that cerebral injections of MCP1 decreased MCH and ORX mRNA and protein levels, similarly to LPS. The MCP1 receptor, CCR2, was also mapped by immunohistochemistry in MCH neurons. We tested therefore whether MCP1 could act directly on these neurons. Secretion and electrophysiology experiments demonstrated that MCP1 application on MCH-neurons in brain tissues decreased their activity. Finally, the use of pharmacological tools and transgenic animals lacking MCP1 and CCR2 confirm the role of MCP1 pathway in LPS-induced anorexia.
This work was supported by the CNRS and FRM.

P3.161

Forced desynchronization of the circadian system through chronic jet lag affects metabolic function

Casiraghi, L.P. (Bernal)[1], Alzamendi, A. (La Plata)[2], Giovambattista, A. (La Plata)[2], Golombek, D.A. (Bernal)[1]
[1]*Universidad Nacional de Quilmes, Department of Science and Technology, Bernal, Argentina,* [2]*Instituto Multidisciplinario de Biología Celular (CONICET-CIC), Neuroendocrinology Research Unit, La Plata, Argentina*

Circadian disruption by shift-work and jet-lag has been established as a health hazard in both humans and in animal models. The mechanisms by which these conditions lead to such a wide range of deleterious effects are still unclear. Internal desynchronization of physiological variables has been proposed as a potential underlying cause. We developed and characterized a model of forced desynchronization of the circadian system, based on a chronic jet-lag (CJL) schedule consisting in 6 hour advances of the light-dark (LD) cycle every 2 days (ChrA). Moreover, we suggest that this is due to the disruption of the coupling of the ventrolateral and dorsomedial regions of the master clock in the suprachiasmatic nuclei (SCN). The SCN clock tightly controls the timing of peripheral organs and metabolism, and we hypothesized that metabolic disturbances should be present in the ChrA model. We evaluated several metabolic variables (weight gain, food intake, leptin and triglycerides blood levels, and amount and histology of fat tissue) in mice subjected to ChrA. Restricting feeding times to periods of darkness in ChrA was assayed as a potential therapy for the circadian disruption. Animals under a CJL schedule of delays of the LD cycle (ChrD), which does not produce desynchronization, were also studied.
Body weight gain was significantly increased in animals under ChrA as compared to controls, an effect evident from the first week to over 60 days from ChrA start. Epididymal fat was also increased in ChrA mice. While restricting feeding times to the dark phase resulted in decreased body weight gain, it appears to be linked to varying food intake. There was no effect of the ChrD schedule on body weight gain as compared to controls.
We found that disruption of the biological rhythms through ChrA disturbs metabolic function at different levels. The effects seem to be linked to forced desynchronization of the circadian system, as no disturbances were detected under the less disrupting ChrD schedule, supporting the notion that advance and delay jet-lag have asymmetric effects on physiology. In conclusion, we confirm the ChrA model as a useful tool for studying health issues related to the stress of normal circadian function and strategies to prevent and overcome these hazards.

P3.162

Vasopressin decreases LTP in the CA2 field of the hippocampus

Desarménien, M.G. (Montpellier)[1], Chafai, M. (Valbonne)[2], Guillon, G. (Montpellier)[3]
[1]*Univ. de Montpellier 1 & 2, CNRS UMR 5203 - INSERM U 661, Institut de Génomique Fonctionnelle, Montpellier, France,* [2]*Institute of Molecular and cellular Pharmacology, CNRS UMR 7275, Valbonne, France,* [3]*CNRS UMR 5203, INSERM U 661, Univ. Montpellier I & II, Montpellier, France*

A growing body of evidence indicates that vasopressin, beside its peripheral endocrine actions, is involved in several brain functions including memory and social behaviour. One of the central targets of vasopressin neurons is the CA2 field of the hippocampus, a region particularly enriched in V1b receptors. The function of this CA2 field remains to be established, but its putative involvement in the theta rhythm and its complex innervation make it particularly interesting. Accordingly, we have tested the action of vasopressin and a specific V1b agonist on synaptic responses recorded from acute sagital slices of mice (C57Bl6-J) dorsal hippocampus. To this end, either Schaffer collaterals or entorhinal cortex (EC) fibres were stimulated and postsynaptic potentials were recorded from the dendritic field of CA2 neurons. High frequency stimulation (HFS) of either fibre populations induced a

long term potentiation (LTP) that installed progressively. Vasopressin (1 min. bath application) transiently decreased the amplitude of the LTP-enhanced excitatory post synaptic potential (EPSP) by ≈15%. Interestingly, this decrease occurred only after LTP-induction; 1 min. bath application of vasopressin did not affect the EPSP before HFS. In another set of experiments, we also noticed that vasopressin did not affect the control or LTP-enhanced EPSPs recorded in CA1 neurons after stimulation of Schaffer collaterals. Interestingly, the effect of vasopressin of the entorhinal cortex - CA2 synapse was mimicked by the specific V1b agonist dLeu4-Lys8-vasopressin, indicating a contribution of this receptor to the vasopressin response. Altogether, these data provide the first demonstration that vasopressin modulates excitatory inputs on CA2 neurons. This modulation may support some of the new roles of vasopressin and the V1b receptor in the regulation of memory and social behaviour.

P3.163

Neurohormonal effects of oxytocin and vasopressin receptor agonists on spinal pain processing in rats

Poisbeau, P. (Strasbourg)[1], Juif, P.-E. (Strasbourg)[1]
[1]*CNRS & Université de Strasbourg, Molecular Determinants of Pain, Strasbourg, France*

Oxytocin (OT) and arginine vasopressin (AVP) are two neuropeptides displaying well-known effects on the reproductive system. Oxytocin and vasopressin were shown to exert potent analgesic effects on the nociceptive system when administered directly in various central nervous structures. On the other hand, little is known about their peripheral (hormonal) actions on nociception and pain responses.
The aim of the present work was to characterize the effects of physiological blood concentrations of OT and AVP on spinal nociception and on pain responses.
To do so, growing doses of OT or AVP were administered intravenously and the nociceptive processing by spinal cord neurons was analyzed in anesthetized male rats in vivo. We observed that the action potentials mediated by C-type nociceptive fibers was strongly reduced (antinociception) after intravenous injections of low doses of OT (< 5µg) or AVP (< 500pg), whereas an increase (pronociception) was observed at higher doses. Interestingly, antinociceptive and pronociceptive effects were fully abolished in the presence of the OT receptor antagonist and the AVP receptor antagonist type 1A (V1A), respectively.
We confirmed this result using a behavioral model of forced swim stress-induced analgesia (SIA) associated with plasmatic release of OT (and not vasopressin). SIA was transiently lost following i.v. administration of OTR antagonist.
Together, the present work provides straightforward evidence that blood levels of OT and AVP modulate nociception and pain responses. The final target structures explaining these effects remains to be identified but are likely to be C-type nociceptors.

P3.164

Vasopressin V1b receptors in the rodent central nervous system detected and activated with new fluorescent ligands

Corbani, M. (Montpellier)[1], Marir, R. (Montpellier)[1], Tomboly, C. (Szeged)[2], Manning, M. (Toledo)[3], Guillon, G. (Montpellier)[1]
[1]*Univ. de Montpellier I et II, CNRS UMR 5203 - INSERM U 661, Institut de Génomique Fonctionnelle, Montpellier, France,* [2]*Biology Research Center, Institute of Biochemistry, Szeged, Hungary,* [3]*University of Toledo, Health Science Campus, Toledo, United States*

Vasopressin and CRF are involved in the stress response mainly by acting at the pituitary and the adrenal levels. Recently, vasopressin has also been shown to regulate, via central V1a, V1b and

oxytocin (OXT) receptors, the mood, the learning and affective disorders involving the limbic system. The role of V1b receptors has been especially emphasized but the lack of selective antibodies or specific radiomarkers has prevented further studies so far. At the same time, knockin AVP-EGFP rats were constructed in order to evidence AVP-ergic neurons (cell bodies and fibres) directly in brain regions (Ueta et al, 2005 Endocrinology,146,406). Based on the vasopressin-derived peptides previously synthesized in our group (Pena et al, 2007, Endocrinology,148, 4136-4146), we have developed new V1b selective ligands coupled to fluorophores chosen for their spectral emission and brightness in order to detect low levels of receptors in native tissues. These ligands have been characterized for their affinity and for functional coupling to phospholipase C or adenylate cyclase on human (Corbani et al, 2011, J Med Chem, 54, 2864-2877) and rat vasopressin (V1a, V1b, V2) and oxytocin receptors. Several ligands exhibiting a highly selective affinity (0.65-15 nM) for V1b-R also behaved as full agonists at V1b-R and very partial agonists (30%) at OXT-R. Since they provided convincing images in confocal microscopy for imaging the V1b receptors on transfected cells and on primary cultures, the fluorescent ligands were challenged on the rat brain to check brain structures where V1b-R expression was suspected. Rat brains were sectioned in coronal or sagittal slices and, using macroconfocal imaging, V1b sites were detected in the most relevant tissues known to be involved in the regulation of stress, anxiety, depression or aggressivity. In addition, taking advantage of the agonist nature of the fluorescent ligands, the activation of the Map kinase pathway in living brain slices by the fluorescent agonists could also be evidenced using a specific anti-pERK antibody. Thus, mapping the *in situ* expression of V1b receptors in connection with their function in normal and stressed animals will become possible using these tools.

P3.165

Characterization of a new murine model of multiple sclerosis using immunization with a CD8 epitope of myelin oligodendrocyte glycoprotein

Nicot, A.B. (Nantes)[1,2,3], Guillot, F. (Nantes)[1,2,3], Brouard, S. (Nantes)[1,2,3], Laplaud, D.A. (Nantes)[1,2,3]
[1]INSERM CR 1064, Nantes, France, [2]l'UNAM, Université de Nantes, Nantes, France, [3]CHU de Nantes, ITUN, Nantes, France

Multiple sclerosis (MS) is an autoimmune, demyelinating and degenerative disease of the central nervous system (CNS). Anti-myelin CD4 T cells are strongly associated with disease development in MS and in several murine experimental autoimmune encephalomyelitis (EAE) models. However, CD8 T cells often outnumber CD4 T cells in the CNS parenchyma of MS patients and recent studies suggest that anti-myelin CD8 T cells may be also involved. In order to better understand the contribution of CD8 T cells, we tested a new model of EAE by immunizing C57Bl/6 mice with an epitope of myelin oligodendrocyte protein (MOG) specifically presented by MHC-I; this has been previously reported to lead to either pathogenic or regulatory CD8 T cells. In our facility, one third of these mice developed chronic EAE with mild clinical signs while all mice immunized with MOG_{35-55} (contaning the CD4 and CD8 epitopes) developed full-blown EAE as expected. Proliferation and FACS analysis of T cell reactivity using splenocytes isolated from CD8 epitope-immunized mice confirmed the emergence of specific MOG-reactive CD8 T cells *in vivo*. Immunohistochemical analysis of the CNS of mice that developed clinical signs indicates that T cells infiltrate the CNS white matter with a caudo-rostral gradient. CD4 T cells outnumbered CD8 T cells in the cerebellum or spinal cord. Immunophenotyping of the infiltrating cells indicated that most T cells did not express the regulation marker FoxP3 but rather expressed activation T cell markers. Astrogliosis and myelin loss were also evidenced in the white matter of most severe EAE mice. Taken together, these data indicate that anti-MOG CD8 T cells can initiate EAE supporting an early role of anti-myelin CD8 T cells in MS pathogenesis.
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P3.166
Endogenous ghrelin tone influences feeding responses to exogenous ghrelin

Zizzari, P. (Paris)[1], Hassouna, R. (Paris)[1], Tomasetto, C. (Illkirch)[2], Epelbaum, J. (Paris)[1], Tolle, V. (Paris)[1]
[1]Centre de Psychiatrie et de Neurosciences, UMR 894 INSERM, Université Paris Descartes, Paris, France, [2]I.G.B.M.C., U 964 INSERM UMR 7104 CNRS, Biologie Moleculaire et Cellulaire des Cancers du Sein, Illkirch, France

Ghrelin is the only orexigenic hormone originating from the gastrointestinal tract and its actions on appetite are relayed through the Growth Hormone Secretagogue Receptor 1a. However, feeding responses to intraperitoneal ghrelin injections vary considerably from one C57BL/6 mouse to the other (Hassouna et al. PLoS One 2012). Interindividual variability is observed amongst animals within the same experimental group, suggesting that environmental factors alone (nutrition, stress,...) cannot explain this variability.
In the present study, each mouse received three ghrelin injections (30 nmol, ip) in the early light phase followed by 2 washout days over a 2-weeks period to test the reproducibility of the response. Mice that were clearly high responders (60%, ie increased food intake over a threshold of 0.42 g 4 hours after the injection) retained the capacity to respond to ghrelin throughout the experimental period, thereby suggesting that an endogenous factor determines the sensitivity to this orexigenic signal. However, ghrelin-induced feeding response did not correlate with the amount of food consumed within 15 minutes prior to the injections (R2=0,001, P=0,887), suggesting that immediate feeding status prior to the injection was not causal in the variability of responses. Ghrelin response was next measured in a group of mice after a 30 minutes restraint-stress. Following restraint-stress, animals divided into both low (50%) and high-responders (50%), indicating that psychological stress does not interfere with ghrelin-induced feeding response. We then hypothesized that endogenous ghrelin tone may differentiate high and low responders and compared food intake in wild-type and ghrl -/- mice. Interestingly, whereas only 60% of wild-type injected mice increased food intake over the threshold, 100% of ghrl -/- mice responded to ghrelin.
These data demonstrate that feeding response to ghrelin varies with individual animals and suggest that endogenous ghrelin tone influences the response to exogenous ghrelin. Whether this variability is purely dependent on the individual, or on the individual under a specific environment only, still needs to be investigated.

P3.167
Transitive inference reasoning is altered in spatial neglect

Petitet, P. (Bron)[1], Van der Henst, J.-B. (Bron)[2], Rossetti, Y. (Bron)[1]
[1]Centre de Recherche en Neurosciences de Lyon (CRNL), INSERM U 1028 - CNRS UMR 5292, Université Lyon 1, Impact Team, Bron, France, [2]Institut des Sciences Cognitives, Laboratoire sur le Langage, le Cerveau et la Cognition (L2C2) CNRS Université Lyon 1 UMR 5304, Bron, France

Spatial neglect is characterized by a deficit for perceiving and acting towards the left of space. In addition, it similarly affects the level of mental space representations, as shown by visual imaging. More recently it has been shown that this deficit tends to expand to the mental representation of numbers and other magnitude scales. At an even higher level of cognitive processing, one may wonder whether spatial neglect may affect reasoning.
Transitive inference (TI) is a type of reasoning frequently used in which a conclusion is inferred from different initial relations by combining them in order to draw new relations. It may be applied to spatial (A is on the left of B and B is on the left of C so A is on the left of C) and non-spatial dimensions (A is nicer than B etc.). It has long been suggested that transitive reasoning involves the manipulation of visuo-spatial mental models. Therefore it may be predicted that neglect patients should exhibit a deficit specific to spatial relations implicating the left-right dimension rather than the other space dimensions

(height, depth). In addition, we may also expect that within the left-right dimension, neglect patients may show more impairment in one direction rather than the other (i.e. left vs. right).
In order to test these hypotheses, we first compared subjects' TI performance (measured by the rate of correct answers and reaction time) for the 3 spatial dimensions:
1) left-right;
2) above-below;
3) in front of-behind.
Participants were presented with two-premise problems (A is on the left of B. B is on the left of C) and had to decide whether a subsequent conclusion could be logically derived from the premises or not (e.g. is A on the right of C? No.). Our data reveal a greater impairment in neglect when TI implicated the left-right dimension rather than the other dimensions. In addition, the spatial reasoning direction (towards the left or right) rather than the semantic side (using the words “left” or “right”) determined the neglect-related asymmetry between the left and the right. These results reveal that spatial neglect affects higher cognitive levels than usually assumed.
Keywords: Spatial neglect, transitive inference, spatial representation

P3.168

Tracking of speech rhythm by neuronal oscillations: an MEG study on natural fast speech perception

Laaksonen, H. (Lyon)[1,2], Jerbi, K. (Bron)[3], Ferragne, E. (Lyon)[1,4], Bedoin, N. (Lyon)[1], Boulenger, V. (Lyon)[1]
[1]CNRS/Université Lyon 2 UMR 5596, Laboratoire Dynamique du Langage, Lyon, France, [2]INSERM U 1028 - CNRS UMR 5292 - CRNL, Centre de Recherche en Neurosciences de Lyon, Bron, France, [3]INSERM U 1028 - CNRS UMR 5292, Centre de Recherche en Neurosciences de Lyon, Bron, France, [4]Université Paris 7, CLILLAC-ARP, Paris, France

Introduction: Recent work suggests a close correspondence between speech rhythm and cortical oscillations, allowing the brain to parse the acoustic signal into linguistic elements critical for language comprehension. Studies show phase-locking of ongoing theta oscillations in auditory regions to the amplitude envelope of the speech signal. Interestingly, an alignment between oscillatory activity in auditory and premotor regions has further been reported in speech-relevant frequency bands, supporting recent dual-stream models of a functional role of the motor system in speech perception. The goal of this study is to uncover the role of the speech production system by investigating how articulatory and auditory regions interact during natural speech perception using MEG.
Methods: Brain activity of 24 normal-hearing participants will be recorded using a 275-channel whole-head MEG system. In the study, participants listen to sentences whose final word is either congruent or not with the preceding context and have to decide whether they make sense. There are 3 experimental conditions: natural speech produced at a normal rate, natural speech produced at a faster rate and speech artificially time-compressed to match the fast speech rate. The control condition consists of amplitude-modulated noise at normal and fast rates. Stimuli of the same condition are grouped into randomized blocks of 3-5 items. Task-related time-frequency analysis and coupling between the speech signal envelope and the MEG signals will be computed using routine spectral analysis methods in FieldTrip toolbox.
Expected results: We expect to find evidence for an entrainment of neuronal oscillations to the amplitude envelope of the speech signal and that the coupling to auditory signal will be stronger for speech stimuli than for amplitude-modulated noise. Also, we expect that comparison of the power and coupling results obtained across the 3 speech conditions will show task-specific modulations of power in frontal regions both in low and higher frequency bands. Coupling analysis will test our hypothesis that processing faster speech rates is associated with changes in the extent or pattern of coupling of the neuronal activity in the articulatory and auditory regions to the speech envelope.

P3.169

Multivoxel pattern analysis of grapheme-color synesthesia

Ruiz, M.J. (Grenoble)[1,2], Dojat, M. (Grenoble)[1], Hupé, J.-M. (Toulouse)[2]
[1]Grenoble Institut des Neurosciences, Isère, Grenoble, France, [2]Centre de Recherche Cerveau et Cognition, Haute-Garonne, Toulouse, France

Grapheme-color synesthesia is a subjective phenomenon in which colors are automatically and arbitrarily associated to letters or digits. The automatic perception of ´synesthetic colors´ in response to achromatic graphemes is especially informative to study color vision isolated from low-level color coding. A popular hypothesis proposes that a ´real colors center´ such as V4/V8, adjacent to the Visual Word Form Area, is co-activated by grapheme processing. However, studies using univariate analyses failed to provide univocal evidence in favor of this assumption [for review see Hupé et al. (2012) Cereb. Cortex 22(7)]. MultiVoxel Pattern Analysis (MVPA), which provides information by classifying patterns of activation with machine learning algorithms, may reveal fine grained processing not detected by single-voxel or Regions Of Interest analysis. We collected so far fMRI data of 3 synesthetes and 3 non-synesthetes. We presented in the scanner black letters and digits that were specifically associated for each synesthete to red, green, blue and yellow. We also presented these colors as moving concentric circles. We hypothesized that ´real color´ (concentric rings) and ´synesthetic color´ (black graphemes) perception would share patterns of activations in synesthetes but not in non-synesthetes (who do not experience any color for black graphemes). Support Vector Classification was able to successfully predict ´real color´ perception (up to 50%, $p < 0.05$, binomial threshold = 37%) and grapheme perception (up to 80%) in anatomically and functionally defined visual areas of individual subjects. As expected, cross-classification between ´real colors´ and ´synesthetic colors´ did not reach significance in non-synesthetes, but neither did it for synesthetes. This result suggests that there is no shared pattern of activation for ´real´ and ´synesthetic colors´. However, cross-classification of different kinds of stimuli leads automatically to a decrease of performance, and classification rate for ´real colors´ may not have been high enough in this data set to allow us detecting the transfer. Individual and group analyses of more subjects are in progress and will allow us to further understand these issues.

P3.170

Social behavioral disorders in MPTP monkey model of Parkinson's disease

Durand, E. (Clermont Ferrand)[1], Petit, O. (Strasbourg)[2], Zimmer, C. (Strasbourg)[2], Tremblay, L. (Bron)[3], Sgambato-Faure, V. (Bron)[3], Chassain, C. (Clermont Ferrand)[4], Laurent, M. (Clermont Ferrand)[1], Pereira, B. (Clermont Ferrand)[5], Silberberg, C. (Clermont Ferrand)[1], Durif, F. (Clermont Ferrand)[1]
[1]Université d'Auvergne, EA 7280, Clermont Ferrand, France, [2]Institut Pluridisciplinaire Hubert Curien-Département Ecologie, Physiologie et Ethologie (IPHC-DEPE), UMR 7178, CNRS-UDS, Strasbourg, France, [3]Centre de Neuroscience Cognitive (CNC), Institut des Sciences Cognitives (ISC), CNRS UMR 5229, Bron, France, [4]CHU Gabriel Montpied, Centre d'Imagerie par Résonance Magnétique, Clermont Ferrand, France, [5]CHU Clermont-Ferrand, Unité Biostatistiques, Clermont Ferrand, France

Parkinson´s disease is a neurodegenerative disorder with progressive impairment in motor and cognitive functioning. Individuals experience not only physical discomfort brought on by the illness, but also considerable psychological distress. In some patients, this psychological distress can result in a disruption of social relationships. It is more likely that social disorders, like cognitive deficits, appear in pre-symptomatic and/or in symptomatic stage of the disease.
The present study focused on social behavior within a group of female long-tailed macaques assessed
1) in baseline condition
2) in pre-symptomatic condition after chronic low doses (CLD)-1-méthyl-4-phényl-1,2,3,6-tétrahydropyridine (MPTP) administration (0.1 mg/kg doses)

3) in pre-symptomatic condition after chronic high doses (CHD)-MPTP administration (0.4 mg/kg doses administration individually adjusted according to the response and until stability of parkinsonian-like symptoms) and
4) in symptomatic conditions when animal developed stable parkinsonian-like symptoms during 1 month minimum.
During these four periods, each testing session included social behaviors, cognitive abilities and motor behaviors assessments. Moreover, dopaminergic denervation was assessed in cortical areas and basal ganglia by fluorine-18-L-dihydroxyphénylalanine (^{18}F-DOPA) positron emission tomography (PET) scans.
Social behavioral disorders were identified within the social group during the pre-symptomatic state following CLD-MPTP protocol including significant increases of aggressive and affiliative behaviors. These observations were associated with the emergence of more frequent and intense conflicts following by post-conflict reconciliation episodes. Unexpectedly, these social behavioral disorders were only identified within the subordinate animals and observed before any motor and/or cognitive impairment for the first time. Otherwise, cognitive deficits, motor and social behavioral disorders, with decrease of submissive behaviors, were identified on the symptomatic state following CHD-MPTP protocol. Finally, PET scans data analyses allowed to highlighted possible implication of cortical structures, namely the insula, the orbitofrontal cortex and the anterior cingulate cortex in these social behavioral disorders.

P3.171

Retinoic acid restores aged-related cognitive deficits and glucocorticoid signalling pathway alterations

Bonhomme, D. (Bordeaux)[1], Touyarot, K. (Bordeaux)[1], Servant, L. (Bordeaux)[1], Henkous, N. (Bordeaux)[2], Higueret, P. (Bordeaux)[1], Béracochéa, D. (Bordeaux)[2], Pallet, V. (Bordeaux)[1]
[1]NutriNeuro, Bordeaux, France, [2]INCIA, Bordeaux, France

It now established that vitamin A and its derivatives, *all-trans* retinoic acid (RA), are required for cognitive functions in the adulthood and that retinoid hyposignalling occurring naturally during aging contributes to the deterioration of hippocampal synaptic plasticity and functions (Etchamendy, et al., 2001). Furthermore, recent data have revealed interaction between retinoid and glucocorticoid signalling pathways. Particularly, an inhibitory effect of RA has been observed on the 11bHSD1 expression, enzyme which regenerates active glucocorticoids. This modulation might contribute to the beneficial effects of retinoids on memory since it has been recently shown that hippocampal 11bHSD1 expression increases with aging and correlates with spatial memory deficits.
In the present study, we therefore aim to test the hypothesis that the beneficial effect of vitamin A supplementation, or RA administration, on memory performances and cerebral plasticity could be also mediated by a modulation of the glucocorticoid pathway. We investigated the effects of vitamin A supplementation (25UI/g retinol, 2 months) and RA treatment (150µg/kg, 5 days) to 14 month-old mice, on contextual serial discrimination task (CSD) which allows the detection of early signs of age-related hippocampal-dependent memory dysfunction. We measured plasma and intrahippocampal corticosterone concentrations (by microdyalisis). Measurement of intrahippocampal expression of hippocampal glucocorticoid, and retinoid target genes by real time RT-PCR will be conducted in order to verify if vitamin A status can modulate glucocorticoid pathway in the hippocampus of aged mice. Our results showed that RA treatment and vitamin A supplementation improve “episodic-like” memory and have an inhibitory effect on corticosterone secretion. Moreover, these behavioural changes were associated to a modulation of hippocampal genes expression involved in glucocorticoid and retinoid signalling pathways including 11bHSD1. Vitamin A supplementation in diet or pharmacological RA treatment could be good strategies respectively to prevent or reverse age-induced hippocampal alterations and memory deficits.

P3.172

Does a neuronal mutation of thyroid hormone receptor α alter anxiety in mice?

Richard, S. (Lyon)[1], Dkhissi-Benyahya, O. (Lyon)[2], Thévenet, M. (Lyon)[3], Flamant, F. (Lyon)[1]
[1]Institut de Génomique Fonctionnelle de Lyon, ENS de Lyon - CNRS UMR 5242 - USC INRA 1370, Lyon, France, [2]Institut Cellule Souche et Cerveau, INSERM/Univ. Lyon 1/INRA, Lyon, France, [3]Centre de Recherche en Neuroscience de Lyon (CRNL), UMR 5292 CNRS/U 1028 INSERM, Lyon, France

In humans, hypothyroidism is often associated with an increase in anxiety and depression disorders. This link has also been reported in rodent models of hypothyroidism. These observations, together with the wide distribution of thyroid hormone receptors in the brain, suggest that thyroid hormone may be involved in the control of emotions. In order to investigate the role of central thyroid hormone receptors in the control of anxiety, we used male and female mice expressing a neuron-specific mutation of thyroid hormone receptor α ($TR\alpha^{AMI}$ Cre3). Anxiety behaviour was evaluated in the elevated-plus maze, light-dark box and open-field test. Circadian rhythms of home cage activity and food intake were also recorded. In the elevated-plus maze and light-dark box, the number of zone transitions was lower in $TR\alpha^{AMI}$ Cre3 mice than in littermate controls, but there was no difference between genotypes in the time spent in the brighter parts of these devices. In the open-field test, there was no difference between genotypes in the total distance travelled by the mice, but the number of entries into the central zone was lower in $TR\alpha^{AMI}$ Cre3 mice than in controls. The present results suggest that $TR\alpha^{AMI}$ Cre3 mice may be slightly more anxious than control mice. A limitation of the $TR\alpha^{AMI}$ Cre3 model is that expression of the TRα mutation is restricted to a fraction of brain neurons, which might explain the mildness of the observed phenotype. Further investigation will be needed to evaluate the involvement of brain thyroid hormone receptors in the control of emotional behaviour.

P3.173

Dissociation of spatial navigation strategies in primates by inhibiting various territories of the striatum

Etienne, S. (Bordeaux)[1,2], Guthrie, M. (Bordeaux)[1,2], Orignac, H. (Bordeaux)[1,2], N'Guyen, T.H. (Bordeaux)[1,2], Gross, C.E. (Bordeaux)[1,2], Boraud, T. (Bordeaux)[1,2]
[1]Université Bordeaux Segalen, Institut des maladies neurodégénératives, UMR 5293, Bordeaux, France, [2]CNRS, Institut des maladies neurodégénératives, UMR 5293, Bordeaux, France

Two separate sub-cortical brain systems have been shown, in rodents, to learn environmental navigation in parallel but at different rates and encoding the environment in different ways. In the rodent, the hippocampal system has been shown to support allocentric (or place-based) strategies and the basal ganglia cue-based and direction-based strategies. This has not been reproduced in the primate and taxonomic differences between the groups mean that it is not possible to extrapolate from rodents to primates. The two groups rely on different basic perceptual systems, olfaction and vibrissae in rodents and vision in primates. As a consequence, in contrast to the place cells of spatial maps formed in the rodent hippocampus, primates form a hippocampal mapping more related to visual orientation. As this issue has consequences for day-to-day functioning in the most common human neurodegenerative diseases, we are in the process of quantifying the interactions between the two neural networks in the non-human primate. We have developed a task that is adapted from the classical X-maze of Packard & McGaugh (1996) that showed the dissociation of learning between the two systems. We have trained three female rhesus macaque monkeys to navigate the maze in a powered wheelchair. We showed that the monkeys can use the three competing strategies (Place-based, Cue-based and Direction-based) depending on the length of the learning. These first results are concordant with those obtained in spatial navigation task led with human subjects in a virtual environment (Iaria and al. 2003). Using a pharmacological approach, we showed that local inhibition of the shell of the nucleus accumbens induces a strong decrease of the place strategy and the inhibition of the dorsomedial striatum reduces the direction strategy. We are still investigating the striatal territory underlying the cue strategy.

P3.174

Potential role of sleep in memory and forgetting - a rat study

Carponcy, J. (Lyon)[1], Fraize, N. (Lyon)[1], Dijoux, F. (Lyon)[1], Youssef, M. (Lyon)[1], Salin, P. (Lyon)[1], Malleret, G. (Lyon)[1], Parmentier, R. (Lyon)[1]
[1]Centre de Recherche en Neuroscience de Lyon (CRNL), UMR 5292 CNRS/U 1028 INSERM, Lyon, France

It is commonly accepted that sleep promotes memory processes. Giuditta et al. (1995) suggest two distinct but interdependent roles of the two different sleep phases, slow wave sleep (SWS) and paradoxical sleep (PS). In a first step, SWS allows the reduction of "non-essential" memory traces, while the consolidation of the remaining relevant informations occurs during PS. We tested this hypothesis that SWS promotes forgetting while PS facilitates the consolidation of information by coupling electrophysiological and behavioral approaches in rats submitted to tasks aimed to assess three distinct forms of memory in a radial maze (Malleret et al., 2010):
1) reference memory (RM) requiring the long-term consolidation of invariable information,
2) working memory (WM) with high level of proactive interference (HIWM) during which storage of variable information is required but forgetting of previous trials is preferable to avoid an overload of irrelevant information, and
3) working memory with low level of interference (LIWM) during which such forgetting is not required.
Each day, at rest (after training), sleep EEG/EMG activity was recorded.
We first observed a transient increase in the amount of PS (+35%) in the RM task the day the animal has learned the rule. In HIWM, no quantitative changes in SWS or PS were noted. However, a significant positive correlation is seen between performance on day n+1 and the amount of SWS (but not PS) shown the day before (day n). In contrast, in LIWM, no notable change was found in the sleep amounts or in the correlations between SWS or PS and WM performance. Our results thus suggest that PS contributes to the long-term storage of information whereas SWS would be required for proper treatment of memory interference and therefore forgetting of irrelevant information required for working memory.

P3.175

Perceptual inference as revealed by dynamic causal modeling of cortical evoked responses: a simultaneous EEG-MEG MMN study

Lecaignard, F. (Bron)[1,2,3], Bertrand, O. (Bron)[1,2], Daligault, S. (Lyon)[3], Delpuech, C. (Bron)[1,2,3], Gimenez, G. (Lyon)[3], Maby, E. (Bron)[1,2], Perrin, M. (Bron)[1,2], Mattout, J. (Bron)[1,2], Caclin, A. (Bron)[1,2]
[1]LNRC, Dycog, Bron, France, [2]University Lyon 1, Lyon, France, [3]CERMEP - MEG Department, Lyon, France

Perception has been suggested to reflect the interplay between top-down inner predictions and bottom-up incoming sensory information. Furthermore, neurophysiological evoked responses such as the Mismatch Negativity (MMN) would result from the neuronal computation of prediction errors [1]. Dynamic Causal Modeling (DCM) enables to test formal hypothesis about this cortical implementation and about the causal relationship between context dependent modulations of effective connectivity and changes in the dynamics of electrophysiological responses [2]. A series of EEG DCM studies have shed light on the generative process of the MMN [3].
In this context, the objective of the present study was twofold:
· to assess the effect of predictability of auditory sequences on the MMN
· to refine the DCM of such responses by combining MEG and EEG measurements.

We therefore conducted a passive frequency-MMN study, using simultaneous EEG and MEG recordings. We compared an unpredictable (U) with a predictable (P) condition, where deviant sounds occurred randomly and in a deterministic fashion, respectively. We hypothesized that error prediction and hence evoked responses would be reduced in condition P.
At the sensor level, both EEG and MEG validated this hypothesis by revealing a significant decrease of MMN amplitude.
We applied DCM to EEG and MEG data separately, in order to assess the cortical network and the modulations of effective connectivity that best explain the oddball effect. Bayesian model comparison was then informed by the two modalities to identify the best model family: a bilateral fronto-temporal network with modulated forward and backward connections. This result is consistent with previous findings [3], except for the contribution of the left inferior frontal gyrus.
Finally, the effect of predictability was assessed by comparing the conditional estimates of effective connectivity modulations, for each connection. MEG revealed a weaker modulation of fronto-temporal connections, in condition P, which was not supported by EEG. This highlights the different sensitivity of the two modalities and speaks in favour of true data fusion.
[1] Friston, Philos Trans R Soc Lond B, 2005
[2] David et al., NeuroImage, 2006
[3] Garrido et al., J Neurophysiol, 2009

P3.176

DHA improves spatial memory and modulates gene expression in the brain of aged mice

Létondor, A. (Bordeaux)[1,2,3], Béracochéa, D. (Talence)[4,5], Krazem, A. (Talence)[4,5], Buaud, B. (Pessac)[3], Vaysse, C. (Pessac)[3], Pallet, V. (Bordeaux)[1,2], Alfos, S. (Bordeaux)[1,2]
[1]Univ. Bordeaux, Nutrition et neurobiologie intégrée, UMR 1286, Bordeaux, France, [2]INRA, Nutrition et neurobiologie intégrée, UMR 1286, Bordeaux, France, [3]ITERG, Institut des corps gras, Pessac, France, [4]CNRS, INCIA, UMR 5287, Talence, France, [5]Univ. Bordeaux, INCIA, UMR 1286, Bordeaux, France

Brain aging is associated with multiple morphological and biochemical changes leading to cognitive decline. Fatty acids are the main components of the brain membranes and among them docosahexaenoic acid (DHA) is the major n-3 polyunsaturated fatty acids (n-3 PUFAs). Several studies suggested that n-3 PUFAs and most particularly DHA are critical for the maintenance of cognitive functions during aging. It has been recently reported that unesterified DHA pool contained in plasma decreases with age in rodents and that brain DHA levels are altered during aging. DHA may modulate brain functions by several mechanisms including the regulation of gene transcription. Indeed DHA is the endogenous ligand of nuclear receptors such as peroxisome proliferator-activated receptors (PPARs) and retinoid X receptors (RXRs) which are transcription factors that modulate, in the brain, the expression of genes involved in synaptic plasticity. RXR is a master regulator that forms heterodimers with numerous nuclear receptors such as PPAR or the retinoic acid receptor (RAR). Recent data suggest that unesterified DHA (active form) is involved in working memory in mice *via* RXRγ activation. The objective of the present study was to evaluate in aged mice the effects of intraperitoneal administration of unesterified DHA on memory performances and synaptic plasticity. In a first experiment, we studied the effect of four doses of DHA injected during four days on the spatial working memory evaluated in a sequential alternation paradigm. Our results showed that DHA (0.1 and 1 mg/kg of body weight) have a beneficial effect on the alternation performances. We then evaluated in aged mice the effects of these efficient doses in a contextual and serial discrimination (CSD) task involving episodic-like memory. Our results showed that DHA improve memory performances of aged mice in the CSD paradigm. In order to understand the molecular mechanisms involved in this beneficial effect we also measured by qPCR the mRNA expression of nuclear receptors (RARs, RXRs and PPARs) and synaptic plasticity markers (GAP-43, RC3 and PSD95) in hippocampus and prefrontal cortex. Our results suggest that unesterified DHA plays an important role in the maintenance of memory processes during aging.

P3.177

Effect of polyphenol-rich plant extract on age-related cognitive decline

Bensalem, J. (Bordeaux)[1,2,3], Gaudout, D. (Libourne)[3], Lafenêtre, P. (Bordeaux)[1,2], Pallet, V. (Bordeaux)[1,2]
[1]*Université Bordeaux, Nutrition et neurobiologie intégrée, UMR 1286, Bordeaux, France,* [2]*INRA, Nutrition et neurobiologie intégrée, UMR 1286, Bordeaux, France,* [3]*Activ'Inside, R & D, Libourne, France*

The health of the aged population is a public health priority. Indeed, the number of seniors over 65 is increasing and is estimated to reach 16% of the world population in 2050.
During aging, cognitive deficits have been observed associated with synaptic plasticity impairments. Recent studies have highlighted the beneficial role of alimentation to prevent this decline. Polyphenols have been identified as potential functional food candidates. They are known to impede the production of free radicals but they could also potentiate the signaling pathways of synaptic plasticity as well as learning and memory performances.
This study aims to investigate the effects of a polyphenol-rich plant extract supplementation on learning and hippocampal-dependent memory and to better understand the neurobiological mechanisms underlying these effects.
The memory deficits of aged mice were first highlighted in the Morris water maze task. Indeed, reference memory depends on the integrity of the hippocampus, a brain structure which is particularly affected during aging. In order to assess the effectiveness of nutritional polyphenols to prevent or delay the occurrence of this age-related cognitive decline, young and old mice were submitted during 6-week to a polyphenol enriched diet. Preliminary results suggest that polyphenols are able to improve the memory retention capacity of supplemented mice. To shed more light on the molecular and cellular mechanisms, we further analyzed the expression of gene encoding proteins involved in synaptic plasticity (BDNF, GAP43, RC3) by RT-qPCR, the phosphorylation of Erk and Akt by western-blotting and the morphological changes of neurons by immunohistochemistry.

P3.178

Native and foreign language discrimination in adults: an optical imaging study

Perugia, E. (Amiens)[1], Mahmoudzadeh, M. (Amiens)[1], Vannasing, P. (Montreal)[2], Lassonde, M. (Montreal)[3], Wallois, F. (Amiens)[1]
[1]*Inserm U 1105 GRAMFC, Université de Picardie Jules Verne, CHU Amiens, Amiens, France,* [2]*Centre de recherche, Centre Hospitalier Universitaire Sainte-Justine, Montreal, Canada,* [3]*CERNEC, Université de Montréal, Montreal, Canada*

Introduction: We have assessed the capabilities of adults in discrimination the languages, comparing their native language (i.e. French) to one belonging to a different rhythmical classes (i.e. Arabic) and, in either case, played both forward (FW) and backward (BW). The brain mechanisms underlying the language discrimination capabilities are studied using optical imaging with functional Near Infrared Spectroscopy.
Methods: The data were recorded from right-handed native French speakers (mean age 26.5) and do not understand Arabic language.
The Snow White story in both French and Arabic language was presented with FW and BW speech. We have recorded the fairy tale in both languages by the same perfect bilingual female speaker and used the Praat software to reverse the utterances. Each session included 72 sentences (i.e. 18 sentences for each language conditions) which lasted 20 sec and where spaced-out randomly from 23 to 27 sec of silent.
Results: The oxy-Hb activation patterns in response to the stimuli were studied comparing and contrasting the language conditions and the hemispheres. The French story played forward elicited a more consisted and robust answer than the others conditions for all subjects; in particular in the left

temporal area. The activation by Arabic FW is different among the subjects, in general lower than the French FW. The story played backward, both in French and the Arabic, elicited a weak and randomly answers: these conditions have neither semantic nor syntax information than can be processed by the subjects.
The subjects´ brain is more activated by the native language (i.e. French) stimuli than foreign one (i.e. Arabic) or the backward conditions. The languages are preferably processed by the temporal area in left hemisphere.
Discussion: Since fairy tale is recorded from the same perfect bilingual female speaker, the discrimination task was focused just on the language. The subjects´ activation patterns were different between the languages played FW with a stronger activation for the French than the Arabic and this is consistent with our expectation and the previous works (Mazoyer, 1993; Schlosser, 1998). The comprehension of the French may be the cause of the greater hemodynamic response for the French story than for the Arabic.

P3.179
Electrophysiological correlates of task-set adaptation in task switching paradigms

Favre, E. (Bron)[1,2], Lio, G. (Bron)[1], Boulinguez, P. (Bron)[1]
[1]Centre de Neuroscience Cognitive (CNC), Institut des Sciences Cognitives (ISC), CNRS UMR 5229, Université Lyon 1, Bron, France, [2]Service de Neurologie C, Hôpital Neurologique Pierre Wertheimer, Hospices Civils de Lyon, Bron, France

A key mechanism of cognitive control is the ability to appropriately maintain or change a task set in unpredictable environment, i.e., to activate appropriate stimulus-response associations rules. Most emphasis has been directed to the processes involved when the task-set must be reconfigured (task switching). However, recent studies suggested that the repetition of the same appropriate stimulus-response association rules may induce progressive task-set adaptation, which in turn may affect task switching operations.
The neural and psychophysiological correlates of task setting and task switching have been studied with event-related fMRI and ERPs, but the conclusions are controversial. One main reason is that blood flow analyses are unable to separate in time the different processes likely entangled in the function. Another one is that classical scalp EEG measurements are based on the mixing of signals of different types that seriously obscures the meaning of the ERPs classically analyzed. Here, we used a task switching paradigm, and performed advanced high-resolution EEG blind source separation to track the electrophysiological correlates of task-set adaptation.
17 healthy participants participated in a color/object naming task. Each task-set could be repeated up to 5 consecutive times, or unpredictably switched to the other task-set. gICA were performed to measure the correlation between the degree of task-set repetition and the amplitude of significant ERPs, directly at the source level.
Task-set repetition induced gradual changes in:
1) An early visual component (109 ms) whose activity decreased as a function of task-set repetition,
2) a late component in the SMA (380 ms) whose activity increased as a function of task-set repetition, and
3) an even later component in the ACC/dorsomedial prefrontal cortex (400 ms) whose activity increased as a function of task-set repetition.
These results provide further evidence for task-set adaptation. It is inferred that these modulations could be related to:
1) decreased visual attention, and
2) increased motor preparation.
The pattern of results revealed by the third component is more obscure. It might be related to increased requirements for conflict monitoring as stimulus-response associations are reinforced.

P3.182

Anti-stress effect of maternal high-fat diet in an animal model of maternal separation in rats

Minni, A. (Bordeaux)[1], Lepinay, A. (Bordeaux)[1], Delage, P. (Bordeaux)[1], Theodorou, V. (Toulouse)[2], Fioramonti, J. (Toulouse)[2], Layé, S. (Bordeaux)[1], Darnaudéry, M. (Bordeaux)[1]
[1]NutriNeuro, INRA UMR 1286, Univ. Bordeaux, Bordeaux, France, [2]Neuro-gastroenterologie et Nutrition, UMR INRA/ENVT 1331, Toulouse, France

In human, early life adversity such as childhood traumas is associated with an increased vulnerability to develop anxiety disorders and irritable bowel syndrome in adulthood. In rats, chronic maternal deprivation promotes a hyper-activity of the Hypothalamo-Pituitary-Adrenal (HPA) axis in response to stress, associated with enhanced anxiety-like behavior in adult offspring. This model also affects gut mucosa immune status and leads to visceral hypersensitivity. Recently, it has been demonstrated that high fat diet ameliorates anxiety- and depressive-like behaviors in mother rats subjected to maternal separation. However, it is unknown whether maternal high fat diet consumption can also protect the progeny against early stress-induced emotional disturbances. The aim of our study was to investigate the influence of maternal high fat diet (20% lipids) during gestation and lactation on behavior and visceral sensitivity in adult offspring previously exposed to maternal separation during the first two weeks of life (3h/day). Our results indicate that maternal separation leads to spatial memory impairments, an exacerbated anxiety-like behavior, HPA alterations and hyper-sensitivity to colonic distension in offspring of mothers fed a standard diet. However, these alterations are counteracted by maternal high fat diet. The comfort food theory suggests that stressful situations promote nutrient-dense foods intake which in turn reduces endocrine and behavioral effects of stress. Interestingly, in an additional study, we report that dams exhibit a marked increase of high fat intake, associated with a decrease of their anxiety-like behavior during the separation sessions. These results reinforce the idea that stress promotes palatable food intake and highlight the critical role of early nutrition in neurodevelopment and behavioral responses later in life.

P3.183

Neuroscreen: high-throughput screening for psycho-active molecules in zebrafish embryos

Benedetti, J. (Paris)[1], Blondeel, S. (Paris)[1], Alavi, M. (Paris)[1], Yanicostas, C. (Paris)[1], Soussi-Yanicostas, N. (Paris)[1]
[1]INSERM U 676, Hôpital Robert Debré, Université Paris 7, Paris, France

Neuroactive small molecules are essential tools for treating mental illnesses as well as dissecting nervous system function. However, very few novel psycho-active drugs only have been discovered in last decades and most of them were discovered serendipitously. Given the unmet need for new psycho-active molecules, we have developed a simple approach that combined an automated motility recording system with the biological complexity of living animals to identify *in vivo* novel psychoactive molecules using zebrafish embryos*. This simple, powerful assay allowed a behavioural “barcode” to be devised whereby complex behaviour can be reduced to a series of single quantifiable behaviours. This behavioural “barcode” then enables us to classify psychotropic drugs and assess their psychoactivity. High-throughput screening of chemical libraries or psychoactive compounds generates huge volumes of data, yet there is still no software available to date that can rapidly and efficiently analyze them all. The *Z* software** that our team has developed lets users screen thousands of datasets in just a handful of seconds and to swiftly identify novel compounds thanks to the power of the comparisons executed.
By combining automated high-throughput screening technologies with our Z software we have already discovered new psycho-active molecules.
This approach allowed us to rapidly identify novel psychotropic molecules and to predict their molecular targets, toxicity and psycho-activity.

- **Keywords:** High-throughput in vivo screening; Zebrafish; Small molecules; Psycho-active molecule; Z software for analysis of high throughput screening; Brain ; Moto-neurons ; Behavior, Motility ; Vertebrate model

*This service ²High-throughput screening for small psycho-active molecules in zebrafish embryos² (reference : MT0478) is proposed by the INSERM transfert: https://migratech.inserm-transfert.fr
**This software ²Z software platform for fast mining of large-scale behavioural screening² (reference: MT0577) is proposed by the INSERM transfert: https://migratech.inserm-transfert.fr

P3.184

A lack of hippocampo-amygdala communication is associated with impairment of contextual fear in absence of the ID gene Il1rapl1

Chun-Lei, Z. (Bordeaux)[1], Xander, H. (Bordeaux)[1], Marilyn, L. (Bordeaux)[1], Elisabeth, N. (Bordeaux)[1], Yann, H. (Bordeaux)[1]
[1]CNRS, Université Bordeaux Segalen, Institut Interdisciplinaire de NeuroSciences, Bordeaux, France

Loss of functional mutations in the gene encoding for the Il1rapl1 protein (*IL1RAPL1*) leads to cognitive disabilities (CDs) in humans. Here we show that constitutive lack of *Il1rapl1* in mice leads to a prominent loss of contextual fear. And many lines of evidence show that hippocampus and amygdala play crucial roles in contextual fear memory. In order to identify the origin of the deficit, *in vivo*, we performed pharmacological and opto-genetic manipulations of amygdala and hippocampal formations in *Il1rapl1* -/y and *+/y* mice. *Ex vivo*, we examined the early gene expression level in subpopulations of basolateral amygdala (BLA) and hippocampal neurons, 2 hour after achievement of contextual learning. *In vitro,* we also completed an extensive morpho-functional study of synaptic properties and plasticity at hippocampo-BLA projections, in control conditions and following contextual fear learning. All together, our data provide an integrative and comprehensive model explaining the lack of contextual fear in Il1rapl1 deficient mice.

P3.185

Effect of the pronunciation of an action verb in different languages and dialects on the performance of a complex motor action

Lammari, F. (Zéralda)[1,2], Rabahi, T. (Villeurbanne)[3], Hariti, H. (Zéralda)[1,2], Fargier, P. (Villeurbanne)[3], Clouzeau, C. (Villeurbanne)[3,4], Massarelli, R. (Villeurbanne)[3]
[1]Université Alger 3, IEPS Institut de l'Education Physique et Sportive, Zéralda, Algeria, [2]Université Alger 3, Laboratoire de Sciences et Pratiques des Activités Physiques, Sportives et Artistiques (SPAPSA), Zéralda, Algeria, [3]Université Claude Bernard Lyon 1, Centre de Recherche et d'Innovation sur le Sport (CRIS, EA 647), Villeurbanne, France, [4]Institut Supérieur d'Ostéopathie (ISOSTEO), Unité de Recherche, Limonest, France

The object of the present research was to study the effect of the pronunciation of an action verb (*jump* at the second person of the imperative tense) upon the height of a *squat vertical jump* (SVJ) in a population of Algerian sportspersons, males (M) and females (F), experts in two domains, basketball (B) and volleyball (V), and in a naïf group. The results show that, in MV players (n=25), the action verb improves the performance of SVJ (by 1.7-2.5 cm) when pronounced in a maternal Arab dialect (*soté, enteg*, *djelleb,* in Kabyl and *soté, negueze* for high plateaus or southern subjects), in classical Arab (*ikfaze*), in English (*jump*) and in French (*saute*). A not significant improvement of the SVJ height (an increase of 0.5-0.7 cm) was observed with the same stimuli in FV (n=33). Among MB players (n=28) the performance was not significantly ameliorated (0.5-0.9 cm) after pronunciation of the action verb in English, French and Arabic dialect while FB athletes (n=30) improved significantly (0.7-1.0 cm), except in classic Arab (still improving by 0.5 cm). Naïf Sport students (32 males and 26 females) showed no statistically significant improvement (yet increasing the height by 0.5-0.7 cm, for both). The

pronunciation of *saute* increased the performance in females. In similar experiments performed in Lyon, naïf male Sport students ameliorated SVJ by 2.0 cm (n=96) and females by 1.1 cm (N=45). The results show that the action verb pronunciation may stimulate a complex motor act performance in sportspersons, but with differences specifically related to the disciplines and between the genders without a clear cut effect of the maternal language.

P3.186

Medial prefrontal cortex inactivation disrupts recent spatial memory retrieval in the water maze: art, fact or artefact?

Cassel, J.-C. (Strasbourg)[1], Cholvin, T. (Strasbourg)[1], Loureiro, M. (Strasbourg)[1], Cassel, R. (Strasbourg)[1], Cosquer, B. (Strasbourg)[1], Geiger, K. (Strasbourg)[1], Kelche, C. (Strasbourg)[1], Pereira de Vasconcelos, A. (Strasbourg)[1]
[1]LNCA UMR 7364 UdS-CNRS, Strasbourg, France

Systems consolidation models propose that recent memory is initially hippocampus-dependent but becomes partially or completely dependent upon cortical areas, particularly upon the medial prefrontal cortex (mPFC), when remote. The implication of the mPFC in recent declarative-like memory, however, is still debated. We investigated the possible participation of the mPFC in recent memory recall of the Morris water maze task in rats. Different doses of muscimol (MSCI: 0, 30, 50, 80 and 250 ng in 1µL PBS) were used to assess the impact of increasing inactivation of the dorsal hippocampus (dHip) or the mPFC on the retrieval of a learned platform location in a water maze (8 days of training; probe trial after 24 hours). The smallest dose of MSCI (30 ng) had no significant effect on recall, be it injected into the mPFC or the dHip. A MSCI dose as low as 50 ng infused into the dHip, however, disrupted spatial memory retrieval; the same was true for doses of 80 and 250 ng. Infusions of MSCI in the mPFC had no effect on performance in the 0-80 ng dose range, but at 250 ng occasioned an as dramatic impairment as after efficient dHip inactivation. These results, which confirm that the recall of a recently acquired spatial memory requires the dHip, do not exclude a contribution of the mPFC. Alternatively, as the mPFC-induced disruption of performance was found only with the highest dose of MSCI, it might have reflected nonspecific effects due to an excessive diffusion radius of the drug. In previous studies showing memory disruptive effects of mPFC inactivation by MSCI, the doses have most often been 2 to 4 times larger than the largest dose used herein. We therefore suggest that the mPFC is probably not critical to recent spatial memory retrieval. A spreading of MSCI to regions bordering the mPFC because of an excessive dosage vs. smaller doses remaining confined to the mPFC could be a plausible reason for the opposite conclusions reported in the literature.

P3.187

Hippocampal NMDA receptor subunits involvement in memory in juvenile and adult rats

Snitcofsky, M. (Buenos Aires)[1], Baez, M.V. (Buenos Aires)[2], Colettis, N. (Buenos Aires)[2], Cercato, M. (Buenos Aires)[2], Aguirre, A. (Buenos Aires)[2], Cheli, V. (Buenos Aires)[2], Jerusalinsky, D. (Buenos Aires)[2,3]
[1]LaNyN, IBCN, Buenos Aires, Argentina, [2]Medicine School, Buenos Aires University, Buenos Aires, Argentina, [3]CBC, Buenos Aires University, Buenos Aires, Argentina

NMDA (N-methyl-D-aspartate) receptors (R) of the dorsal hippocampus are required to consolidate inhibitory avoidance (IA) memory in rats and their inhibition with the channel blocker MK-801 immediately after IA training, gives rise to amnesia .This retrograde amnesia was prevented by previous exposure to an open field (OF) -two sessions of 3 minutes each, 24 hours apart- that led to habituation of exploration.

Most of the hippocampal NMDAR subtypes contains GluN2A or GluN2B subunits. Inhibition of GluN2B-containing NMDAR by the selective antagonist ifenprodil enabled long-term memory (LTM) formation of IA with a subthreshold (0.3 mA) training in adult rats, but not in juvenile; and caused facilitation when the training was just overtreshold (0.5mA), suggesting that hippocampal GluN2B containing receptors negatively modulate this memory. Intrahippocampal injection of ifenprodil also facilitated memory formation of the hippocampus-dependent object location task in adult rats.
It was previously shown that a 5 min OF session led to STM as well as LTM formation. Although OF habituation was impaired by intrahippocampal MK801, ifenprodil did not alter this memory, suggesting that it does not depend on GluN2B containing receptors.
Hippocampal protein extracts obtained 70 min after rat exposure to a 5 min OF session were analyzed by Westernblot for the NMDAR subunits. While GluN2A significantly increased, GluN2B appeared to remain unchanged, showing similar results to those obtained in other related models (i.e. LTP induction in hippocampal slices).
These results corroborate that, although NMDAR in the dorsal hippocampus are required during IA consolidation, receptor subtypes containing GluN2B seem to inhibit memory consolidation of this tasks. Meanwhile, GluN2A increased when memory consolidation is still going on, as happens during LTP establishment, suggesting that they might be directly involved in synaptic plasticity underlying this process.

P3.188

Syllable breaking after telencephalic cooling unveils the presence of nonlinearly interacting timescales in birdsong motor pathway

Goldin, M.A. (Buenos Aires)[1], Alonso, L.M. (Buenos Aires)[1], Alliende, J.A. (Buenos Aires)[1], Goller, F. (Salt Lake)[2], Mindlin, G.B. (Buenos Aires)[1]
[1]University of Buenos Aires, Physics Department, Buenos Aires, Argentina, [2]University of Utah, Dpartment of Biology, Salt Lake, United States

We study the emergence of complex behaviors in the birdsong motor pathway. Stereotyped motor activities may originate in precise telencephalic control or may arise from its interaction with downstream nuclei. Here we present a minimalistic dynamical model that accounts for the diverse respiratory patterns found in the song of birds. Air-sac pressure gestures are obtained as the output of an excitatory-inhibitory non-linear interaction between two neuron populations, driven by an upstream telencephalic instruction. Considering a simple and precise time pattern, we can build a map of frequency and amplitude of this instruction. This picture, called bifurcation diagram, accounts for most of the syllabic morphologies found in canaries (*Serinus canaria*). It also illustrates patterns of subharmonicity and bifurcation, which makes predictions for how cooling of HVC should affect song. We then tested these predictions by bilateral cooling of the telencephalic nucleus HVC in canaries. Cooling is thought to reduce the frequency of HVC motor output. Cooling resulted in song and subsyringeal air sac pressure patterns that matched the predictions of the model: syllables were stretched initially and syllable breaking occurred upon further cooling.

P3.189

The activation of the septum lateral increased the release of brain histamine

Farias, P. (Santiago)[1], Diaz, J. (Santiago)[1], Ocampo-Garces, A. (Santiago)[1], Torrealba, F. (Santiago)[1]
[1]Universidad Catolica de Chile, Santiago, Chile

The lateral septum (LS) processes the affective significance of sensorial information coming from hippocampus and directs its outputs forward to hypothalamic areas important for motivated, goal-

directed behavior important to survival. The tuberomammillary nucleus of the hypothalamus (TMN), the only source of brain histamine, is one of the principal targets of the LS, and may be implicated in motivation. Brain histamine is important in arousal and alertness. Novelty has motivational effects and can direct and reinforce behavior.
We have proposed that LS could change the activity of histaminergic neurons, modifying histamine release through GABAergic inputs. The reduction in GABAergic input to the TMN should increase vigilance. We first demonstrated using anterograde axonal tracing, immunocytochemistry and electron microscopy that LS input to TMN is indeed GABAergic, and these terminals make symmetric synaptic contacts with TMN dendrites. We measured extracellular histamine and GABA levels in the LS and ventral hypothalamus using microdialysis, and simultaneously made polysomnographic recording to correlate the increase in histamine levels with alertness. Novel objects increased alertness level, increased histamine release and decreased GABA levels in the LS. Reverse microdialysis with 3.5μM muscimol, a GABAA receptor antagonist, in the LS for 30 min did not alter histamine release. Reverse microdialysis into the LS with 0.1mM picrotoxin plus 10mM glutamate increased general arousal.
The LS controls mood and motivation, TMN is important in motivation too, therefore we propose that the LS controls TMN activity and wake state to maintain arousal at levels appropriate for the current behavior.

P3.190

Effect of contingent and no-contigent intra-core metabotropic receptors group I antagonist and group II agonist administration in the stress induced reinstatement in extinguished cocaine-conditioned animals

De Giovanni, L.N. (Cordoba)[1], Virgolini, M.B. (Cordoba)[2], Cancela, L.M. (Cordoba)[2]
[1]National University of Córdoba, IFEC-CONICET, Cordoba, Argentina, [2]IFEC, Cordoba, Argentina

Previous results from our lab showed that the intra-core administration of metabotropic group I (mglu I) antagonist and metabotropic group II (mglu II) agonist blocked the stress-induced reinstatement in an extinguished cocaine-induced conditioned place preference (CPP) in rats. Moreover we determine that animals which were injected systemically with MK 801 (NMDA antagonist) but follow this injection did not were exposed to the CPP apparatus (no contingent administration) did not show the blockade of stress-induced reinstatement. In the present experiments, our goal was to determine if a no contingent intra-core administration of LY 479234 (mglu II agonist) or MPEP (mglu I antagonist) could influence the stress-induced reinstatement. Male Wistar rats (220-300g) were conditioned with cocaine (10 mg/kg i.p.) during four alternated drug/vehicle sessions, and later extinguished with successive vehicle associations. The following day, animals were microinfunded with MPEP (1 ug/side), LY 479234 (0.5 ug/side) or vehicle and, after the administration they were left undisturbed in their home cages. Two days after the same animals were exposed to a 30 min-restraint exposure, and then were tested in the CPP. Results demonstrate that, neither MPEP nor LY 479234 did not exert and influence in the stress-induced reinstatement when the animals were infuded in a non-contingent way. These results suggest that the blockade of stress-induced reinstatement in extinguished cocaine conditioned animals induced by MPEP or LY 479234 requires a contingency relationship, with the CPP exposure. This result support the hypothesis that the involvement of a nucleus accumbens core in the stress-induced reinstatement depends of the contingency between the agonist/antagonist administration and the CPP exposure.
Financial Support: FONCYT, Ministerio de Ciencia y Tecnología, CONICET and SECyT.

P3.191

Hyperactivity and abnormal social and vocal behaviours in *ProSAP1/Shank2* $^{-/-}$ mice, a model of autism spectrum disorders

Ey, E. (Paris)[1,2,3], Leblond, C.S. (Paris)[1,2,3], Faure, P. (Paris)[4], Torquet, N. (Paris)[1,2,3], Le Sourd, A.-M. (Paris)[1,2,3], Bourgeron, T. (Paris)[1,2,3]
[1]Institut Pasteur, Génétique Humaine et Fonctions Cognitives, Paris, France, [2]Institut Pasteur, CNRS UMR 3571, Paris, France, [3]Université Paris Diderot, Sorbonne Paris Cité, Paris, France, [4]Université Paris 6, CNRS UMR 7102, Paris, France

Mutations in genes coding for synaptic proteins were shown to increase susceptibility to autism spectrum disorders (ASD). Among the causative proteins, the cell adhesion molecules neuroligins and neurexins as well as the scaffolding protein PROSAP2/SHANK3 were repeatedly associated in

independent patients with ASD. Recently, the scaffolding protein PROSAP1/SHANK2 has also been associated with ASD. Functional impacts of these mutations on synaptic density were also ascertained.
Following these studies, animal models were developed to better characterise the role of susceptibility genes in ASD. In a collaborative study, we analysed transgenic knockout mice lacking *ProSAP1/Shank2*. These mice display abnormal glutamatergic receptor expression and neurotransmission. We investigated the development and the adult behaviour of *ProSAP1/Shank2* $^{-/-}$ mice to understand the contribution of *ProSAP1/Shank2* deletion to symptoms in social, communicative or stereotyped behaviours.
Comparisons were drawn between *ProSAP1/Shank2* $^{-/-}$, *ProSAP1/Shank2* $^{+/-}$ and wild-type littermates. Pups and adults were tested to examine the developmental trajectory of abnormalities. We conducted social interactions tests, recordings of ultrasonic vocalisations, self-grooming and digging measures, as well as general activity and anxiety measures.
Abnormalities in body weight as well as in vocal behaviour emerged in the first two weeks of life of *ProSAP1/Shank2* $^{-/-}$ pups, and persisted during adulthood. Adult *ProSAP1/Shank2* $^{-/-}$ males and females were hyperactive. Only *ProSAP1/Shank2* $^{-/-}$ females groomed themselves for longer time in comparison with their littermates. Impairments in social interactions emerged mostly in free social interactions, and appeared together with abnormalities in usage and structure of ultrasonic vocalisations.
All together, this study suggests that mutations in genes causing ASD in humans can alter glutamatergic neurotransmission and cause alterations in social interactions and communication in mice. Together with other mouse models of ASD, the *ProSAP1/Shank2* $^{-/-}$ mice may provide a comprehensive framework to identify new knowledge-based treatments of ASD.

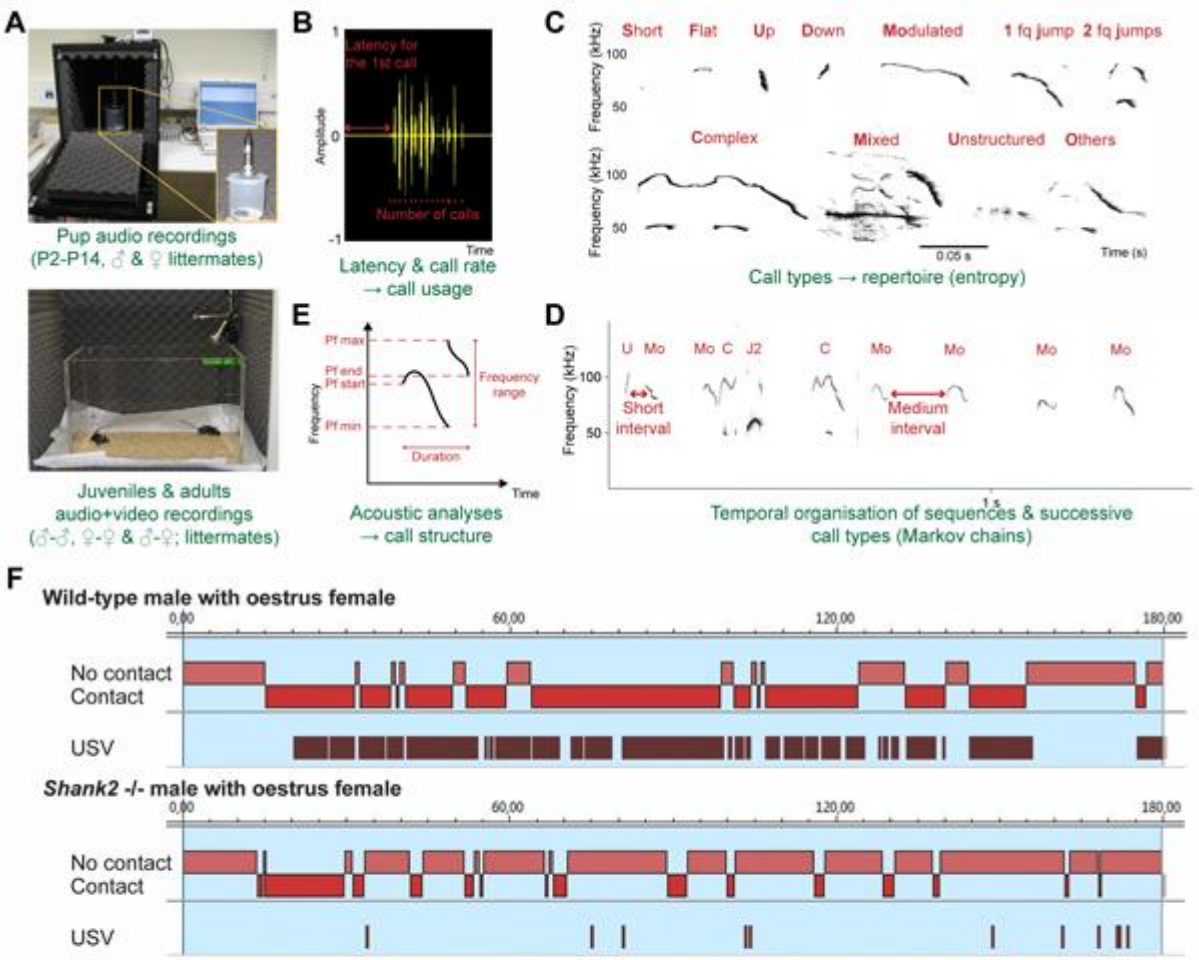

Figure 1: Methods used to analyse the vocal behaviour of mouse models of ASD during development and during social interactions. **A.** Settings to record ultrasonic vocalisations: during development (upper part) and during free social interactions (lower part) in juveniles and adults. **B.** Measurements of the latency for the first call and the call rate on waveform files to characterise call usage. **C.** Spectrograms of the 11 call types used to classify ultrasonic vocalisations to determine the vocal repertoire in each context; entropy is used to compare between contexts or genotypes the global distribution of call types within a repertoire. **D.** Spectrogram of a call sequence to illustrate the analysis of the temporal organisation and succession of calls within a sequence. **E.** Acoustic variables related to duration, peak frequency (Pf) and frequency modulation measured on each call to characterise call acoustic structure. **F.** Combination of social interactions and ultrasonic vocalisations emission during male-female interactions (simple coding with The Observer from Noldus Information Technology, the Netherlands). In wild-type mice (upper panel), most ultrasonic vocalisations (USV) were emitted during physical contacts. In contrast, in *ProSAP1/Shank2* -/- mice (lower panel), the few vocalisations recorded were emitted when animals were apart.

[Methods social/vocal interactions mice]

P3.192

Enhanced emotional reactivity and reduced susceptibility to seizure in mice lacking high-affinity zinc inhibition of NMDA receptors

Ouagazzal, A.-M. (Illkirch)[1], Vergnano, A.M. (Paris)[2], Reiss, D. (Illkirch)[3], Filliol, D. (Illkirch)[3], Neyton, J. (Paris)[2], Paoletti, P. (Paris)[2], Kieffer, B.L. (Illkirch)[4]
[1]IGBMC, CNRS (UMR 7104), Illkirch, France, [2]IBENS, CNRS (UMR 8197), INSERM (U 1024), Paris, France, [3]IGBMC, Illkirch, France, [4]IGBMC, INSERM (U 964), Illkirch, France

Emerging evidence suggests that zinc in its ionic form (Zn^{2+}) acts as a neuronal messenger to regulate brain functions, but the underlying mechanisms are poorly understood. Among the potential synaptic Zn^{2+} targets, the NMDA receptors (NMDARs) containing the GluNR2A subunit exhibits an exquisite sensitivity for extracellular Zn^{2}. The GluN2A subunit, which is widely expressed in adult brain, harbors in its N-terminal domain a high-affinity Zn^{2+} binding site that mediates an allosteric inhibition of the NMDAR complex. To address the *in vivo* relevance of Zn^{2+} modulation of NMDARs, we generated knock-in (KI) mice carrying a point mutation (GluN2A-H128S) in the zinc binding site of the GluN2A subunit. We previously demonstrated that the former mutation selectively eliminates high-affinity (nanomolar) zinc inhibition of NMDARs, results in hyperalgesia to radian heat and capsaicin and completely abolishes analgesia induced by exogenous Zn^{2+} under acute and chronic pain conditions (Nozaki et al., 2011, Nature Neurosci, 14:1017-1022). In the present study we explored whether the elimination of GluN2A Zn^{2+} site impacts higher brain functions. KI mice had a normal phenotype in various tasks assessing social behavior, a simple form of non-associative learning (habituation to novel environment), working memory (spontaneous alternation in a Y-maze) and aversive learning that requires instrumental response (two-way active avoidance). By contrast, KI mice displayed increased anxiety-like behavior in the elevated plus-maze test and enhanced fear responses in the fear conditioning paradigm that was manifested by a high level of freezing during training and contextual testing. Unexpectedly, KI mice were also less susceptible to epileptic seizures and neuronal damage induced by convulsants agents (pentylene-tetrazol and kainic acid). Altogether these data provide the first *in vivo* evidence that inhibition of GluN2A NMDARs by endogenous zinc modulates expression of fear-related behavior, seizure susceptibility and excitotoxicity. They further suggest that alterations of synaptic Zn^{2+} homeostasis may contribute to the pathogenesis of psychiatric and neurological disorders, such as anxiety, epilepsy and dementias.

P3.193

Influence of tVTA over substantia nigra dopamine neurons activity

Sanchezcatalan, M.J. (Strasbourg)[1], Bourdy, R. (Strasbourg)[2], Veinante, P. (Strasbourg)[2], Freund-Mercier, M.J. (Strasbourg)[2], Georges, F. (Bordeaux)[3], Barrot, M. (Strasbourg)[1]
[1]CNRS 3212, INCI, Nociception et Douleur, Strasbourg, France, [2]CNRS 3212, INCI, Univ. Strasbourg, Nociception et Douleur, Strasbourg, France, [3]CNRS, Institut Interdisciplinaire de Neurosciences, Bordeaux, France

The tail of the ventral tegmental area (tVTA) is a GABAergic mesopontine structure, which projects heavily to midbrain dopamine neurons. This connectivity suggests that the tVTA could regulate the activity of dopamine neurons, so it has been proposed as a major brake for dopamine systems. Several neuroanatomical and electrophysiological studies have demonstrated the VTA-DA neurons/ tVTA connection, showing that the tVTA stimulation is able to decrease dopamine activity in VTA, whereas tVTA inhibition increases it. Nevertheless, there are fewer studies concerning the relation of tVTA over substantia nigra (SN) dopamine neurons. Concretely, it has been previously demonstrated in our laboratory that SN neurons projecting to the dorsal striatum (DS) receive apposition of fibers arising from the tVTA, showing a tVTA/SNc/DS circuitry. Moreover, the electrical or chemical tVTA stimulation is able to inhibit SN dopamine neurons, whereas tVTA inhibition increases it.
Thus, in the present study, we have explored the basal firing rate of substantia nigra pars compacta dopamine neurons in Sprague-Dawley rats with bilateral excitotoxic lesions of tVTA. To this end, we used extracellular recording techniques in anesthetized animals. On the other hand, we have investigated, in another group of animals, the consequences of the bilateral excitotoxic lesion of the tVTA on motor performance in the rotarod test. At the end of the experiments, rats were deeply anesthetized and brains were removed for the verification of the tVTA lesion and SNc recordings. Our results demonstrate that the basal firing rate of SN dopamine neurons is higher in animals with bilateral tVTA lesion, indicating that the tVTA exerts basal inhibitory control over SN dopamine neurons. In addition, bilateral lesions of the tVTA led to an increase in motor performances and further in motor skills learning. These electrophysiological and behavioral data suggest that the tVTA exerts an inhibitoy control over SN dopamine neurons, as well as, the tVTA may trigger or modulate motor responses.
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P3.194

Partitioning stem from options with color cues improves reading and solving of multiple-choice questions

Lions, S. (Valparaiso)[1], Pena Garay, M. (Santiago)[2]
[1]*Universidad de Valparaiso, Centro Interdisciplinario de Neurociencias, Valparaiso, Chile,*
[2]*Universidad Catolica de Chile, Santiago, Chile*

Color contributes to visual processing through scene segmentation. We then explored how color cues could improve reading and solving of multiple-choice questions. Two groups of secondary students solved simple mental arithmetic multiple-choice questions presented as text on a computer screen. In the Yellow-Group, options appeared on white background but the stem appeared on yellow while in the White-Group, both question's parts appeared on white background. We found a significantly higher performance in the Yellow-Group. This effect was mainly observed in students with a history of lower academic reading scores. It was associated with a lower frequency of premature stem-to-options saccades occurred before a full exploration of the stem took place, and to a longer latency when providing incorrect answers. Our results suggest that color cues may improve reading and solving of multiple-choice questions because color assists in segmenting the text into two functionally distinct parts that are more exhaustively explored in space and time.

P3.195

Exploring the role of dorsal streams in prehension: natural and pantomimed grasping movements in central vs. peripheral vision

Fukui, T. (Bron)[1], Revol, P. (Bron)[1], Salemme, R. (Bron)[1], Pisella, L. (Bron)[1], Rossetti, Y. (Bron)[1]
[1]*Centre de Recherche en Neurosciences de Lyon, INSERM U 1028 - CNRS UMR 5292 - Univ. Lyon 1, Equipe Impact, Bron, France*

To study how visual perception influences action execution, we investigated kinematic properties of natural and pantomimed reach-to-grasp movements by manipulating object familiarity and gaze direction. Patient I.G. with a lesion of bilateral posterior parietal cortex and control participants were required to perform a pantomimed prehension task followed by a real grasping task. In the pantomime session, participants gazed at a target object or a fixation point (20 deg leftward deviated from object position) and were subjected to a 3-s visual occlusion (manipulated by crystal shutter goggles), during which the experimenter removed the object. The participants were then required to pretend to make a reach-to-grasp action toward the location where the object had been presented. In the real grasping session, a 3-s delay was also inserted in this session, as well as in the pantomimed session, and the target object to be grasped was not removed during this delay. The vision during the movement was provided. Gaze direction was fixed during a trial including visual occlusion period. We used a can of juice, a can of cachou, and a battery cell (size AA) as familiar objects and same-shape gray objects as non-familiar objects. Gaze condition (central and peripheral visual conditions) was blocked and each object was presented in a random fashion. We calculated the slope of the regression between peak grip aperture and object size. Patient I.G. data were very different between real and pantomimed grasping, which was not the case for controls except for a common lack of influence of gaze direction on pantomimed and not on real grasping. In real grasping, both controls and patient I.G. showed smaller values when the object was presented in peripheral vision than when it was presented in central vision. Object familiarity seemed to affect the performance of the control participants, but not patient I.G. These results are discussed in the framework of dorsal (dorso-dorsal / ventro-dorsal) / ventral streams theory.

P3.196

Context is essential to expression of amphetamine-induced locomotor sensitization, but its long lasting maintenance depends on abstinence time

Engelke, D.S. (Sao Paulo)[1], Karcher, D. (Sao Paulo)[1], Filev, R. (Sao Paulo)[1], Santos-Junior, J.G. (Sao Paulo)[2], Mello, L.E. (Sao Paulo)[1]
[1]Federal University of São Paulo (UNIFESP), Sao Paulo, Brazil, [2]School of Medical Science of Santa Casa of São Paulo, Sao Paulo, Brazil

Animal models of drug addiction are very important to understand the neural basis of this illness that comprises several mechanisms of neural adaptations. Locomotor sensitization has been extensively used to investigate incentive salience and the transition from use to abuse. Furthermore, there are several studies reporting the influence of contextual aspects in the acquisition and expression of locomotor sensitization. However, these studies do not explore whether its maintenance is context specific. Thus, the aim of this study was to evaluate the contextual influence in the amphetamine-induced locomotor sensitization and how long it is maintained. First, adult male C57bl/6 mice were individually maintained in a contextual activity box for 40 min (baseline activity). They were divided in three groups, according to the treatment performed during five consecutive days: CTX+AMPH (N=10) - animals treated with amphetamine (1mg/kg, i.p) and, after each injection, submitted to the contextual activity box; AMPH (N=10) - animals treated similarly to the previous group, however, in acquisition days they received the injection and came back to home cage; CTRL (N=10) - animals treated daily with saline and submitted to the contextual activity box every day. In acquisition phase, locomotor activities were accessed for 40 min just after the first and last injections. After three (3), fourteen (14) and twenty eight (28) withdrawal days, all animals were challenged with amphetamine (1mg/Kg) and the expression of locomotor sensitization was evaluated. The group CTX+AMPH presented significantly higher locomotor activity when compared with the other experimental groups at the 3rd and 14th (but not 28th) day. We suggest that contextual factors during acquisition phase play a role in the expression of locomotor sensitization. Moreover, the lack of differences at the 28th day of withdrawal could be explained by memory decay, which leads to impairment in the recognition of the context. This result suggests that long term effects of abstinence could be due memory decay triggering the drug seeking behavior.

P3.197

Effects of cold exposure on behavioral and electrophysiological parameters related with hippocampal function in rats

El Marzouki, H. (Kenitra)[1], Aboussaleh, Y. (Kenitra)[2], Suer, C. (Kayseri)[3], Dolu, N. (Kayseri)[3], Artis, A.S. (Istanbul)[4]
[1]Universite Ibn Tofail, Neurosciences et Nutrition, Kenitra, Morocco, [2]Université Ibn Tofail, Faculté des sciences, Neurosciences et Nutrition, Kenitra, Morocco, [3]Université Erciyes, Faculté de médecine, Physiologie, Kayseri, Turkey, [4]Université Medeniyet, Faculté de médecine, Physiologie, Istanbul, Turkey

Physical or psychological stress induces a rapid reaction activating of the autonomous nervous system leading to release of adrenaline from the adrenal medulla;and slower response activating of the hypothalamus-pituitary-adrenal axis leading to secretion of glucocorticoids.The most important target of glucocorticoids is the hippocampus which has the highest concentration of receptors in the brain.The Morris water maze test has been widely used to test the spatial learning performance of rats.Long-term potentiation is manifested as increased synaptic efficiency lasting over an hour that is

induced by brief high-frequency stimulation of presynaptic neurons and widely accepted as a model of the synaptic plasticity that is thought to underlie learning and memory processes.
Our experiments were designed to investigate and to compare the effects of high ACTH levels,achieved by cold stress application, on spatial memory performance and synaptic plasticity in the dentate gyrus. In this study male Wistar rats were divided into 3 groups: the control,15 min and 2h cold stress groups.The animals in the cold-stress group were placed in a cold room for 15 min/day or 2 h/day for 5 days between 8:00 a.m. and 10:00 a.m. to avoid corticosterone circadian rhythm.Control animals were acclimatized to standard animal laboratory conditions.All rats were housed in a room under a 12/12 h light-dark cycle.Morris water maze and long-term potentiation recordings were taken and blood was obtained for ACTH measurements.
The ACTH levels of the cold stress groups were significantly higher than the control group.The results for the MWM testing demonstrate that escape latency and distance moved were not significantly different in the cold stressed groups from the control group at the end of training period.However cold stressed rats spent more less time in target quadrants than the control rats in probe trail.The LTP responses obtained by perforant path stimulation were found to be more depressed in the cold stressed rats than those in the control rats.These findings indicate that the exposure to cold stress affects aspects of local circuit activity and plasticity in the dentate gyrus.It is possible that these alterations underlie some of the behavioral consequences of the stress experience.

P3.198

Drug reinforcement impairs declarative-like memory while promoting striatum-dependent learning

Husson, M. (Talence)[1], Baudonnat, M. (Talence)[1], Guillou, J.-L. (Talence)[1], Vandesquille, M. (Talence)[1], Corio, M. (Talence)[1], Crespin, M.-L. (Talence)[1], Béracochéa, D. (Talence)[1], Mons, N. (Talence)[1], David, V. (Talence)[1]
[1]*Univ. de Bordeaux I, Incia - Cnrs Umr 5287, Talence, France*

The multiple memory systems hypothesis posits that different neural circuits function in parallel and may compete for information processing and storage. For example, instrumental conditioning depends on the striatum, whereas spatial memory is processed by a circuit centered on the hippocampus. However, the nature of the task itself is not sufficient to select one system over the other. As compared to aversive events, the impact of rewards on interactions between memory systems remain poorly understood. We have implemented an experimental set-up to compare the long-term effects of food (crisps) and drug (intra-VTA morphine) rewards on various forms of memory. This paradigm is based on a Y-maze discrimination task which can be acquired using either a cued or a spatial learning strategy. Subsequent use of cued and spatial learning strategies are further assessed using a competitive version of the Morris water-maze task. Behavioral testing was completed with brain analysis of the phosphorylated form of cAMP Response Element Binding (pCREB) protein, and recording of field potentials evoked by stimulation of the amygdalo-striatal and amygdalo-hippocampal pathways in freely-moving mice. We found that drug-induced activation of the reward system impaired spatial but not cued memory. This spatial impairment was related to a dramatic decrease in pCREB expression within the dorsal hippocampus (CA1 and CA3 subfields) and the prefrontal cortex. Interestingly, both food and drug rewards activated CREB in the ventral striatum (nucleus accumbens). In contrast, only drug reward persistently upregulated CREB expression within the dorsomedial striatum for at least 72h. CREB upregulation was related to a potentiation of neuronal activity in the amygdalo-striatal pathway, and a persistent use of cued-learning strategy in the water-maze. Decreasing (Rp-cAMPS) CREB activity within the dorsal striatum prevented the spatial deficit. We conclude that drug-induced activation of the reward system has a negative impact on hippocampus-dependent memory and related signaling pathways. This declarative-like deficit, together with a facilitated control of behavior by conditioning processes, could contribute to the instatement of addictive behaviors.

P3.199

Neural networks underlying liking and wanting responses to food odors in healthy women

Jiang, T. (Lyon)[1], Soussignan, R. (Dijon)[2], Schaal, B. (Dijon)[2], Carrier, E. (Lyon)[3], Royet, J.P. (Lyon)[1]
[1]INSERM U 1028 - CNRS UMR 5292 - CRNL, Centre de Recherche en Neuroscience de Lyon, Equipe Codage et Mémoire Olfactive, Lyon, France, [2]Centre de Sciences du Goût et de l'Alimentation, Dijon, France, [3]Clinique St Vincent de Paul, Lyon, France

The current way of life in industrialized countries with abundant, palatable foods of easy access, is considered as a major drive of overeating and obesity. In this “obesogenic environment”, food intake does not only satisfy energetic needs, but also depends on psychological and societal demands. Individual reactions to foods are largely controlled by anticipatory and consummatory reward responses, that control both quantitative and qualitative intake. Recent studies on the neural substrates of reward responses in rodents have revealed different processes involved in reward processing, such as liking and wanting and both of these processes can be dissociated at the psychological level in humans as well. However, the neural substrates underlying them remain unclearly defined. As odor cues of food participate into both anticipatory (before ingestion) and consummatory (during ingestion) reward processes the study of the brain substrates of odor cued reward responses could extend our understanding of reward neural underpinnings. In this aim, we have examined the brain correlates of liking and wanting to food odors using event-related fMRI in 12 healthy women. Each participant was scanned during two sessions over two consecutive days, one day in the pre-prandial state and another day in the post-prandial state. During each session, BOLD signals in responses to liking and wanting judgments were acquired in two separate runs. Liking and wanting judgments were rated for each food odor with a five key-press button box. Functional analyses were focused on regions of interest involved in reward processing (*e.g.,* striatum, orbitofrontal cortex). Preliminary results indicate that liking and wanting tasks were processed by partially distinct neural networks that are variables as a function of the odor stimulus (food *vs*. no-food) and the participant's motivational state (hungry *vs*. satiated).

P3.200

Mice gamble for food reward: inter-individual differences and behavioural profiles

Granon, S. (Orsay)[1], Pittaras, E. (Brétigny-sur-Orge)[2], Cressant, A. (Orsay)[3], Serreau, P. (Orsay)[4], Bruijel, J. (Orsay)[4], Dellu-Hagedorn, F. (Bordeaux)[5], Rabat, A. (Brétigny-sur-Orge)[2]
[1]CNPS, UMR 8195, CNRS, Centre de Neuroscience Paris Sud, Orsay, France, [2]IRBA Institut de Recherche Biomédiale des Armées, ENOP, Brétigny-sur-Orge, France, [3]CNRS UMR 8195, Orsay, France, [4]CNPS, UMR 8195, CNRS, Orsay, France, [5]CNRS, INCIA, UMR 5287, Bordeaux, France

Background: One of key questions in Neuroscience is to understand how we choose one option instead of another one when we are in uncertain or ambiguous situation.
We know that our decisions have short- and long-term consequences. Therefore, it may be better to favour a small immediate profit in order to get one more important later. The Iowa Gambling Task (IGT; Bechara et al., 1994) has been developed to study such real-life decision-making process since subject wins (reward) or loses (penalty) money, both happening with unpredictable and variable probabilities. Developing a rodent version of this task allows 1) the study of brain circuits, pharmacological or molecular mechanisms that impact on decision-making processes and 2) the identification of inter-individual differences.
Methods: 72 male C57Bl/6J mice were used to adapt the gambling task. We first adapted the task in an operant chamber from rat's works (Rivalan et al., 2009). We tried two different penalties and two different rewards. As our results were not conclusive, we adapted the task to a maze version, inspired by previous works (Van Den Bos et al., 2006). We modified the protocol in order to match as closely as possible the human task. In addition, we performed different other behavioural tasks to measure anxiety, locomotion and social behaviours.

Results: First we showed that transferring directly the rat gambling task to mice was not successful because of species particularities. For example, we observed that mice were not sensitive to delay penalties, as rats were. In the maze version, where quinine pellets were used as penalties, we found that mice showed a clear preference for small immediate rewards to maximize their benefits in the long term. We also found inter-individuals differences and specific behavioural traits for each subgroup.
Conclusion: We were able to design a Mouse Gambling Task (MGT). Like in humans and rats, despite the uncertainty of the options, the choices made by the mice gradually evolved toward favourable options, and inter-individuals differences emerged. We discriminated subgroups based on individual MGT performance and characterised them behaviourally in order to establish behavioural profiles.

P3.201

Role of the *Caps1* gene on neurohormone secretion and mouse behavior

Kappeler, C. (Créteil)[1], Grannec, G. (Paris)[2], Brose, N. (Göttingen)[3], Giros, B. (Paris)[2], Leboyer, M. (Créteil)[1,4,5], Nosten-Bertrand, M. (Paris)[2], Jamain, S. (Créteil)[1]
[1]Psychiatrie Génétique, INSERM U 955, Hôpital H. Mondor, Créteil, France, [2]Physiopathologie des Maladies du SNC, INSERM U 952, CNRS-UMR 7224, UPMC, Paris, France, [3]Department of Molecular Neurobiology, Max Planck Institute of Experimental Medicine, Göttingen, Germany, [4]Université Paris Est, Faculté de Médecine, Créteil, France, [5]AP-HP, Hôpital H. Mondor – A. Chenevier, Pôle de Psychiatrie, Créteil, France

Bipolar disorder (BD) is a complex psychiatric disorder characterized by alternating periods of mania and depression, each period corresponding to important fluctuations in mood, appetite, sleep and energy levels.
We recently carried out a whole-genome linkage analysis and showed that the 20p12 and the 3p14 regions were more frequently shared by sib-pairs affected by early-onset BD than expected by chance. The gene encoding the synaptosomal associated protein SNAP25, essential for neurotransmitter release, is located in the 20p12 region and has been associated with BD. Our team recently identified a variant, within the *SNAP25* promoter region, more frequent in patients with early-onset BD and associated with a higher level of *SNAP25* mRNA in the prefrontal cortex. Interestingly, the Calcium-dependent activator protein for secretion (*CADPS*) gene is located in the 3p14 region and encodes a protein involved in the release of catecholamine and monoamines that directly interacts with SNAP25.
Since abnormal functioning of synaptic transmission likely plays a role in the etiology of psychiatric disorders, we initiated a behavioral and physiological characterization of the *Caps1* knockout mice. We particularly focus on phenotypes that are relevant for BD symptoms, including altered circadian rhythms, hyperactivity, anxiety, and depressive behaviors.
The hypothalamo-pituitary-adrenal (HPA) axis is a key regulator for mood, anxiety and sexuality behaviors that are impaired in BD patients. Abnormal levels of corticotrophin releasing hormone and cortisol have been described in psychotic disorders including BD. We have thus specifically tested the HPA axis by measuring neurohormone secretion combined to behavioral analysis in the *Caps1* knockout mice maintained under normal conditions or in response to stress.

P3.202

Fos imaging of age-dependent effects of estradiol on working memory

Al Abed, S. (Bordeaux)[1], Sellami, A. (Bordeaux)[1], Brayda-Bruno, L. (Bordeaux)[1], Lamothe, V. (Bordeaux)[1], Potier, M. (Bordeaux)[1], Bennetau-Pelissero, C. (Bordeaux)[1], Noguès, X. (Bordeaux)[1], Marighetto, A. (Bordeaux)[1]
[1]*NeuroCentre Magendie INSERM U 862, Bordeaux, France*

In addition to its effects on the reproductive axis, the hormone 17β estradiol (E2) may play a role in cognition and potentially exert protective effects against aging-related memory disturbances. E2 can indeed increase GluA receptor expression, dendritic spine density and LTP, and thereby counteract aging effects on certain aspects of morphological and functional plasticity of hippocampal neurons. However, the effects of E2 on memory are far to be homogeneous among studies. Our hypothesis is that E2 effect may depend on both the age of the subject and the form of memory concerned. We recently found in mice that a chronic (water-drinking) treatment with E2 alleviated the aging-related impairment in long-term memory and had no effect on this memory in adulthood. Here we explored the effects of the same treatment on short-term/working memory. Young and aged mice were trained in a radial-maze task assessing both retention and organization (proactive interference) of working memory. Fos immunodetection in hippocampal, prefrontal, striatal and amygdalar areas was performed after the last training session. Behavioural and Fos data converged to support our hypothesis. In contrast to its effect on long-term memory, E2 failed to improve the working memory impairment seen in aged mice but significantly affected retention and organization of working memory in young mice: retention was enhanced but organization was reduced. At the brain level, analyses of Fos activation patterns showed that E2 altered hippocampal function only: it reduced DG hyperactivation in aged mice (same tendency in CA3), but produced overactivation of the same field in young mice. Analyses of Fos between-structure correlation matrices confirmed that E2 also affect functional connectivity in an opposite direction depending on the age. Present diversity of E2 effects may be explained by its capability to enhance LTP (reduce LTD), and highlights the need for integrative approaches to improve our understanding of the role of estrogens in cognitive aging.

P3.203
Eye movements as an excellent probe of multi-stable motion direction perception

Meso, A. (Marseille)[1], Masson, G. (Marseille)[1]
[1]*Institut de Neuroscience de la Timone, CNRS/Aix-Marseille Université, Marseille, France*

Eye movements are thought to provide an informative window into cognitive and perceptual processes. We exploited this in psychophysical experiments with multi-stable motion stimuli in which observers perceived direction dynamically. Given a continuous space over which a percept varies i.e. from horizontal through diagonal to vertical motion directions, explicit perceptual reports are limited to instances in time and constrained by the context in which the reports are made, normally a discrete forced choice. To probe the useful information within the eye movements, we used a slow moving barber pole stimulus (6deg/s), with an obliquely oriented sinusoidal luminance grating presented behind a square aperture generating the multi-stability. First we sought to identify what component of the eye movements, was coupled with perception during both short presentations (< 1s) and extended trials over which switches occurred (15s). We used acceleration and speed criteria to identify blinks, saccades and smooth components (containing both smooth pursuit and ocular following responses). We found these smooth components to be coupled to perceived directions at the end of short trials and also to the sparsely sampled transitions during the longer trials. Second, we found that the question of which precedes the other between the eyes and the reports showed individual differences probably due to transformations from neural representation to motor response. Third, the eyes provided a rich source of information in estimated eye directions and their distributions over trial conditions, indicating where noise driven transitions were more likely. Eye movements are therefore a very useful and so far underused probe of multi-stable perception.

P3.204

Don't be too strict with yourself! Rigid negative self-representation in healthy subjects mimics neurocognitive profile of depression for autobiographical memory

Piolino, P. (Paris)[1], Sperduti, M. (Paris)[1], Martinelli, P. (Paris)[1], Kalenzaga, S. (Paris)[1], Gallarda, T. (Paris)[1], Amado, I. (Paris)[1], Krebs, M.-O. (Paris)[1], Oppenheim, C. (Paris)[1]
[1]Université Paris Descartes, Centre de Psychiatrie et Neurosciences Inserm U 894, Paris, France

Autobiographical memory comprises representation of specific (episodic) and generic personal information (semantic). Depression is characterized by a shift from episodic to semantic memories retrieval (overgeneralization). Theoretical models have proposed that this effect could be linked to reduced executive resources. Moreover, overgeneral memories, accompanied by the well known negativity bias lead to a general and pervasive negative self-representation. Interestingly, even in non clinical population executive function and autobiographical memory specificity are closely linked, and in turns overgeneral memories could lead to depressive responses. We made the hypothesis that healthy subjects showing a rigid negative self-image would mimic the neurocognitive profile of patients. We measured executive functions and the self-image in 20 young healthy subjects, and recorded their brain activity, by means of fMRI, while they retrieved episodic autobiographical memories. We reported that a more rigid negative self-image corresponded to lower executive functions and less vivid memories. Moreover higher negative self-image resulted in attenuated activity in the orbitofrontal and the anterior cingulate cortex that contrarily positively correlated with executive and memories performances. These results are in line with role of these regions in executive functions and autobiographical memory retrieval. More importantly these regions have been constantly shown to exhibit altered functionality and connectivity in depression. We proposed that rigid negative self-image could represent a marker or a vulnerability trait of depression that is linked to reduced executive functions efficiency and indirectly to episodic autobiographical memory decline. Our results are encouraging for psychotherapeutic approaches promoting cognitive flexibility in depression and other psychiatric disorders.

P3.205

Effect of chronic pharmacological activation of oxytocin receptors on the behavioral and molecular phenotypes in the prenatal stress rat model of psychiatric disorders

Mairesse, J. (Villeneuve d'Ascq)[1], Gatta, E. (Villeneuve d'Ascq)[1], Morley-Fletcher, S. (Villeneuve d'Ascq)[1], Soichot, M. (Lille)[2], Reynaert, M.-L. (Villeneuve d'Ascq)[1], Marrocco, J. (Messina)[3], Deruyter, L. (Villeneuve d'Ascq)[1], Van Camp, G. (Villeneuve d'Ascq)[1], Bouwalerh, H. (Villeneuve d'Ascq)[1], Pittaluga, A. (Genova)[4], Allorge, D. (Lille)[2], Nicoletti, F. (Rome)[1,5,6], Maccari, S. (Villeneuve d'Ascq)[1]
[1]University Lille North of France, UMR 8576 CNRS, Neuroplasticity Team, Villeneuve d'Ascq, France, [2]Faculté de Médecine de Lille, UDSL, Lille, France, [3]IRCCS Centro Neurolesi 'Bonino Pulejo', Messina, Italy, [4]University of Genova, Dept Exptl. Medecine, Genova, Italy, [5]University Sapienza, Dept Physiology and Pharmacology, Rome, Italy, [6]IRRCS, Neuromed, Pozzili, Italy

Activation of oxytocin receptors in the SNC is a new potential strategy in the treatment of psychiatric disorders involving emotions and social behavior. Prenatal restraint stress (PRS) in rats programs the offspring to develop a pathological behavioral phenotype associated with abnormalities of the hypothalamic-pituitary-adrenal (HPA) axis. Chronic systemic treatment of PRS rats with the oxytocin receptor agonist, carbetocin (1 mg/kg daily for 3 weeks), normalized the HPA response to a mild stress and the related changes in the expression of corticosteroid receptors and type-2 11β-hydroxysteroid dehydrogenase in the hippocampus. All pathological behaviors of PRS rats (anxiety- and depressive-like behavior, and impaired social behavior and social memory) were corrected by carbetocin treatment. The negative correlation between anxiety-like behavior and social behavior in the entire cohort of rats suggested that reduction of anxiety lies at the core of the “therapeutic effect” of carbetocin in PRS rats. Carbetocin treatment also corrected the defect of depolarization-evoked glutamate release in ventral hippocampal synaptosomes of PRS rats, a neurochemical parameter that showed a tight correlation with both anxiety and social behavior. Remarkably, carbetocin had little, if

any, effect in unstressed control rats, suggesting that its action was “disease dependent”. Thus, at least in this particular setting, activation of oxytocin receptors had no impact on normal brain functioning but corrected the pathological program induced by early life stress. These data strongly encourage the use of oxytocin receptor agonists in the treatment of stress-related disorders.

P3.206

Role of retinoid X receptors in control stress adaptation processes in social defeat model

Krezel, W. (Illkirch)[1], Podlesny, A. (Wroclaw)[2], Krzyzosiak, A. (Illkirch)[3]
[1]CERBM-GIE, IGBMC,Département de Neurobiologie et Génétique, Illkirch, France, [2]Technical University of Wrocalw, Wroclaw, Poland, [3]IGBMC, Illkirch, France

Preclinical genetic and pharmacological studies indicated that compromised signaling through retinoid X receptor (Rxr) gamma in the nucleus accumbens (NAc) is associated with depressive-like behaviors. In order to understand physiological functions of Rxr gamma in stress adaptation processes and its implication in the etiology of depressive-like behaviors we studied its signaling in mouse model of social defeat stress. We found that in wild type C57BL6N mice stress led to hyperactivity of NAc shell as monitored by enhanced expression of several early responsive genes which were also identified by DNA microarray analyses among the ten top transcripts increased by genetic ablation of Rxrg. Absence of reduced expression of Rxrg in response to chronic stress suggests its up-stream position in the process of stress-related adaptive changes in the NAc. The effects of Rxr-specific pharmacological treatments in stressed mice should further reveal the role of Rxrs in mechanisms of stress adaptation. Potential role of Rxrs in modulation of stress adaptation processes may be instrumental for understanding of clinical depression associated with abnormal signaling of isotretinoin or n-3 polyunsaturated fatty acids, the pharmacological modulators of transcriptional activities of Rxrs.