SAN2011

XXVI Congreso Anual
de la Sociedad Argentina
de Investigación en Neurociencia

Huerta Grande, Córdoba
18-22 Octubre 2011.

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Course | Organizing Committee

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Course Program

"Electrophysiological Recordings and Optical Imaging in Neuroscience"

Tuesday, October 18th | Day 1: Electrophysiology

8.00	**Registration**
09:00 - 11:00	**Lecture I** *"Membrane potential, action potential, excitatory and inhibitory synaptic currents"* **Mariano Casado,** *Neuroscience Section, IBEns (CNRS UMR 8197 / INSERM U 1024) Ecole Normale Supérieure, Paris, France*
11:05 - 11:20	**Coffee break**
11:30 - 12:55	**Lecture II** *"Extracellular and intracellular solutions, basic properties, ions"* **David Ogden,** *Laboratoire de Physiologie cérébrale, UMR8118, Université Paris Descartes, Paris, France*
13.00 - 14:00	**Lunch**
14:00 - 18:00	**Lecture III** *"Patch clamp techniques: voltage clamp, current clamp, perforated patch"* **Mariano Casado & Marco Diana,** *Institut de Biologie. CNRS UMR 8197 – INSERM U 1024.Ecole Normale Supérieure, Paris, France*
18:00 - 18:30	**Coffee break**
18:30 - 20:30	**Lecture IV "***Electronics, the construction of a (basic) amplifier and test with model cell"* **Boris Barbour,** *Neuroscience Section, IBEns (CNRS UMR 8197 / INSERM U 1024) Ecole Normale Supérieure, Paris, France & David Ogden*
20:45 - 21:15	**Discussion. Round table**
21.30 - 22:30	**Dinner**

Wednesday, October 19th | Day 2: Imaging

08:15 - 09:15	**Breakfast Meeting**
09:15 - 11:15	**Lecture V** *"Introduction to conventional and confocal microscopy"* **Stepahne Dieudonné,** *Institut de Biologie, CNRS UMR 8197 INSERM U 1024. Ecole Normale Supérieure, Paris, France*
11:15- 11:30	**Coffee break**
11:30 - 13:30	**Lecture VI** *"Introduction to 2-photon microscopy"* **Stepahne Dieudonné**
13.45 - 14:30	**Lunch**
15:00 - 17:00	**Lecture VII** *"Caged compunds and Ch- Rhodopsins. Principles, compounds"* **David Ogden & Marco Diana**
17:30 - 18:45	**Lecture VII** *"Voltage sensitive dyes: principles – compounds – applications"* **David Ogden**
19:10 - 20:30	**9th lecture.** *"Data analysis, programs"* **Boris Barbour**
20:45 - 21:30	**Round Table**
21.45 - 22:45	**Dinner**

Meeting Program

Thursday, October 20th

8.00 **Registration**

9:00 - 9:15 **Opening**

9:30 - 12:20 **Symposium I. Non classical synaptic transmission**

- **Gabor Tamas.** *Department of Physiology, Anatomy and Neuroscience, University of Szeged. Hungary.* *"Unitary volume transmission by neurogliaform cells: broadening the functional scope of single neurons"*
- **Ian Forsythe.** *MRC Toxicology Unit. University of Leicester Lancaster Road, Leicester LE1 9HN, UK.* *"The role of nitric oxide in neuronal homeostasis and intrinsic plasticity"*
- **David Ogden.** *Laboratoire de Physiologie cérébrale, UMR8118, Université Paris Descartes, France.* *"Metabotropic glutamate receptor signalling in cerebellar Purkinje neurons studied with flash photolysis"*
- **Boris Barbour.** *Neuroscience Section, IBEns (CNRS UMR 8197 / INSERM U 1024) Ecole Normale Supérieure, France.* *"Pure spillover transmission in the cerebellum"*

12:30 **Lunch**

14:30 - 16:20 **Young Investigators Colloquium I**

Fabricio Ballarini. *Instituto de Biología Celular y Neurociencias , Fac. de Medicina, UBA, Argentina.* *"Novel lessons improve memory in elementary school children: evidence of behavioral tagging in humans"*

Mario Perello. *Instituto Multidisciplinario de Biología Celular (IMBICE) (CONICET/CICPBA), Argentina.* *"Ghrelin mediates stress-induced food reward behavior in mice"*

Lionel Muller Igaz. *Grupo de Neurociencia de Sistemas, Dpto. Fisiologia, Facultad de Medicina, UBA, Argentina.* *"Novel animal models to study the role of TDP-43 in neurodegenerative disease"*

Estefanía Bello. *INGEBI-CONICET, Argentina.* *"Mice lacking dopamine D2 autoreceptors reveal their fundamental role on dopamine neurotransmission"*

16:30 - 19:00 **Poster session I**

19:30 **"De Robertis" Lecture.** **Martín Giurfa** *Centre Recherches Cognition*

Animale, CNRS, Université Paul Sabatier, Toulouse, France. «Accessing the neural bases of cognitive processing in a simple brain (that turns to be not so simple)»

21:00	**Dinner**
23:00	**Bonfire**

Friday, October 21st

8:30 - 10:20 **Young Investigators Colloquium II**

Fernando Sepúlveda. *Departamento de Fisiología, Universidad de Concepción, Chile. "Membrane perforation induced by Aß is modulated by membrane levels of NR2B and LRP6 protein"*

Tomás Falzone. *Instituto de Biología Celular y Neurociencia IBCN-CONICET-UBA, Argentina. "Abnormal endo-lysosomal membrane dynamics and retrograde axonal transport defects induced by proteasome inhibition"*

Jesica Raingo. *Instituto Multidisciplinario de Biología Celular (IMBICE) (CONICET/CICPBA), Argentina "Alternative splicing of calcium channels adjust neuronal activity"*

Esteban Beckwith. *Fundación Instituto Leloir-IIBBA. CONICET, Argentina. "Retrograde BMP signaling modulates the pace of the circadian clock"*

10:30 - 13:00	**Poster session II**
13:00	**Lunch**
14:30	**"Ranwell Caputo" Lecture. Santiago Quiroga.** *Facultad de Ciencias Químicas. Universidad Nacional de Córdoba, Argentina. "Regulation of membrane expansion at the nerve growth cone: Axonal specification and elongation"*
15:30 - 18:00	**Poster session III**
18:00 - 20:40	**Symposium II "Brain Development and Evolution"**

Pasko Rakic *Department of Neurobiology. Yale University, US. "Evolution of the Neocortex: Evo-Devo approach"*

Diego Gelman. *Instituto de Neurociencias de Alicante, Spain. "Generation of cortical interneuron diversity in the mouse"*

Suzana Herculano-Houzel. *Instituto de Ciências Biomédicas-Universidade Federal do Rio de Janeiro, Brazil. "Building a bigger brain: New views on brain*

	scaling the development and evolution of mammalian brains" **Lucía Franchini.** *Instituto Investigaciones Ingeniería Genética y Biología Molecular, Buenos Aires, Argentina. "Human brain evolution: searching the genetic basis underlying our unique cognitive capacities"*
21:00	**Dinner**
22:00	**Plenary meeting SAN. SfN local chapter**
24:00	**Neuro party**

Saturday, October 22nd

9:30	**Symposium III** *"Applied neuroscience: Music, herbs hypnosis and jokes"* **Draulio Araujo.** *Instituto do Cérebro da UFRN, Universidade Federal do Rio Grande do Norte, Brazil. "Seeing with the Eyes Shut: Neural Basis of Enhanced Imagery following Ayahuasca Ingestion"* **Tristan Bekinschtein.** *MRC Cognition and Brain Sciences Unit – Cambridge UK. "Neuroscience of what makes us laugh"* **Yann Cojan.** *Center for Neuroscience. University of Geneva, Switzerland. "Hypnosis and the brain: investigation of hypnotic suggestions using fMRI"* **Mariano Sigman.** *Departamento de Física, Universidad de Buenos Aires, Argentina. The taste of music"*
12:30	**"Héctor Maldonado" Lecture: John Hildebrand.** *Department of Neuroscience. University of Arizona, USA "Learning from Insect Brains: Explorations of a 'Simple' Olfactory System"*
13.45	**Farewell Barbecue**

COURSE ABSTRACTS

Course
"Electrophysiological Recordings and Optical Imaging in Neuroscience"

Neurons are excitable cells that express themselves electrically. These signals - changes in membrane potential caused by ions moving through channels - take place on a millisecond timescale and over a range of millivolts, involving tiny currents of pico- and nano ampères. The nature of these signals and their time scale impose severe restrictions on the experimental approaches for studying the physiology of neurons. The two most suitable methods are electrophysiology and imaging. However, the technical nature of these disciplines makes them inaccessible to many neurobiologists. This invisible barrier is an obstacle to the development of neuroscience. The goal of the course is to present in a simple way the principles, capabilities and limitations of electrophysiological and imaging techniques. It will provide a theoretical grounding of each technique, and illustrate each using real examples, in many cases produced in the laboratory of the instructor. The course will consist of two modules: the first devoted to the study of the bases of the electrophysiology and the second to imaging techniques.

Tuesday, October 18th
Day 1: Electrophysiology

09:00 - 11:00 | **Lecture I**

"Membrane potential, action potential, excitatory and inhibitory synaptic currents" **Mariano Casado**

This first lecture is intended to level the knowledge on basic neurophysiology among participants of the course.

11:30 - 12:55 | **Lecture II**

"Extracellular and intracellular solutions, basic properties, ions"
David Ogden

A discussion will be provided on the most common solutions used in cellular physiology, the basis for choosing specific ions, and also the underlying cellular principles.

14:00 – 18:00 | **Lecture III**

"Patch clamp techniques: voltage clamp, current clamp, perforated patch"
Mariano Casado & Marco Diana

The instructors will present a comparative analysis of different electrophysiological techniques, its applications and limitations. Typical problems, such as series resistance in patch-clamp recordings (whole cell configuration) will be evaluated. Practical approach. The choice of the appropriate techniques for each experimental condition and according to the parameter that needs to be measured.

18:30 - 20:30 | **Lecture IV**

"Electronics, the construction of a (basic) amplifier and test with model cell" **Boris Barbour & David Ogden**

Practical approach. An introduction will be provided on the electronics used in patch-clamp, voltage-clamp amplifiers with microelectrods, current-clamp, etc. Students will be divided in small groups for discussion of details in their design of basic models of amplifiers. Demonstrations. Tests using oscilloscope and model cell.

20:45 - 21:15 | **Discussion. Round table**

A final session will be held among students and faculty in order to answer remaining questions related to the subjects of this first part of the course.

Wednesday, October 19th
Day 2: Imaging

08:15 – 09:15 | **Breakfast Meeting**

A breakfast will be served to students and professors before the first lecture. This seeks to give students the opportunity to have informal chats with the professors in order to discuss about their interests, future plans, career.

09:15 – 11:15 | **Lecture V**

"Introduction to conventional and confocal microscopy" **Stephanee Dieudonné**

This part is intended to level basic knowledge of students about the basis of optics and the principles and application of conventional and confocal microscopy. Concepts such as light source, laser, lamps, detectors will be discussed. As for the first lecture in the first section, this talk is meant to provide the students with the tools to understand the rest of the section.

11:30 – 13:30 | **Lecture VII**

"Introduction to 2-photon microscopy" **Stephanee Dieudonné**

The basis and applications of two photon microscopy will be explained. A comparison will be drawn with confocal microscopy.

15:00 – 17:00 | **Lecture VII**

"Caged compunds and Ch- Rhodopsins. Principles, compounds" **David Ogden & Marco Diana**

Ca2+ and glutamate uncaging as well as the novel Ch- Rhodopsin/allo-rhodopsin tools will be studied.

17:30 – 18:45 | **Lecture VIII**

"Voltage sensitive dyes: principles – compounds – applications" **David Ogden**

19:10 – 20:30 | **Lecture IX**

"Data analysis, programs" **Boris Barbour**

A free software, WinWCP, for data acquisition will be presented. Examples from previous sections will be used to explain the principles of signal analysis in electrophysiology (i.e., spike sorting – cross correlogram – statistics). An open source -free - software for data analysis developed in Barbour's lab will be introduced.

20:45 – 21:30 | **Round Table**

Final discussion on the contribution, perspectives and challenges of imaging techniques on neurophysiology. Remaining questions will be taken.

SAN2011 MEETING ABSTRACTS

9.30-12.20
Symposium I Non classical synaptic transmission

"Unitary volume transmission by neurogliaform cells: broadening the functional scope of single neurons"

Gabor Tamas - *Research Group for Cortical Microcircuits of the Hungarian Academy of Sciences, Department of Physiology, Anatomy and Neuroscience, University of Szeged, Közép fasor 52., Szeged, H-6726 Hungary*

The presentation addresses mechanisms linking the activity of single neurons with network events by defining the function of a cell type in the cerebral cortex. The key hypothesis is that neurogliaform cells cells achieve their function in the cortex through an extreme form of spatial unspecificity of release. We showed neurogliaform cells reach GABAA and GABAB receptors on target cells through unitary volume transmission going beyond the classical theory which states that single cortical neurons act in or around synaptic junctions. Moreover, enrichment of the neurosteroid sensitive GABAA delta receptor subunits on neurogliaform cells suggests that these neurons could gate hormonal actions in neocortical circuits. We propose that the spatial unspecificity of neurotransmitter action leads to unprecedented functional capabilities for a single neuron simultaneously acting on neuronal, glial and vascular components of the surrounding area allowing neurogliaform cells to synchronize metabolic demand and supply in microcircuits.

"The role of nitric oxide in neuronal homeostasis and intrinsic plasticity"

Ian D. Forsythe - *Dept Cell Physiology & Pharmacology, University of Leicester, Leicester LE1 9HN. UK.*

It is well established that synaptic plasticity is a crucial element of brain function, underlying neuronal development and adaptations associated with learning and memory. It is increasingly recognised that the postsynaptic neuron excitability is also malleable, and so must also be considered when

investigating the physiological changes associated with network activity. The observation that synaptic signalling can influence neuronal excitability through changes in ion channel activity is known as intrinsic plasticity.
Previously we have demonstrated that the auditory brainstem expresses high levels of neuronal nitric oxide synthase (nNOS) and that stimulation of the calyx of Held synapse in the medial nucleus of the trapezoid body (MNTB) triggers nitric oxide signalling to suppress Kv3 potassium channels (Steinert et al., Neuron 60: 642-656, 2008). Now we show that NO additionally enhances Kv2 potassium currents, thereby shifting the basis of action potential repolarization in both the MNTB and CA3 region of the hippocampus (Steinert et al., Neuron 71:291-305 2011). This shows that synaptic activation of nitrergic signalling is a broad mechanism for intrinsic plasticity and suggests that part of the mechanism involves activation and/or suppression of different families of voltage-gated potassium channels. The net effect of this nitrergic modulation is generation of short duration action potentials which can be sustained at higher firing frequencies, thereby tuning the target neurons to the recent history of synaptic activity.

"Metabotropic glutamate receptor signalling in cerebellar Purkinje neurons studied with flash photolysis"

David Ogden, Marco Canepari, Celine Auger - *Laboratoire de Physiologie cérébrale, UMR8118, Université Paris Descartes, Paris, France.*

Flash photolysis to release L-glutamate or receptor specific ligands can be used in combination with pharmacological agents to study postsynaptic signalling independently of interference by drugs with the presynaptic processes. We have used this approach to study two pathways mediated by mGluR1 receptors originating with parallel fibre burst stimulation. One is the slow EPSC, mediated by a calcium permeable cation channel that produces a slow excitation of Purkinje neuron PN dendrites and a slow increase of intracellular calcium ion concentration. It operates independently of phospholipase C activation but dependent on tyrosine kinase/phosphatase. A second type of response is seen only after depolarisation or firing of the

PN. After stimulation or glutamate photorelease the response comprises a precise delay of 100 ms followed by a transient K conductance resulting from a transient Ca increase apparently mainly in spines. This latter pathway is dependent on PLC and is due to Ca release from stores. The two pathways appear reciprocally related in amplitude and are distributed only over about 20% of the dendritic field. The size of the mGluR1 mediated excitation seen with PF stimulation was found to depend on whether AMPA receptors were inhibited. AMPA receptor antagonists enhance signalling via metabotropic receptors, an effect prevented by inhibition of protein tyrosine kinase. This observation suggests that PF synapses with AMPA receptors active participate less in the mGluR1 response. Conversely synapses with strong mGluR1 signalling may have decreased AMPA receptor activity and will respond with mGluR1 signalling to the lower glutamate concentrations seen with spillover.

"Pure spillover transmission in the cerebellum"

Boris Barbour –*Neuroscience Section, IBEns (CNRS UMR 8197 / INSERM U 1024) Ecole Normale Supérieure*

Classic central synaptic transmission by fast neurotransmitters - glutamate, GABA or glycine - involves liberation from vesicles directly opposite postsynaptic receptors at junctions containing both a presynaptic active zone and a postsynaptic specialisation. Such classic transmission is thought to underlie much of the information transfer and processing in the brain. However, there also exist a substantial number of reports of signalling by the same transmitters outside this classic framework, whereby liberation and/or receptor activation occur beyond synaptic boundaries. We term these processes collectively parasynaptic signalling. After a review of the principles governing activation of receptors inside and outside active synapses, a novel, pure-spillover glutamatergic transmission will be presented. This operates in the cerebellar cortex, between climbing fibres and molecular layer interneurones. Possible functions for this parasynaptic signalling will be discussed.

19.30-20 :30
"De Robertis" Lecture

"Accessing the neural bases of cognitive processing in a simple brain (that turns to be not so simple)"

Martín Giurfa - *Centre de Recherches sur la Cognition Animale – CNRS, Université Paul Sabatier – Toulouse, 31062 Toulouse cedex 4, France*

Equipped with a miniature brain smaller than one cubic millimeter and containing only 950000 neurons, honeybees could be indeed considered as having rather limited cognitive abilities. However, bees display a rich and interesting behavioral repertoire, in which learning and memory play a fundamental role in the framework of foraging activities. We focus on the question of whether adaptive behavior in honeybees exceeds simple forms of learning and whether the neural mechanisms of complex learning can be unraveled by studying the honeybee brain. Besides elemental forms of learning, in which bees learn specific and univocal links between events in their environment, bees also master different forms of non-elemental learning, including categorization, contextual learning and rule abstraction, both in the visual and in the olfactory domain. Different protocols allow accessing the neural substrates of some of these learning forms and understanding how complex problem solving can be achieved by a relatively simple neural architecture. We apply different forms of neuronal inactivation to determine the circuits that are necessary and sufficient to mediate different forms of cognitive processing in bees. Calcium imaging techniques allow visualizing how sensory representations evolve during learning and retention. Moreover, structural analyses detect specific, plastic changes in brain areas as a consequence of long-term memory formation. These results underline the enormous richness of experience-dependent behavior in honeybees, its high flexibility, and the fact that it is possible to formalize and characterize in controlled laboratory protocols basic and higher-order cognitive processing using an insect as a model.

14.30-15.30
"Ranwell Caputo" Lecture

"Regulation of membrane expansion at the nerve growth cone: Axonal specification and elongation"

Santiago Quiroga - *Dpto. de Química Biológica y CIQUIBIC, Fac. de Ciencias Químicas, Univ. Nacional de Córdoba-CONICET*

Axonal growth is one of the hallmarks of neuronal polarization, and requires membrane expansion by exocytosis of plasmalemmal precursor vesicles (PPVs) at the growth cone. We have demonstrated that IGF-1 stimulates the exocytosis of PPVs via activation of PI3k. Few details are known about the PPVs targeting mechanisms. Our results show that a cascade critical for the regulation of membrane expansion in neurons includes TC10 and the exocyst complex. We have also examined the role of growth factors in polarization and established that IGF-1 is (one of) the growth (s) factor (s) initiating polarization and that a particularly early event, in neurons that do not yet exhibit an axon, is the segregation of activatable IGF-1 receptors (IGF-1r) to one neurite. Activation of the IGF-1r requires its insertion into the plasmalemma controlled by TC10 activity and the exocyst complex. Since IGF-1 activates TC10 and triggers exocyst assembly it may regulate the insertion of its own receptor. This is a positive-feedback mechanism that could rapidly amplify the membrane expansion response to IGF-1. We propose, therefore, that the process of IGF-1r insertion to neurite membrane/ receptor activation /and further membrane expansion may be (one of) the self-reinforcing mechanism(s) necessary for neuronal polarization.

18.00-20.40
Symposium II Brain Development and Evolution

"Evolution of the Neocortex: Evo-Devo approach"

Pasko Rakic - *Department of Neurobiology and Kavli Institute for Neuroscience at Yale University, New Haven, CT*

The cerebral cortex is the crowning achievement of evolution and the biological substrate of human uniqueness. However, the genetic origin and cellular mechanisms generating the distinct evolutionary advancements are not well understood. In my presentaion I will describes how the applications of methods of molecular genetic and cell biology applied to concepts of modern developmental neurobiology have given us insights into the evolutionary mechanisms involved in building the primate cerebrum that could not be predicted just few years ago. Although the basic principles of brain organization and development in all mammals may be similar, the modifications of developmental events during evolution produce not only quantitative (e.g., the number of neurons, timing and sequence of cellular events) but also qualitative changes (e.g., the elaboration of new neuronal types, addition of specialized cytoarchitectonic areas and new connections). Relatively small genetic differences between mammalian species are predominately expressed in the brain, and usually act early during embryogenesis at the time of the progenitor's exit from the cell cycle. Indeed, some of the modifications of primate cortical development can often be traced to the action of phylogenetically conserved genes that generate a different outcome depending on their evolutionary context. Thus, the molecular and cellular events in developing human, non-human primates and rodent embryos elucidate both similarities as well as differences that may be relevant for understanding development and evolution of cerebral cortex. We now can propose models of how gene expression within the cell progenitors controls neuron number, regulates neuronal migration and their allocation into proper regions, promotes the differentiation into specific phenotypes, establish connections and determines the expression of particular neurotransmitters and receptors in specific synaptic fields. After basic neural network is set, the final pattern of axonal connections is selected through functional validation of some synapses and elimination of the others. The development and evolution of the cerebral cortex is viewed in the context of the radial unit hypothesis, the postulate of an embryonic protomap and the concept of competitive neuronal interactions. The recent data on the differences in gene expression, molecular pathways and novel cellular interactions that have led to these evolutionary advances

provide also insight into the pathogenesis and therapies for human-specific neuropsychiatric disorders.

Recent Reviews on the subject:

Rakic P, Ayoub AE, Breunig J-J, Dominguez MH. 2009 Decision by Division: Making Cortical Maps. Trend in Neuroscience, 32: 291-301

Rakic, P. 2009 Evolution of the neocortex: Perspective from developmental biology. Nature Review Neurosci. 10, 724-735

"Generation of cortical interneuron diversity in the mouse"

Diego Matías Gelman - *Instituto de Neurociencias de Alicante, Spain*

Gamma-aminobutyric acid-containing (GABAergic) interneurons play major roles in the function of the cerebral cortex. Through mostly inhibitory mechanisms, interneurons regulate the activity of pyramidal cells, prevent hyperexcitability, and synchronize the rhythmic output of cortical activity. Our knowledge on interneuron specification has grown incredibly during the last ten years thanks to the invaluable use of transgenic mice lines. In addition, growing evidence suggest that disruption of interneuron function is common to several psychiatric conditions, such as schizophrenia. They show an extraordinary diversity and, thus, understanding cortical interneuron development seems crucial to shed light into their role in cortical processing, both in health and disease. They arise from the subpallial region of the telencephalon, the most rostral part of the brain, mainly from three defined structures: the medial ganglionic eminence, the caudal ganglionic eminence and the preoptic area. Interneuron diversity is accomplished by the combined expression of different transcription factors during development. So, time and space are key features of interneuron fate. The talk will be focused on the molecular mechanisms controlling the development of cortical interneurons in the mouse.

"Building a bigger brain: New views on brain scaling the development and evolution of mammalian brains"

Suzana Herculano-Houzel - *Instituto de Ciências Biomédicas, Universidade Federal do Rio de Janeiro, Rio de Janeiro, Brasil*

Until recently, bigger mammalian brains were considered to consist homogeneously of larger numbers of larger neurons, with a glia/neuron ratio that accompanies increasing brain size, and a relatively larger cerebral cortex that, in evolution, comes to dominate brain volume and, supposedly, function. In that scenario, the human brain is traditionally considered special: larger than expected for a mammal, or even for a primate.

Recent findings from our lab, however, call for a reconsideration of many of these basic tenets of brain scaling and evolution. Applying a novel method (the Isotropic Fractionator; Herculano-Houzel and Lent, 2005) to a number of species, we have found that different neuronal scaling rules apply to the brain of three different mammalian orders (Herculano-Houzel et al., 2006, 2007; Sarko et al., 2009). In contrast, the scaling rules that apply to non-neuronal cells appear conserved across mammals. We also find that the human brain conforms to the primate scaling rules in both size and number of cells (neuronal and non-neuronal; Azevedo et al., 2009); that, despite the increase in relative size of the cerebral cortex in larger brains, numbers of neurons scale concertedly in the cerebellum and cerebral cortex of a number of mammals (including humans); and that major changes in the cellular composition of the brain occur postnatally (Bandeira et al., 2009). The implications of these findings for understanding the developmental mechanisms of generation of diversity in brain evolution will be discussed in this talk.

References

Herculano-Houzel S, Lent R. Isotropic fractionator: a simple, rapid method for the quantification of total cell and neuron numbers in the brain. J Neurosci. 2005 Mar 9;25(10):2518-21.

Herculano-Houzel S, Collins CE, Wong P, Kaas JH. Cellular scaling rules for primate brains. Proc Natl Acad Sci U S A. 2007 Feb 27;104(9):3562-7.

Herculano-Houzel S, Mota B, Lent R. Cellular scaling rules for rodent brains. Proc Natl Acad Sci U S A. 2006 Aug 8;103(32):12138-43. Epub 2006 Jul 31.

Bandeira F, Lent R, Herculano-Houzel S. Changing numbers of neuronal and non-neuronal cells underlie postnatal brain growth in the rat. Proc Natl Acad Sci U S A. 2009 Aug 18;106(33):14108-13.
Sarko DK, Catania KC, Leitch DB, Kaas JH, Herculano-Houzel S. Cellular scaling rules of insectivore brains. Front Neuroanat. 2009;3:8.
Azevedo FA, Carvalho LR, Grinberg LT, Farfel JM, Ferretti RE, Leite RE, Jacob Filho W, Lent R, Herculano-Houzel S. Equal numbers of neuronal and nonneuronal cells make the human brain an isometrically scaled-up primate brain. J Comp Neurol. 2009 Apr 10;513(5):532-41.

"Human brain evolution: searching the genetic basis underlying our unique cognitive capacities"

Lucía Florencia Franchini - *INGEBI-CONICET, Buenos Aires, Argentina*

It has been hypothesized that the evolution of the unique human cognitive capacities is due to the acquisition of new temporal and spatial expression patterns of preexisting genes rather than changes in the protein-coding sequences. Using a combination of bioinformatics and functional studies including the generation of transgenic zebrafish and mice we are investigating differences in gene regulation which may have contributed to the evolution of the human brain.

I will present here our results involving the functional characterization of regulatory elements of two key brain development genes that evolved faster in the human lineage. We have uncovered a novel enhancer of DLL1, a gene involved in the proliferation/differentiation switch of neuronal precursors that positively evolved in primates. We have found that the primate-specific changes are crucial for this regulatory element function during brain development. In addition, we are functionally characterizing the largest cluster of the most rapidly evolving human elements yet identified (termed [HARs]) located within 648 kb of the NPAS3 gene. We tested the ability of NPAS3-HARs to function as developmental enhancers. Our results indicated that the regulation of NPAS3 and DLL1 has been shaped during human evolution, suggesting that changes in its expression pattern could have been crucial for the evolution of the human brain.

9.30-12.30
Symposium III Applied neuroscience: Music, herbs hypnosis and jokes

"Seeing with the Eyes Shut: Neural Basis of Enhanced Imagery following Ayahuasca Ingestion"

Draulio B. de Araujo - *Instituto do Cérebro da UFRN, Universidade Federal do Rio Grande do Norte, Brazil*

The hallucinogenic brew Ayahuasca, a rich source of serotonergic agonists and reuptake inhibitors, has been used for ages by Amazonian populations during religious ceremonies. Among all perceptual changes induced by Ayahuasca, the most remarkable are vivid "seeings". During such seeings, users report potent imagery. Using functional magnetic resonance imaging during a closed-eyes imagery task, we found that Ayahuasca produces a robust increase in the activation of several occipital, temporal and frontal areas. In the primary visual area, the effect was comparable in magnitude to the activation levels of natural image with the eyes open. Importantly, this effect was specifically correlated with the occurrence of individual perceptual changes measured by psychiatric scales. The activity of cortical areas BA30 and BA37, known to be involved with episodic memory and the processing of contextual associations, was also potentiated by Ayahuasca intake during imagery. Finally, we detected a positive modulation by Ayahuasca of BA 10, a frontal area involved with intentional prospective imagination, working memory and the processing of information from internal sources. Therefore, our results indicate that Ayahuasca seeings stem from the activation of an extensive network generally involved with vision, memory, and intention. By boosting the intensity of recalled images to the same level of natural image, Ayahuasca lends a status of reality to inner experiences. It is therefore understandable why Ayahuasca was culturally selected over many centuries by rain forest shamans to facilitate mystical revelations of visual nature.

"Neuroscience of what makes us laugh"

Bekinschtein, Tristan - *MRC Cognition and Brain Sciences Unit – Cambridge UK*

Why is something funny? There are several conceptual frameworks attempting to explain humour, and a handful of them are scientifically testable. We concentrate here in accounts of verbal humour and explore the neural implementation of wordgames in jokes. Complementarily, we fit the brain responses to a prediction error model where the participants hear the set up line and have to decide whether it will be a joke or not. We expect to be able to map behaviourally and neurophysiologically why we laugh with abstract stimuli (language), while having a good time.

Reference:

Bekinschtein TA, Davis MH, Rodd JM, Owen AM. Why Clowns Taste Funny: The Relationship between Humor and Semantic Ambiguity. J Neurosci. 2011 Jun 29;31(26):9665-71.

"Hypnosis and the brain: investigation of hypnotic suggestions using fMRI"

Yann Cojan - *LABNIC, University of Geneva, Switzerland*

The recent development of neuroimaging techniques has made possible the investigation of brain regions that underlie the hypnotic state. We will present a brief history of hypnosis and the neuroscientific theories proposed to explain the effect of hypnosis on brain functions, then we will discuss the latest results obtained from these techniques, with particular emphasis on studies we conducted on hypnotic paralysis using fMRI.

"The taste of music"

Mariano Sigman - *Departamento de Física, Universidad de Buenos Aires*

Zarlino, one of the most important music theorists of the XVI century, described the minor consonances as `sweet' (dolci) and `soft' (soavi). Hector Berlioz, in his Treatise on Modern Instrumentation and Orchestra speaks about the `small acid-sweet voice' of the oboe. In line with this tradition,

we discovered reliable empirical associations between taste words and musical compositions. Trained musicians asked to improvise on the basis of the four canonical taste words (sweet, sour, bitter, and salty) produce reliable and consistent musical patterns. For instance, bitter improvisations are low-pitched and sour improvisations are high-pitched and dissonant. Blind decoding methods and classification by non-trained musicians can classify binary choices of improvisations at performance level well above chance. Extending the synergic dialog between music and science, we show composition procedures that can generate music from taste-words algorithmically. These results extend the correspondences between language and music, two ubiquitous manifestations of human culture, beyond the well-studied domain of timing, prosody and syntax, into the domains of semantics. We will discuss how these ideas can be extended beyond this narrow domain of semantics to investigate the meaning of music.

12.30-13.30
"Hector Maldonado" Lecture

"Learning from Insect Brains: Explorations of a 'Simple' Olfactory System"
John Hildebrand - *Department of Neuroscience. University of Arizona, USA characteristic behavioral responses.*

The insects are remarkably speciose, diverse, and successful animals from which we can learn much about the evolution, specialization, operation, and adaptedness of neural systems and behavior. Explorations of the diminutive brains of insects reveal principles and mechanisms of neural development and function and at the same time help us understand both beneficial and harmful insect behaviors. Key to that understanding is the insect's sense of smell. Evolutionarily ancient and impresisvely powerful, insect olfaction is of paramount importance for much of what most insects do. This presentation will highlight investigations of behaviors based on complex olfactory stimuli and the underlying neural processing of those stimuli in an experimentally favorable system, the giant sphinx moth Manduca sexta. This work aims at understanding: the neurobiological mechanisms through which information

about olfactory stimuli is encoded, processed, and integrated with inputs of other modalities in the brain; how the innate or learned behavioral significance of a natural, multicomponent olfactory stimulus is encoded in the brain; and how this sensory information ultimately initiates and controls characteristic behavioral responses.

YOUNG INVESTIGATORS ABSTRACTS

14.30-16.20
Young Investigators Colloquium I

"Novel lessons improve memory in elementary school children: evidence of behavioral tagging in humans"
F. Ballarini, C. Martinez, D. Moncada, H. Viola - I*nstituto de Biología Celular y Neurociencias, Facultad de Medicina, UBA*

Education is the most traditional means with formative effect on the human mind, being learning and memory its fundamental support. We have previously showed that in rats receiving weak training protocols which only induce short-term memory, a long-term memory (LTM) was promoted if the training session took place close to a novel experience. This process called behavioral tagging begins with the setting of a learning tag established by the weak training and requires synthesis of plasticity-related proteins induced by novelty. The main goal was to study whether performance on cognitive tests could be improved by a novel experience occurring before or after the acquisition session. A short story was read to a total of 800 elementary school students (between 7 and 10 years) and 24h later we evaluated how much they remembered about it. Memory improvements were observed in groups of students who received a novel lesson (using two different kinds) 1h before or after the reading of the story, but not 4h. Interestingly, if the lesson was familiar, the improvement was not observed. Because LTM could be improved only by novelty and in a critical time window, our results suggest the existence of a behavioral tagging mechanism operating also in humans.

"Ghrelin mediates stress-induced food reward behavior in mice"
Mario Perello - *Instituto Multidisciplinario de Biología Celular (IMBICE) (CONICET/CICPBA)*

Most humans experience altered feeding behaviors upon stress, with approximately 40% eating more than usual. Such changed eating behavior

likely contributes to the increased prevalence of obesity in humans with chronic stress. However, the molecular substrates and neurocircuits involved in the complex behaviors responsible for stress-based eating remain mostly unknown, and few animal models to probe mechanisms orchestrating these behaviors have been reported. Here, we describe a new mouse model in which food reward behavior, as assessed using a conditioned place preference (CPP) task, is monitored in animals following exposure to chronic social defeat stress (CSDS), a model of prolonged psychosocial stress. We demonstrate a) chronic stress in mice increases both CPP for and intake of high fat diet, b) such stress-induced food reward behavior is dependent on signaling by the hormone ghrelin, and c) ghrelin signaling specifically in catecholaminergic neurons not only mediates ghrelin's orexigenic and food reward behavioral effects, but also is sufficient to mediate stress-induced food reward behavior. This mouse model thus has allowed us to ascribe a role for ghrelin-engaged catecholaminergic neurons in stress-induced food reward behaviors.

Ref: J Clin Invest 2011. 121(7):2684-92.

"Novel animal models to study the role of TDP-43 in neurodegenerative disease"

Lionel Müller Igaz - *Grupo de Neurociencia de Sistemas, Dpto. Fisiologia, Facultad de Medicina, UBA*

TAR DNA-binding protein 43 (TDP-43) has been recently identified as the major disease protein in frontotemporal dementia (FTD) and amyotrophic lateral sclerosis (ALS). Transgenic (Tg) mice conditionally expressing human wild type TDP-43 (hTDP-43-WT) and hTDP-43 with a defective nuclear localization signal (hTDP-43-ΔNLS) directed by the CaMKII□ promoter were generated to elucidate mechanisms of neurodegeneration in TDP-43 proteinopathies. Expression of hTDP-43 WT or hTDP-43-ΔNLS led to neuron loss in selectively vulnerable forebrain regions, corticospinal tract degeneration and motor spasticity recapitulating key aspects of FTD and primary lateral sclerosis. The presence of phosphorylated and ubiquitinated

TDP-43 inclusions was variable among various lines. Remarkably, neurodegeneration correlated not with the presence of inclusions but with a dramatic downregulation of endogenous mouse TDP-43 of affected neurons. Finally, mice expressing hTDP-43-ΔNLS exhibited profound changes in gene expression particularly in the up-regulation of genes involved in chromatin assembly. Our data suggest that perturbation of highly regulated endogenous nuclear TDP-43 results in loss of functions and changes in downstream gene regulatory pathways that trigger degeneration of selectively vulnerable neurons.

"Mice lacking dopamine D2 autoreceptors reveal their fundamental role on dopamine neurotransmission"

E.P. Bello, Y. Mateo, D.M. Gelman, D. Noaín, J.H. Shin, V.A. Alvarez, D.M. Lovinger & M. Rubinstein - *INGEBI-CONICET, Buenos Aires, Argentina*

Dopamine (DA) D2 receptors expressed in DA neurons (D2 autoreceptors) exert a negative feedback regulation that reduces DA neuron firing, DA synthesis and DA release. Since D2 receptors are mostly expressed in postsynaptic neurons, pharmacological and genetic approaches have been unable to definitively address the in vivo contribution of D2 autoreceptors to DA-mediated behaviors. To circumvent this difficulty, we generated mice deficient in D2 receptors in midbrain DA neurons (Drd2loxP/loxP; Dat+/IRES-cre, referred to as autoDrd2KO mice), as we could confirm by an in situ hybridization analysis. AutoDrd2KO mice showed elevated DA synthesis and release and displayed hyperlocomotion and supersensitivity to the psychomotor effects of cocaine whereas showed normal motor reactions when challenged on a rotarod or in approach/avoidance conflict tests. Interestingly, these mice also exhibited increased place preference for cocaine and enhanced motivation for food reward. Our results highlight the importance of D2 autoreceptors in the regulation of DA neurotransmission and demonstrate that D2 autoreceptors are important for normal motor function, food-seeking behavior, and sensitivity to the locomotor and rewarding properties of cocaine.

8.30-10.20
Young Investigators Colloquium II

"Membrane perforation induced by Aß is modulated by membrane levels of NR2B and LRP6 protein"

F.J. Sepulveda[1], RW Peoples[2], C.. Opazo[1], LG Aguayo[1] - [1]*Department of Physiology, University of Concepcion, Chile.* [2]*Department of Biomedical Sciences, Marquette University, USA*

Alzheimer's disease (AD) is a progressive neurodegenerative brain disorder that leads to major debilitating cognitive deficits. We have recently postulated that the initial step in synaptic failure induced by Aß is the consequence of membrane pore-formation, however the mechanism is still not been completly elucidated and there is still an active discussion about the mechanisms involved in the formation of Aß perforations. In this study, we found that the perforating action of Aß was modulated by membrane proteins that are believed to be involved in AD. For example, proteolytic treatment on hippocamapl neurons with trypsin significantly decreased the membrane levels of the NR2B subunit of NMDA receptors and LRP6, a co-receptor for Wnt signaling. Interestingly, under the same experimental conditions, the perforations induced by Aß occurred much faster than under control conditions. Moreover, the decrease of levels of NR2B and LRP6 in the membrane produced changes in Aß clustering in the membrane of hippocampal neurons suggesting that these protein can modulate the distribution of Aß in the neuronal membrane. In conclusion, the present data indicate that membrane proteins, such as NR2B and LRP6, can modulate the formation of Aß perforations by sequestering Aß at the neuronal surface.

"Abnormal endo-lysosomal membrane dynamics and retrograde axonal transport defects induced by proteasome inhibition"

Gabriela M. Otero, Lucas E. Cromberg & **Tomás L. Falzone** - *Instituto de Biologia Celular y Neurociencia IBCN-CONICET-UBA*

Extreme polarized neurons depend on a regulated system of vesicle and

protein delivery. Anterograde and retrograde axonal transport mediated by kinesin and dynein motors ensures the correct distribution of neuronal cargos supporting polarization. Abnormal protein accumulation together with ubiquitin proteasome degradation defects have been suggested as mechanism involved in the progression of Alzheimer disease (AD). To test the hypothesis that delivery defects of the protein degradative machinery may lead to the accumulation of toxic proteins in AD, we assessed for the movement of the proteasome. Live imaging experiments tracking fluorescent proteasome subunit revealed processive anterograde and retrograde trajectories compatible with motor dependent fast axonal transport. Interestingly, proteasome inhibition (MG132) induced selective changes in retrograde transport. Double color tracking proteasomes and lisosomes revealed a coordinated retrograde transport that is reduced by MG132. Moreover, MG132 induced kinesin-1 motor accumulation at growth cones and kinesin-1 reduction by siRNA increases lisosomal transport. Taken together, our results suggest a mechanism of crosstalk between proteasome and lisosome degradation that may be mediated by local accumulation of kinesin-1 motor.

"Alternative splicing of calcium channels adjust neuronal activity"

Jesica Raingo - *Instituto Multidisciplinario de Biología Celular (IMBICE) (CONICET/CICPBA)*

Voltage operated calcium channels (CaV) translate electric signals into chemical signals in neurons. CaV modulate gene expression, neurotransmitter release and/or signaling cascades depending on their type and localization. Thus, the fine tuning of CaV activity became essential for neuronal functions. The G protein coupled receptor (GPCR) pathways efficiently control CaV activity. These GPCR pathways are extremely specific for each neuronal function and differ among neuron types. What makes these processes so precise is a hard-to-answer question. The GPCR downstream players unlikely give the high specificity as they are widely expressed among neuron. On the other hand, the CaV undergo an extensive pre-mRNA alternative splicing generating many different isoforms of the channel, some of which are

specific for neuron types. We have showed that different CaV alternative splicing isoforms have different sensitivity to GPCR pathways. Here, I will discuss the result of CaV alternative splicing on sensitive and sympathetic neurons and also the behavioral outcome observed in genetically modified mice that express only a particular alternative splicing isoform.

"Retrograde BMP signaling modulates the pace of the circadian clock"

Esteban J. Beckwith - *Laboratorio de Genética del Comportamiento, Fundación Instituto Leloir, Buenos Aires, Argentina.*

Living organisms use circadian rhythms to maintain internal temporal order and anticipate daily environmental changes. Clocks employ self-sustained biochemical oscillators and become evident at molecular, physiological and behavioral levels. In Drosophila, a group of neurons expressing the Pigment Dispersing Factor represent the central oscillator of the fly brain. As a result of a miss-expression screen we identified a fly strain that causes period lengthening of daily activity rhythms. The transposon landed within schnurri (shn), a nuclear component of the decapentaplegic/bone morphogenetic protein pathway. shn overexpression in the PDF circuit was necessary and sufficient to generate a 25.5h period of locomotor behavior specifically in adult flies. Interestingly, overexpression of constitutively active BMP receptors also gave rise to long period phenotypes. In contrast, down-regulation of the endogenous receptors led to arrhythmicity. The period phenotype of shn overexpression was rescued by per, tim and sgg overexpression, three key players in the molecular clock. A detailed analysis of PER subcelular localization showed a delayed PER nuclear entry in the mutant compared to that of wild type flies, pointing to a specific effect of shn deregulation on the core clock mechanism.

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P-172 - Florencia María Dadam - Participation of sex chromosome complement in brain pattern activation during acute sodium depletion
P-175 - María Belén Pardi - A switch from integrative to orthogonal input processing by hippocampal adult-born neurons
P-178 - Silvio Gabriel Temprana - Spatio-temporal pattern of efferent connectivity of adult-born dentate granule cells

SESSION 2 - FRIDAY 21ST - 10:30 hs 13:00 hs

P-170 - Bárbara Yael Braz - Role of long term depression in corticostriatal postnatal maturation in vivo
P-173 - Mariela Escande - Changes in pallidal activity during cortical activation depend on striatal output in an animal model of Parkinson's disease
P-176 - María Soledad Pitra - Serotonergic system involvement during states of body hypertonicity
P-179 - Margarita Yang - GABAergic regulation of the cardioinhibitory response in the crab Neohelice granulata

SESSION 3 - FRIDAY 21ST- 15:30 hs 18:00 hs

P-168 - Rodrigo Javier Alvarez Juliá - Research project: assessing the physiological impact of NMDAR ablation in interneurons on prefrontal cortex activity during postnatal development
P-171 - María Ana Calviño - Strategies toward identifying the source of endogenous 5-HT involved in the SSRI effect
P-174 - Camilo Juan Mininni - Stable Neural Ensemble Recordings in Awake Head-fixed

Rat
P-177 - Elisa Schneider - Neural basis of goal directed behaviour

Neurochemistry and Neuropharmacology

SESSION 1 - THURSDAY 20TH - 16:30 hs 19:00 hs

P-181 - Ignacio Bergé - Pharmacology of nicotinic receptors from C. elegans muscle
P-184 - Pablo Ariel - Casalis Neuroprotective effect of a new rhEPO analogue with low erythropoietic activity
P-187 - María Alejandra Esparza - Latrunculin A in the nac inhibited the expression of motor sensitization and increased AMPA receptors after cocaine in chronically stressed animals
P-190 - Damián Gustavo Maur - Role of nitric oxide in the alterations in HPA axis induced in prenatally stressed offspring
P-193 - Juan Ignacio Muñoz - BDNF and NR2B/PSD-95 complex modulation by Atorvastatin leads to an improvement in the spatial learning and memory after transient focal cerebral ischemia
P-196 - Mariana Raineri - Neuroprotective properties of modafinil on methamphetamine-induced glial activation in mouse striatum

SESSION 2 - FRIDAY 21ST - 10:30 hs 13:00 hs

P-182 - Juan Carlos Boffi - Stereospecific modulation of α9α10 nicotinic cholinergic receptors by L-ascorbic acid
P-185 - Natalí Luján Chanaday Ricagni - Nerve terminal alterations in frontal cortex from rats with experimental autoinmune encephalomyelitis (EAE)
P-188 - Nicolás Fernández Hurst - Involvement of the Gabaergic system in the induction of the neuropathological alterations in experimental autoinmune encephalomyelitis (EAE)
P-191 - Daniel Minter - In search of a specific agonist for the α9α10 nicotinic cholinergic receptor
P-194 - Claudia Pascovich - Melanin concentrating hormone (MCH) decreases presumed serotonergic neuronal activity in the dorsal and median raphe nuclei
P-197 - María Inés Riberi - Differential expression of α1 subunit and β2-3 subunit of GABA-A receptors in neonatal chick forebrain submitted to an acute stess. Modulation by noradrenaline hicks forebrain

SESSION 3 - FRIDAY 21ST- 15:30 hs 18:00 hs

P-180 - Andrea Beltrán González - Potentiation of the homomeric rho1 GABAc receptor function by H2O2 is mediated by the intracellular cysteine residue Cys-364
P-183- Lucila Guadalupe Cáceres - Neonatal X radiation: hippocampal morphology and anxiety state
P-186 - Santiago Cuesta - Effect of vanadium exposure through lactation: Biochemical

and histological studies in neonate wistar rats brain
P-189 - Javier Gasulla - Homomeric ρ1 GABAc receptor function is potentiated by S-nitrosylation
P-192 - Bethania Mongi Bragato - Preproenkephalin knockout mice did not show sensitization to the behavioral effects induced by cocaine and failed to show the cocaine-induced increases in ERK activation and AMPA cell surface expression in striatum and nucleus accumbens
P-195 -María Constanza Paz - The brain RAS is involved in the neuroadaptative responses induced by amphetamine in a two-injection protocol
P-198 - Maximiliano Rivera - Effects of α-and β-thujone on anxiety behavior in neonatal chicks: involvement of GABAA receptors

Neuroendocrinology and Neuroimmunology

SESSION 1 - THURSDAY 20TH - 16:30 hs 19:00 hs

P-199 - Tathiana Alvarenga - The effect of sleep loss on the reproductive function of male rats
P-202 - Mario Bibolini - Inhibitory role of Diazepam on autoinmune inflammation in rats with experimental autoinmune encephalomyelitis
P-205 - Nadia Kazlauskas - Effect of maternal viral infection on postnatal development in a mouse model of autism
P-208 - Verónica Trujillo - Model of depression in rats: Effect of tianeptine on hipocampal GR and MR expression, MR/GR balance, anxiety and hedonic behavior

SESSION 2 - FRIDAY 21ST - 10:30 hs 13:00 hs

P-200 - Gustavo Adolfo Argañaraz - Involvement of kinin B2 receptors in the development of the pilocarpine-induced epilepsy in female rats
P-203 - Agustina Soledad Cabral - Ghrelin indirectly activates hypophysiotropic CRF neurons
P-206 - Javier María Peralta Ramos - Cell death mediated apoptosis by a Candida albicans infection in Central Nervous System (CNS)
P-209 - Lesly Spring Valdivia Torres - Cold exposure activates Thyrotropin Releasing Hormone (TRH)-producing neurons in specific brain nuclei

SESSION 3 - FRIDAY 21ST- 15:30 hs 18:00 hs

P-201 - Natalia Baez - Immunologic studies in a model of Autism Spectrum Disorders (ASDs)
P-204 - Evelin Mariel Cotella - Early maternal separation and stress during adulthood: implications for glucocorticoid receptors and anxiety-like behavior under treatment with tricyclic antidepressant amitriptyline
P-207 - Ezequiel Martin Salido - Early retinal changes in an experimental model of type 2 diabetes characterized by postprandial hyperglycemia

Sensory Systems

SESSION 1 - THURSDAY 20TH - 16:30 hs 19:00 hs

P-211 - Mercedes Bengochea - Morphological and physiological characterization of columnar neurons from the second optic ganglion of the crab Chasmagnathus granulatus
P-214 - Esteban Calcagno - The role of visual cues in auditory distance perception
P-217 - Diego Carlos Fernández - Distal axonopathy of the visual pathway in experimental diabetes
P-220 - Emiliano Marachlian - Learning modifies odor mixtures representation in the honey bee antennal lobe
P-223 - Soledad Lucía Uran - Moderate noise exposure differentially affects hippocampal and auditory structures in developing rats

SESSION 2 - FRIDAY 21ST - 10:30 hs 13:00 hs

P-212 - Melina Bordone - Study of visual pathways in an experimental model of glaucoma in rats
P-215 - Damián Dorfman - The exposure to an enriched environment prevents acute visual ischemic injury in adult rats
P-218 - María Eugenia Gómez Casati - More than just supporting: new insights into the roles of supporting cells in the postnatal and adult inner ear
P-221 - Jorge Ariel Rasmussen - Polarity in sensory neuron axonal transport: relevance to regenerative capacity in the central and peripheral nervous system

SESSION 3 - FRIDAY 21ST- 15:30 hs 18:00 hs

P-210 - Nicolás Adalberto Belforte - Neuroprotective effects of brief ischemia pulses in an experimental model glaucoma
P-213 - Carlos David Bruque - Neurogenesis in the zebrafish retina: glutamatergic control of cell proliferation
P-216 - Pablo Esteban Etchemendy - Perception of principal pitch in asymmetrical vibratos: a new experiment and revision of models
P-219 - Lucas David Jungblut - Behavioral response to conspecific alarm cues in tadpoles of Rhinella arenarum: the involvement of the olfactory and vomeronasal organs
P-222 - Manuel Soliño - Protein nitration: Is it part of the oxidative stress associated with light induced retinal damage?

Sensory Systems

SESSION 1 - THURSDAY 20TH - 16:30 hs 19:00 hs

P-229 - Walter Bast - A semiconductor-based photoconductive stimulation device

P232 - Marisa Ghersi - Ghrelin increases glutamate release from rats hippocampal synaptosomes
P235 - Juan Goutman - Facilitation and depression determine timing of synaptic responses at the inner hair cell ribbon synapse
P-238 - María Victoria Oberholzer - M1 muscarinic receptor positively modulates neurotransmission in CA1 area of rat hippocampus
P241 - Adriana Saal - Decisions on magnitude and parity in two consecutive tasks
P-244 - Juan Pablo Vivar - A methodological approach to monitor fluctuations on vesicular membrane potential
P-247 - Javier Zorrilla de San Martin - Functional development of the medial olivocochlear efferent innervation before the onset of hearing

SESSION 2 - FRIDAY 21ST - 10:30 hs 13:00 hs

P-224 - Ezequiela Adrover - Prenatal stress effect on glutamate transporters, GLT1 and GLAST
P-227 - Jimena Ballestero - Short -term synaptic plasticity at the medial olivocochlear hair cell synapse in α9L9iT knock-in mice
P-230- María Georgina Davies Sala -Local network activity controls neuronal maturation in the adult dentate gyrus
P-233 - Belén Goitia - Effect of repetitive cocaine "binge" administration on GAD65/67 and T-type subunits levels from GABAergic somatosensory thalamic neurons in mice
P-236 - Guillermina Hernando - GABA receptors in Caenorhabditis elegans embryonic muscle cells
P-239 - Estefanía Piegari - The effect of calcium dyes on the observed dynamics of calcium signals
P-242 - María Fernanda Tolosa - Functional changes of Cys-loop receptors generated by electromagnetic fields
P-245 - Carolina Wedemeyer - GABA regulates the release of acetylcholine (ACh) at the medial olivocochlear (MOC) efferent-inner hair cell synapse through presynaptic GABAB(1a,2) receptors

SESSION 3 - FRIDAY 21ST- 15:30 hs 18:00 hs

P-225 - Yanina D.Alvarez - The immediately releasable pool of mouse chromaffin cell vesicles is coupled to P/Q calcium channels by the synaptic protein interaction site
P-228 - Walter Bast - Design strategies for improving dynamic clamp performance
P-231 - Mariano Nicolás Di Guilmi - Reduction of presynaptic calcium influx with higher amplitude excitatory post-synaptic currents in S218L Cav 2.1 knock-in migraine mouse model
P-234 - Nicolás González- Muscarinic and adrenergic neurotransmission at CA1 area of rat hippocampus
P-237 - Francisco López Aguilera - Angiotensin II AT2 receptor blocker reverses the neuroprotective action of preconditioning in the brain of hypoxic-ischemic neonatal rats
P-240 - Marilina Raíces - 5-Bromo-2`deoxyuridine is toxic to olfactory epithelium cells

at concentrations widely used for studies of cells lineage

P-243 - Cecilia Tubert - Functional maturation of striatal cholinergic interneurons during adolescence

P-246 - Sung Min Yang - Voltage-dependent conductances modulate the amplification and propagation of signals in nospiking neurons

• CELLULAR AND MOLECULAR NEUROBIOLOGY

Poster Number 1 | Session 1

"Modulation of CaV1.3 activity by melanocortin receptor type 4 (MC4R)"

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MC4R mutations are, by far, the most common cause of monogenic obesity in humans. It has been shown that MC4R is highly expressed in the hypothalamic paraventricular nucleus (PVN), where it increases expression of appetite related-genes. The mechanisms by which MC4R regulates gene expression are unclear. Calcium influx through L-type calcium channels induces gene expression in neurons. The PVN neurons express the L channel isoform, CaV1.3, which is activated by negative potentials and has a narrow distribution in the brain. Here, we tested if MC4R regulates CaV1.3 activity using the patch clamp technique in HEK293 cells. We developed an in vitro system co-expressing CaV1.3, the auxiliary subunits and a MC4R plasmid with the soluble GFP sequence. We found that MC4R co-expression did not modify the CaV1.3 biophysical properties, such as voltage activation and kinetic. However, we observed an inverse correlation between the amount of transfected MC4R cDNA and CaV1.3 current levels, suggesting an effect of MC4R basal activity on CaV1.3 function. In line with this possibility, we found that mu-opioid receptor, which has no basal activity, did not affect CaV1.3 current levels. Our results suggest that CaV1.3 could be a target of MC4R to regulate gene expression in PVN neurons.

Cellular and Molecular Neurobiology
Poster Number 2 | Session 2

"Impaired mitochondrial dynamics and cell death in manganese-induced Parkinsonism"

Agustina Alaimo[4], Roxana Mayra Gorojod[4], Nicola Simola[2], Juan Beauquis[1,3], Flavia Saravia[1,3], **Micaela Morelli[2], Mónica Lidia Kotler[4]**
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Mitochondria form a dynamic network by opposing actions of fusion/fission proteins. Changes in their expression or localization alter mitochondrial morphology and may promote apoptosis. Increasing evidence correlates these events with the occurrence of neurodegenerative diseases. Therefore, we focused on this topic in Manganese (Mn)-induced Parkinsonism, a disorder associated with Mn accumulation preferentially in the basal ganglia. Using MitoTracker staining we observed increased mitochondrial network fission in Mn-treated rat astrocytoma C6 cells. Moreover, Mn provoked a marked decrease on the fusion protein Opa-1 levels as well as a dramatically increase in fission protein Drp-1 levels in both cytosol and mitochondria. Mdivi-1, a newly pharmacological Drp-1 inhibitor, prevented cell death, reduced apoptotic nuclei and maintained mitochondrial network integrity. In addition, we analyzed the histological changes in rat brain sections after Mn-intrastriatal/nigral administration. Our results revealed that altered mitochondrial dynamics plays a role in Mn-induced cell death. This knowledge may provide new therapeutic tools for the treatment of Manganism and possibly other neurodegenerative diseases.

Cellular and Molecular Neurobiology
Poster Number 3 | Session 3

"Notch Pathway involvement in the remyelination and in the romyelinating effect of Apotransferrin (aTf)"

Florencia Almeira Gubiani[1], Alejandro Schinder[2], Ana María Adamo[1]
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A promyelinating effect of aTf in a demyelination model with cuprizone (CPZ) has been demonstrated. We examined the Notch signaling pathway in the remyelination process and in the aTf-mediated reversion of demyelination. Twenty-one-day-old Wistar rats were fed with a 0.6% CPZ diet for 2 weeks. After withdrawal of CPZ, animals were injected with either aTf or saline (C). The subventricular zone (SVZ) and corpus callosum (CC) of rats were used for Western blot and RT-PCR. We evaluated the expression of down stream genes Hes1, Hes5 and MAG to explore Notch pathway activation and its relationship with OPC proliferation and differentiation. Results showed an increase in Hes1 expression in the SVZ of CPZ animals when compared to C and no changes in Hes5 or MAG expression. aTf injection increased Hes1 and MAG expression after 6h and 24h respectively. In the CC, we found decreased expression of MAG that was reverted by injection of aTf. In order to study the progenitors` migration to the demyelinated CC, CAG-GFP

retrovirus was injected into SVZ concomitantly with saline, aTf or a γ-secretase inhibitor. Immunostaining at 7 days post injection showed higher incorporation of CAG-GFP virus into SVZ of CPZ- animals when compared with C, as well as colocalization with NG2 and NESTIN.

Cellular and Molecular Neurobiology
Poster Number 4 | Session 1

"Sprouty4 is a negative modulator of TrkA signaling and neuronal differentiation induced by NGF"

Fernando Cruz Alsina[1], Dolores Irala[1], Paula Aldana Fontanet[1], Francisco Javier Hita[1], Fernanda Ledda[1], Gustavo Paratcha[1]

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The Sprouty (Spry) family of proteins represents endogenous regulators of downstream signaling pathways induced by receptor tyrosine kinases (RTKs). Despite of the essential contribution of nerve growth factor (NGF) for neuronal development and function, the molecular mechanisms that control NGF-induced TrkA signaling are not totally understood. Using a cDNA microarray screening, we identify Spry4 as a potential modulator of intracellular signaling pathways and biological processes induced by NGF and its receptor TrkA. qRT-PCR assays confirme that Spry4, but not Spry1-2, is significantly induced by NGF in PC12 and primary dorsal root ganglia (DRG) neurons. Ectopic expression of wt Spry4 causes a significant reduction in MAPK and Rac1 activation and neurite outgrowth induced by NGF. Ectopic expression of a mutated form of Spry4 (Y53A), in which a conserved tyrosine residue was replaced, fail to block both TrkA-mediated Erk/MAPK activation and neurite outgrowth induced by NGF, suggesting that an intact Tyr 53 site is required for the inhibitory effect of Spry4 on NGF signaling. Together, these findings establish a new physiological mechanism through which Spry4 regulates neurite outgrowth reducing not only the MAPK pathway but also restricting Rac1 activation in response to NGF.

Cellular and Molecular Neurobiology
Poster Number 5 | **Session 2**

"GIT1 mediates filopodium formation induced by stress-regulated neuronal glycoprotein M6a"

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Gpm6a was identified as a stress-responsive gene down-regulated in the hippocampus of chronically stressed animals. This effect is reversed by antidepressants. The role of M6a in neurite and filopodium formation has been demonstrated recently. Neuroplasticity is impaired by chronic stress exposure. Mechanisms by which M6a mediates its neuroplastic effect and chronic stress response remain unclear. In the present study we analyzed a possible signaling pathway by which M6a regulates neurite and filopodium formation. Coimmunoprecipitation followed by mass spectrometry revealed G protein-coupled receptor kinase-interacting protein 1 (GIT1) as a potential M6a interacting partner. GIT1 regulates spine morphogenesis and synapse formation by targeting actin regulators and locally modulating Rac activity. The effect of the coexpression of wt and mutant GIT1-GFP with wt M6a-RFP was analyzed in neuroblastoma cells using fluorescent microscopy. Neurite outgrowth and filopodium formation was quantified. The coexpression of mutant GIT1 with M6a caused a significant decrease in M6a-induced filopodium formation. No effect was observed when mutant forms of Rac1 were employed. We suggest that GIT1 mediates the function of M6a in filopodium formation through a pathway that does not involve Rac1.

Cellular and Molecular Neurobiology
Poster Number 6 | **Session 3**

"RAGE and NFkB are involved in neuronal death and reactive gliosis induced by Sleep Apnea"

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Sleep apnea (SA) produces cognitive alterations. In an experimental model of

SA by intermittent hypoxia (IH) we previously showed reactive gliosis, neuronal degeneration, overexpression of the Receptor for Advanced Glycation End Products (RAGE) and its ligand S100B. Since S100B/RAGE/NFkB pathway can promote either cell survival or death; we studied NFkB activity using reporter mice and performed loss of function assays in vivo and in vitro in neuro-glial culture. NFkB activity was increased after IH. Intrahippocampal injections of RAGE neutralizing antibodies decreased neuronal alterations and reactive gliosis in IH animals but were detrimental in normoxia. Endogenous S100B blockage did not reverse neurodegeneration but reduced reactive gliosis after IH. NFkB blockage with SFZ (NFkB inhibitor) improved neuronal survival after IH but was detrimental in normoxic conditions. Since oxidative stress is recognized as the main detrimental effect of SA, primary neuro-glial mixed cultures (astrocytes and neurons) were exposed to 150 uM H2O2 for 30 min and showed changes in astroglial morphology that were reversed by RAGE blockage. Taken together, these results indicate that over-activity of the S100B/RAGE/NFkB pathway could promote reactive gliosis and neuronal degeneration after IH.

Cellular and Molecular Neurobiology
Poster Number 7 | **Session 1**

"Effects of environmental enrichment on hippocampal astroglial populations in APP transgenic mice"

Juan Beauquis[2,3], Patricio Pavía[2], Paulina Roig[3], Verónica Galván[4], Alejandro F De Nicola[3,1], Flavia Saravia[2,3,1]

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Environmental stimulation plays a protecting role in the pathogenesis of Alzheimer¥s disease (AD). Our aim was to explore the effect of environmental enrichment (EE) in a model of AD, focusing on astroglial changes in the hippocampus. Transgenic mice (Tg, PDAPP-J20; Swe and Ind APP mutations) and non-transgenic siblings (NTg) were housed in EE or in standard conditions (SC) for 3 months (5 to 8 months of life). Using immunohistochemistry we found a decreased density of GFAP+ astrocytes in the CA1 field in TgSC and an increase in TgEE. The volume of astrocytes was measured using confocal microscopy in 2 subpopulations: amyloid plaque-associated (PA) and non-plaque-associated (NPA) astrocytes. PA astrocytes were larger than control astrocytes, suggesting an increased reactivity, with no effect of EE. NPA astrocytes of Tg had a decreased volume when compared

with those from NTg, possibly indicating astroglial atrophy. Interestingly, EE was able to increase the volume to control levels. Sholl analysis showed that astrocyte ramification was increased in TgSC (NPA and PA) but not in TgEE compared with controls. Our results indicate that EE influences astrocyte morphology in APP Tg mice, suggesting a role in the evolution of AD, modulating not only neuronal but also glial populations.

Cellular and Molecular Neurobiology
Poster Number 8 | Session 2

"Effects of monochromatic light in the expression patterns of the transcription factors LIM 1+2 and Islet-1 in horizontal neurons"

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Horizontal cells provide the pathways for local and long range interactions between photoreceptors. They are related to lateral inhibition, contrast and color processing. It is known that light modifies the synaptic connection pattern, however, little or nothing is known about how each subtype of horizontal neurons (H1-H4) respond to a particular wavelength. To analyze the effect of spectral deprivation, chickens were reared with cycles of 12 h light-12 h dark with white or longer (LWL), medium (MWL) or short wavelength light (SWL) for 12 days). Retinas were analyzed after labeling with calretinin (CR) and the transcription factors, Islet-1, LIM 1 +2. Animals reared in MWL showed an increase in the number of horizontal cells islet-1+, and a decrease in LIM 1+2. Conversely, LWL stimulated chicks showed Islet-1 diminished and LIM1+2 increased. Besides, in the SWL group we observed a decrement in LIM1+2. Also changes were observed in the pattern of horizontal cells connections with cones. These results show that monochromatic stimulation change gene expression as well as modify the connectivity of horizontal neurons. Postnatal visual experience showed plastic changes in the neuronal circuits in which subtype H1 and H3 of horizontal cells are involved. PIP00404

Cellular and Molecular Neurobiology
Poster Number 9 | Session 3

"Rapid Endocytosis in mouse chromaffin cells is regulated by

intracellular calcium sources"

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Endocytosis is critical for maintaining membrane homeostasis and secretion reliability in neuroendocrine cells and neurons. Chromaffin cell rapid endocytosis (RE) is a Ca2+ dependent process, which can overcome the previous exocytosis if the Ca2+ entry is ≥ 75pC (excess retrieval, EX). In order to study if intracellular Ca2+ sources may regulate RE, we performed patch clamp capacitance measurements and applied specific pharmacological agents. The global participation of Ca2+ release from endoplasmic reticulum (Er) was studied by depleting this organelle with thapsigargin+caffeine pretreatment, and the specific contribution of ryanodine receptors was studied by ryanodine application. We observed similar effects with both treatments (a reduction of RE by 40 and 60% respectively). We next studied the participation of Ca2+ release through IP3 receptors (IP3R). On one hand, IP3R inhibition with 2-APB, partially reduced RE (by 50%), blocked EX and made the kinetic slower. But on the other hand, heparin, a more specific IP3R blocker, did not mimick those effects. This could be explained by the reported inhibitory effect of 2-APB over the SERCA, what would reduce the Er Ca2+ content. These results together suggest that RE is regulated by Ca2+ release through ryanodine receptors.

Cellular and Molecular Neurobiology
Poster Number 10 | Session 1

"Mechanisms involved in Chromaffin Cell Rapid Endocytosis"

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Endocytosis is critical for maintaining membrane homeostasis and secretion reliability in neuroendocrine cells. Chromaffin cell rapid endocytosis (RE) is Ca2+ dependent and presents two kinetic components, R1<1s and R2~8s. In this work we started to study which endocitotic mechanism (kiss and run, bulk, clathrin dependent endocytosis (CDE)) are associated to R1 and/or R2 components. We used drugs against different steps of endocytosis and measured cell membrane

capacitance by the patch clamp technique. Dynasore (80uM), a non competitive inhibitor of dinamin (a GTPase involved in the closure of fission pore in kiss and run and CDE) partially inhibited RE (by 50%). Although it is classically assumed that CDE is a slow process, it was recently described a CDE in beta cells with a time evolution similar to our R2 component. Considering this fact, we used chlorpromazine 15uM, a drug that interferes with the assembly-disassembly of clathrin in the coated pits. RE was partially inhibited (by 60%), R2 component was 3.5 times slower and surprisingly R1 component was almost lost (it appeared in the 15% of the cases, while in control cells in the 75%). These preliminary results suggest that a significant portion of RE in chromaffin cells might be accounted by a clathrin dependent process.

Cellular and Molecular Neurobiology
Poster Number 11 | **Session 2**

"Kinases activation by uPA-uPAR complex, located in raft microdimains, is responsible of neurite outgrowth"

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Neuritogenesis requires control of mechanisms that include the activation of kinases and reorganization of the cytoskeleton of axonal growth cones.The complex formed by the urokinase-type plasminogen activator (uPA) and its receptor (uPAR) acts on: proteolysis of the extracellular matrix located on the cell surface and signal transduction which promotes changes in the cell behavior. We showed that the uPA stimulates neuronal migration and neurite outgrowth independently of their proteolytic role, throughout activation of the Focal Adhesion Kinase (FAK). Now we investigated: whether the uPA:uPAR must be located in raft in order to initiate the phosphorylation of FAK; the activation of extracellular signal-regulated kinase (ERK) promoted by uPA and the relationship between the temporo-spatial expression of uPAR and the development of chicken Optic Tectum (OT). We observed that: (I) the maintenance of ratf is necessary for the uPA:uPAR effect in phosporilation events; (II) a short stimulation with 10nM uPA promoted an increase of ERK phosphorylation. (III) Immunohistochemical analysis of OTs of different development stages showed that uPAR expression is related to neuronal populations submitted to intensive migration during the lamination process.
Supported by UBA-CONICET.

Cellular and Molecular Neurobiology
Poster Number 12 | Session 3

"CG6115, a novel gene involved in neurodegeneration in Drosophila melanogaster, is needed in muscle during development"

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To identify novel genes related to neurodegeneration a missexpression screen has been carried out in our laboratory. It consisted of examining locomotor behavior in young and aged flies. Flies that showed a progressive loss of rhythmic activity could reveal novel genes involved in neurodegenerative mechanisms. One of the mutants, 100B, shows a striking downregulation of CG6115 expression. CG6115 encodes a gene of unknown function. Homozygous mutants cannot progress beyond second instar larvae and have an abnormal feeding behavior. CG6115 relevance in different tissues was assayed and muscle related GAL4s were found to mimic homozygous mutant behavior in larvae. Interestingly, adult flies expressing the RNAi specific to CG6115 in pigment dispersing factor positive neurons, when screened in locomotor behavior, showed progressive arrythmicity and period lengthening. In addition, flies expressing CG6115 RNAi under the glass Multimer Reporter promoter show severe eye defects in approximately 23% of the individuals. A preliminary analysis of brain slices of young and aged flies showed that expression of the specific RNAi in the brain causes progressive vacuolization. Taken together we propose CG6115 could play a role in neurodegeneration.

Cellular and Molecular Neurobiology
Poster Number 13 | Session 1

"Wnt3a triggers polarization of hippocampal neurons via activation of the IGF-1 receptor/PI3k pathway"

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The selection of the future axon in cultured hippocampal neuron requires the activation of IGF-1 receptor (IGF-1r), PI3k and the accumulation of PIP3 at the growth cone (Shi et al., 2003; Menager et al., 2004; Sosa et al., 2006). Growth factors belonging to the Wnts family have been also implied in the regulation of axonal development (Arevalo and Chao, 2005). It is not known, however, if Wnts have any participation in the regulation of initial axonal outgrowth and the establishment of neuronal polarity. We used cultured hippocampal pyramidal neurons and growth cone particles (GCPs) isolated from fetal rat brain to show that stimulation with Wnt3a is sufficient to trigger neuronal polarization in the absence of IGF-1 or a high level of insulin. We also show that Wnt3a triggers the activation of IGF-1r and PI3k (polarized to one minor neurite) in neurons in stage 2 of development. In addition, we show that the presence of activatable IGF-1r and PI3k activation are necessary for Wnt3a polarizing effects. Finally, using crosslinking and immunoprecipitation experiments, we show that Wnt3a directly binds to IGF-1r. We conclude that Wnt3a triggers polarization of hippocampal neurons via direct activation of the IGF-1r/PI3k pathway.

Cellular and Molecular Neurobiology
Poster Number 14 | **Session 2**

"Class 3 Semaphorins in a mouse model of autism spectrum disorder"

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Methyl Cytosine Binding Protein 2 (MeCP2) is a transcriptional repressor that binds to methylated DNA. Alterations in the expression levels of Mecp2 have been related to autism spectrum disorders. Studies in mouse models of Mecp2 deficiency demonstrated that this protein is important for neuronal maturation, neurite complexity and synaptic plasticity, although the underlying mechanisms are not completely understood. Our working hypothesis proposes that Mecp2 plays a role in the formation of neural circuits during development. In particular, we aim to evaluate the role of this protein in axonal guidance processes during early postnatal stages. In the present work, we used mouse models of Mecp2 deficiency to analyze the expression of members of the Class 3 Semaphorins, a family of guidance molecules that play an important role in the establishment of neural connectivity. Our results show differential expression of Semaphorins receptors in hippocampus from Mecp2-null mice during early postnatal development. These

results suggest that Mecp2 regulates this family of guidance cues in a direct or indirect manner. Future studies will explore how these changes in guidance cues levels could affect the formation of the hippocampal circuitry.

Cellular and Molecular Neurobiology
Poster Number 15 | Session 3

"Experimental approaches for disrupting APP/Aß interaction"

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Increasing evidence suggest that the interaction of Amyloid beta (Abeta) with its parental protein, the Amyloid beta Precursor protein (APP), might play a pathogenic role in Alzheimer¥s disease. It was suggested that the binding site sequence on APP is located in its extracellular juxtamembrane domain. Here, we describe two different experimental approaches for further defining the binding sequence. First, we generated a series of point mutations within APP juxtamembrane domain. These mutations were directed to specific aminoacids critical for beta sheet conformation. Second, we used purified enantiomer or the racemic mix of different profens which have been described to bind to APP yuxtamembrane domain specifically. The effects of these approaches on Abeta-APP interaction was mainly analyzed by using cell cultures and coprecipitations assays. The relevance of the results will be discussed.

Cellular and Molecular Neurobiology
Poster Number 16 | Session 1

"Irreversible incorporation of L-Dopa into the C-terminus of α-tubulin as a possible cause of side effects during administration of L-Dopa to parkinson patients"

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L-Dopa can be added to the C-terminus of α-tubulin by tubulin tyrosine ligase, one of the enzymes involved in the postranslational cyclic tyrosination/detyrosination of tubulin. Now, we show that after its incorporation into tubulin present in soluble extracts from rat brain, L-Dopa cannot be released by tubulin carboxypeptidase, the other enzyme participating in the cycle. L-Dopa-tubulin can form microtubules as well as Tyr-tubulin. Amount of Dopa-tubulin in soluble extracts was inferred by analyzing the tyrosination state of tubulin by Western blots revealed with anti-total, Tyr-, Glu- and Δ2-tubulin. We suggest that during a prolonged administration of L-Dopa to Parkinson patients, L-Dopa-tubulin within cells gradually increases (and other tubulin species decrease), resulting in the formation of microtubules containing L-Dopa-tubulin that affect some of their properties, and this could be in some way responsible for the side effects of prolonged treatment with L-Dopa.

Cellular and Molecular Neurobiology
Poster Number 17 | **Session 2**

"Cholesterol modulates the rate and mechanism of acetylcholine receptor internalization"

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Stability of the nicotinic acetylcholine receptor (AChR) at the cell surface is key to the correct functioning of the cholinergic synapse. Cholesterol (Chol) is necessary for homeostasis of AChR levels at the plasmalemma and for ion translocation. Here we characterize the endocytic pathway followed by muscle-type AChR in Chol-depleted cells (Chol(-)). Under such conditions, the AChR is internalized by a ligand-, clathrin-, and dynamin-independent mechanism. Expression of a dominant negative form of the small GTPase Rac1, Rac1N17, abolishes receptor endocytosis. Unlike the endocytic pathway in control CHO cells, accelerated AChR internalization proceeds even upon disruption of the actin cytoskeleton. Under Chol(-) conditions, AChR internalization is furthermore found to require the activity of Arf6 and its effectors Rac1 and phospholipase D. The Arf6-dependent mechanism may constitute the default endocytic pathway followed by the AChR in the absence of external ligands, membrane Chol levels acting as a key homeostatic regulator of cell surface receptor levels.

Cellular and Molecular Neurobiology
Poster Number 18 | Session 3

" Partial characterization of the PMCA-acetylated tubulin complex"

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Plasma Membrane Calcium ATPase (PMCA) is an integral membrane protein that pumps calcium ions from the cytoplasm to the extracellular space. We previously showed that this enzyme interacts with acetylated tubulin, resulting inhibited. Now, we determined that: 1) PMCA from a detergent solubilized membrane preparation coelutes with acetylated tubulin on molecular exclusion chromatography with a MW= 450 KDa (approx.). The discrete MW of the PMCA/tubulin complex suggests that proteins are not part of a membrane fragment but it is formed by a reduced number of tubulin and PMCA (or other) molecules. 2) In vitro, PMCA interacts preferentially with microtubules containing acetylated tubulin. This finding is compatible with a scenario within the cell where PMCA acts as an anchorage site of microtubules with plasma membrane. 3) By testing separately each cytoplasmic fragment of PMCA, we suggest that the cytoplasmic domains CD2 and CD3 are the sites through which PMCA interacts with acetylated tubulin.

Cellular and Molecular Neurobiology
Poster Number 19 | Session 1

"Expression of the bHLH-Transcription Factor Ascl1 Is Specifically Required for the Development of Spinal CSF-contacting Neurons"

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One central problem in developmental neurobiology is to understand the mechanisms that control neuronal diversification in the nervous system. We identified that cerebrospinal fluid contacting neurons (CSF-cN) in the spinal cord are a subset of V2 neurons that differentiate late during mouse embryonic development. In searching for potential controllers of CSF-cN specification, we found that the proneural protein Ascl1 is expressed in a group of ventral neural

tube progenitors, correlating spatially and temporally with CSF-cN development. Expression analysis and short-term lineage tracings indicated that this transcription factor is expressed in CSF-cN progenitors. In addition, we found that in Ascl1-/- mice CSF-cN population is missing, while other cell types generated earlier from the same domain remain unaffected. This phenotype is not rescued by another proneural protein, Neurogenin2, suggesting that Ascl1 activity is specifically required. Finally, taking advantage of the temporally restricted expression of Ascl1 in Foxn4 null mice, we concluded that Ascl1 exerts its actions around the time of CSF-cN differentiation. In summary, our results show that Ascl1 is expressed in spinal ventral late CSF-cN progenitors and that it plays an essential and specific role in their genesis.

Cellular and Molecular Neurobiology
Poster Number 20 | **Session 2**

"Blood Brain Barrier (BBB) and behavioral early alterations by endovenous (ev) administration of Shiga toxin 2 (Stx2) from enterohemorrhagic Escherichia coli (STEC) in mice"

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Stx2 from STEC causes hemorrhagic colitis, Hemolytic Uremic Syndrome, and neurological damage. Since the entrance to the brain of Stx2 is through the BBB, the aim of this study is to determine BBB and behavioral alterations in mice after ev Stx2 administration. NIH mice were anesthetized, perfused and their brains were processed to perform confocal histofluorescence in the hippocampus by binding of Lycopersicum esculentum lectins to endothelial cell membranes after 0, 24 and 48h. Shirpa test, blood urea, creatinine and weight variation measurements were also done. An increased microvessel area and intensity, mild neurological dysfunction, and increased levels of blood urea and creatinine were observed after 48h of ev Stx2 treatment ($p<0,05$). Significant changes in body weight were observed since 24h. The early changes found in the microvasculature profile correlates well with neurological dysfunctions. Therefore the analysis of the microvasculature profile could be a suitable pathologic marker during the acute stage of brain STEC intoxication in animal models and in patients by using cerebral angiography.

Cellular and Molecular Neurobiology
Poster Number 21 | Session 3

"GLUN2A-containing NMDA receptor expression increases after LTP induction in adult rat hippocampus"

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NMDA receptors (NMDAR) are tetramers containing 2 GLUN1 and 2 GLUN2 or GLuN3 subunits. The major hippocampal GluN2 subunits are GLuN2A and GLuN2B. GLuN2B predominates at early stages, while GLuN2A becomes prevalent later. Rapid electrical changes suggested that synaptic GLuN2A increased while GLuN2B decreased after LTP induction in acute slices that was attributed to an increase in GLuN2B endocytosis followed by delivery of available GLuN2A-NMDAR to the synaptic membrane. Little is known about what happens with the expression (transcription-translation) of GLuN2 subunits during LTP. To evaluate NMDAR subunits expression after LTP induction, field potential recordings from hippocampal fresh slices of young adult rats (P42-60), stimulated at the level of Schaffer collaterals, were performed. LTP was induced by theta burst stimulation (TBS). Then, the expression of NMDAR subunits was analyzed by WB at different periods after LTP induction. 70 min after LTP induction, GLuN1 and GLuN2A expressions were significantly different from control slices. To elucidate if these changes are also expressed in membranes, further assays are being carried out.

This work was supported by UBA (UBACyT), CONICET (PIP) , and LIA DEVENIR (CNRS/CONICET)

Cellular and Molecular Neurobiology
Poster Number 22 | Session 1

"Possible mechanisms involved in the elongation/retraction of CAD cells neurites"

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CAD cells (derived from a mouse brain tumor) have a rounded shape and proliferate in standard culture medium. When deprived of serum, cells stop proliferation and emit long processes similar to normal neurons. Neurites rapidly retract when serum is re-added and cells resume growth. We found that microtubules (MTs) in these cells are highly dynamic, many of the major MAPs are absent, tubulin is mainly tyrosinated, and the acetylated isoform is not detected. We have determined that integrity and disassembly of MTs is required for neurite elongation and retraction, respectively. A transient increase of acetylated tubulin neither increased stability of MTs nor impeded retraction of neurites. Conditions that enhance the interaction of MTs with Na,K-ATPase do not alter elongation or retraction. Treatment of cells with lysophosphatidic acid or with adenosine deaminase, in the absence of serum, induced neurite retraction. Lack of ATP or the disassembly of microfilaments prevent retraction, suggesting that the actomyosin system is part of the retraction mechanism.

Cellular and Molecular Neurobiology
Poster Number 23 | **Session 2**

"Retinal damage by low light intensity exposure: a model of retinal degeneration in mammals"

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Retinal degeneration by defect in phototransduction mechanism is characterized by apoptosis of photoreceptors cells. Constant low light exposure produces photoreceptor cell death through the activation of downstream signal transduction. The present study examined the time course and molecular apoptotic mechanism occurring during retinal degeneration after continuous low light intensity exposure. Wistar rats were exposed to constant illumination with cool white fluorescent light (LL) of 200 lux for 1 to 10 days and compared with controls kept in the dark (DD) or exposed to a regular 12:12 h (LD) cycle. Histological analysis showed a significant reduction of the outer nuclear layer (ONL) after 4 days of LL as compared with LD or DD controls. Retinal analysis by flow cytometry showed an increase of apoptosis in light-exposed rats. Moreover there was a progressive collapse of the outer segments (OS) and inner localization of rhodopsin immunoreactivity in the photoreceptor somas during LL exposure. The study of activated caspase-3 demonstrated a caspase-3 independent mechanism by both Western blot and enzyme activity assays.

Cellular and Molecular Neurobiology
Poster Number 24 | Session 3

"Protein Kinase D1 (PKD1)-dependent neurotrophin receptor TrkA trafficking and sorting"

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After determining the role of several proteins, such as PKD1, in the regulation of intracellular trafficking, we have decided to study the role of these regulatory proteins in the establishment of neuronal polarity, specifically through the analysis of TrkA receptor traffic and sorting. This receptor is actively localized in the axonal terminal, reaching that specific neuronal region by ligand (NGF)-dependent transcytosis at the dendrites. Without NGF, EGFP-TrkA normal distribution is mostly somatodendritic. In the absence of an active PKD1, we have observed a remarkable decrease in the TrkA distribution at the neuronal processes, together with its accumulation in large vesicles that colocalize with Golgi at the neuronal soma. In addition, there was a significant reduction in the length of dendrite branches. We have also analyzed the effect of inactive PKD1 when NGF was present. Even though the length of dendrites was reduced as it was observed without NGF, there was a change in the arborization pattern and in the morphometric variables of dendritic branches. These results would confirm that PKD1 play a key role in the fission regulation of vesicles carrying TrkA, and that the presence of this receptor at the neuronal surface would be essential for dendrite length development.

Cellular and Molecular Neurobiology
Poster Number 25 | Session 1

"Neurotoxicity of Glyphosate involves changes in Wnt5a expression in vivo and in hippocampal neurons in culture"

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Glyphosate is known as the active ingredient of Roundup, it is widely used as a non-selective herbicide. There are few evidences about its toxicity and the mechanism of action on mammals. In this work, we study the potential effect of

glyphosate on nervous system during development. We designed in vivo assays and neuronal cultured experiment to analyze its potential toxicity. Results showed that animals exposed to the glyphosate during gestational period revealed an impairment of reflex responses and locomotor activity. Furthermore, in vitro assays from hippocampal cultured neurons exposed to glyphosate showed a delay in their differentiation. Thus, glyphosate treated neurons did not polarize after 24h in culture and neurons exposed for 2 days showed one axon and few dendrites which exhibited a significant decrease in length and complexity. Importantly, glyphosate treated neurons showed a significant decreased in the expression of Wnt5a, an essential factor for normal axon outgrowth and development. Glyphosate effect on neuronal morphology was reverted when exogenous Wnt5a factor was added to the medium. These evidences suggest that the glyphosate is a potential neurotoxic which affect the neuronal development and functioning involving essential Wnt-signalling pathways.

Cellular and Molecular Neurobiology
Poster Number 26 | **Session 2**

"Identification of Axonal transport properties of VMAT-YFP fluorescent vesicles"

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Parkinsonís disease (PD) is characterized by reductions in dopamine (DA) neurotransmitter release, abnormal protein accumulation, and selective degeneration of DA neurons. DA neurotransmission rely on ATP-dependent internalization by the Vesicular Monoamine Transporter 2 (VMAT2). VMAT2 reductions in mice induce PD phenotypes and neurodegeneration suggesting the relevance of its distribution along neurons. To identify VMAT2 axonal transport properties we generate a protein fusion to YFP (VMAT2-YFP). Initially, sucrose flotation assays in transfected N2a cells revealed VMAT2-YFP association to membranes. Movies from transfected primary hippocampal neurons under live imaging experiments revealed a vesicular axonal transport for VMAT2-YFP with 22% of anterograde, 20% of retrograde and 58% of stationary particles. Average speeds for anterograde (1.11um/sec) and retrograde (0.62um/sec) vesicle displacements correspond to motor dependent movement. These results suggest that VMAT2 axonal transport is necessary for normal DA neurotransmission and implies a high relevance for the identification of the motor moving VMAT2. In

addition, VMAT2 axonal transport defects might have a significant impact in the manifestation of PD phenotypes and in the progression of DA neurodegeneration.

Cellular and Molecular Neurobiology
Poster Number 27 | **Session 3**

"Changes in levels of BDNF / proBDNF and its receptors, p75ntr and TrkB, in a Model of Status Epilepticus in vitro"

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Previous studies from our laboratory showed that neuronal death induced by Status Epilepticus (SE) depends on the levels in the plasma membrane of the receptors TrkB and p75ntr (Unsain et al, 2008, 2009). To determine the detailed mechanisms of this phenomenon, we characterize and evaluate whether an in vitro model of SE is possible to reproduce the same results as those obtained in vivo. A culture of hippocampal neurons were treated with Mg+2-free medium, which induces sustained electrical hyperactivity, similar to that observed in SE in vivo, until the return of maintenance medium. Immediately after starting this procedure, there was an increase in intracellular Ca+2 levels. It was noted that 3 hours of hyperactivation induces neuronal death, which begins to be significant at 6 hours after the conclusion of the SE. Hyperactivation caused significant increases in the mobilization to the plasma membrane of TrkB and p75ntr. Preliminary observations indicate that this time there is an increase in levels of BDNF, and in the release of proBDNF. The results indicate that this model is appropriate to deepen understanding of the involvement of BDNF / proBDNF and their receptors in the mechanisms of neuronal death and survival

Cellular and Molecular Neurobiology
Poster Number 28 | **Session 1**

"Functions of Retinoic Acid during photoreceptor development in vitro"

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Retinoic acid (RA) has a critical role in promoting cell differentiation in several tissues, including the retina. In addition, RA induces apoptosis in diverse cell types. In this work we investigated the effects of RA in retina rod photoreceptors (PHRs). Cultures prepared from newborn rat retinas, grown in chemically defined media, were supplemented with RA and/or docosahexaenoic acid (DHA), a molecule that promotes PHR survival. RA advanced PHR differentiation: it increased opsin and peripherin expression and promoted axon outgrowth in PHRs. Activation of p38 was involved in RA effects. RA promoted the phosphorylation of p38, while a p38 inhibitor blocked RA effect on PHR differentiation. RA also accelerated the onset of PHR apoptosis, which occurs in cultures lacking PHR trophic factors. RA-induced apoptosis was blocked with a caspase inhibitor and also prevented by pre-incubating the cultures with DHA. In summary, this work shows that RA induced both the early differentiation and apoptosis of PHRs in vitro and this death can be prevented by DHA. These results suggest that since RA is critical to stimulate PHR early differentiation, PHRs require a simultaneous provision of survival factors to advance their differentiation and avoid cell death during development.

Cellular and Molecular Neurobiology
Poster Number 29 | Session 2

"Eph-ephrin system is expressed in the chicken regenerated retina by stem cells and transdifferentiation"

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Chick embryo has the capability to regenerate its retina by stem cells of the margin zone (MZ) or transdifferentiation of the retinal pigmented epithelium. To obtain a functional retina, the retinal ganglion cells (RGC) must form topographic connections in the tectum. Ephs and ephrins are the main molecular system involved in this process. Our aim was to analyze the expression of Eph-ephrin system to know if the regenerated retina has the potential to connect to the tectum. Retinectomies and FGF-2 treatment were performed in 4 days embryos (E4). We used immunocitochemistry of 3 and 7 days postretinectomy (3d, 7d), and of E7 and E11 control retina. We compared the expression of Eph-ephrin between 3d-E7 and 7d-E11. 3d and 7d transdifferentiated retina presented lower expression of Ephs and ephrins than E7 and E11 retina. Regenerated retina from MZ presented

a similar level of Ephs and ephrins expression than E7 and E11. Both kinds of regenerations express the Eph-ephrin system which gives the RGCs the potential to form ordered retinotectal connections. However, as the transdifferentiated retina does not form optic nerve, regeneration from CM cells offers better results.
Founded by UBA-CONICET, Miami University.

Cellular and Molecular Neurobiology
Poster Number 30 | Session 3

"EphA4 vs EphA3: fight for the ephrin-As"

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Ephrin-As of the caudal tectum repel temporal (T) axons of retinal ganglion cells (RCG) by activating EphAs. We established that EphA3 stimulates nasal (N) RGCs axon growth to caudal tectum and inhibits branching rostrally to their target area. Our aim was to study the molecular way of action of EphA3. We used retinal explants from chicken embryos treated with EphA3Fc/Fc, PIPLC (sheds ephrin-As) or KYL (inhibits ephrin-A-mediated EphA4 activation). We performed immunocytochemistry, immunoprecipitation and Western blot. NRGCs present higher levels of ephrin-As and EphA4ptyr602. Ephrin-As and EphA4 coexpress mainly in NRGCs. As TRGCs grow longer axons and present less filopodia and NRGCs present the higher respond to EphA3Fc, it was suggested that ephrin-As-mediated EphA4 forward signaling decreases axon growth and stimulates branching whereas the opposite effects produced by EphA3 are mediated by decreasing EphA4 signaling throughout competition of EphAs binding to ephrin-As. We showed that removing axonal ephrin-As or inhibiting EphA4 activation reproduces the effects of EphA3.These results support a novel mechanism of action of EphA3 whereby plays their roles by diminishing the ephrin-As-mediated EphA4 forward signaling.
Founded by UBA-CONICET.

Cellular and Molecular Neurobiology
Poster Number 31 | **Session 1**

"The ETS transcription factor, Etv5, mediates biological response to NGF"

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Nerve Growth Factor (NGF) is a prototypic neurotrophic growth factor that control many aspects of sensory and sympathetic neuronal development. The identification of transcription factors and downstream target genes that mediate NGF-dependent neuronal differentiation and target field innervation is currently a major challenge. Using microarray screening, we identify the ETS transcription factor Etv5 (Erm) as a potential mediator of NGF signaling. Real time PCR assays confirmed that this transcription factor is significantly induced by NGF in different neuronal cells, suggesting that it could be involved in the biological responses induced by this neurotrophin. Pharmacological assays also revealed that activation of Erk/MAPK pathway is required for the induction of Etv5 mRNA in response to NGF. Down-regulation of Etv5 using small interference RNA knock-down experiments inhibited NGF-induced neurite outgrowth. Together, these data establish Etv5 as an essential molecule of the transcriptional program linking neurotrophin signaling to neuronal differentiation. ular and Molecular Neurobiology

Cellular and Molecular Neurobiology
Poster Number 32 | **Session 2**

"Functional analysis of single nucleotide polymorphisms (SNPs) present in GPM6A'S transmembrane domains coding region"

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Neuronal glycoprotein M6a is involved in neuronal plasticity (e.g. neurite outgrowth, filopodium and synapses formation) through unknown mechanisms. M6a has four transmembrane domains (TMs) and shares structural similarity

with the tetraspanin protein family. The function of several tetraspanins is mediated by their ability to self-associate and interact with different membrane proteins and lipids through specific alpha-helix residues. Our aim is to assess the association between genetic variants in GPM6A's TMs and filopodium induction in hippocampal cultured neurons. Three replacement polymorphisms, here named SNP1 (F93C) and SNP2 (I97S) in TM2 and SNP3 (W141R) in TM3, were found in the TMs coding regions of the GPM6A gene. We used site-directed mutagenesis to construct a panel of mutants with the SNPs introduced into the M6a-EGFP-C1 plasmid. To study the ability of M6a to induce filopodium formation, the different mutants were overexpressed in primary culture of hippocampal neurons. Our results showed that overexpression of all SNPs significantly decreased filopodium density compared with control group. These results lead to the conclusion that M6a SNPs reduce neuronal plasticity, probably due to altered transmembrane helix interactions.

Cellular and Molecular Neurobiology
Poster Number 33 | **Session 3**

"Role of ENA/VASP in the adult brain of Drosophila melanogaster"

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Neuronal polarity is essential for input/output processing and appropriate flow of information in neuronal networks (Sanchez-Soriano et al., 2007). The polarized morphology of neurons is maintained and dynamically modified by microtubules and the actin cytoskeleton during brain development (Conde and Caceres, 2009). Among actin regulatory proteins, ENA/VASP is a conserved family which is critical for filopodia formation and elongation. In a previous work we showed that ENA/VASP downregulation in culture of mouse hippocampal neurons generate axonal retraction, which subsequently induces neuronal death through an apoptotic mechanism (Franco et al., 2010). To further characterize the mechanism of axonal retraction triggered by ena in vivo we downregulated the expression of ena in the circadian circuit of adult brain of Drosophila. Preliminary results indicate that reducing ena levels leads to axonal retraction and decrease the number of active zones within the circadian circuit. Taking in account these results we will further evaluate synaptic activity on these neurons and the behavioral phenotype induced after down-regulating ena levels in the circadian circuit.

Cellular and Molecular Neurobiology
Poster Number 34 | Session 1

"Protein tyrosine phosphatase PTP1B is required for synapse formation"

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Protein tyrosine phosphatase PTP1B dephosphorylates B-catenin and promotes its binding to the N-cadherin cytoplasmic domain. We have recently shown that PTP1B localizes in dendrites, suggesting a role in the synaptic compartment. Here we show, by double fluorescence time-lapse analysis, that GFP-PTP1B invades transiently dendritic spines of hippocampal neurons in culture. GFP-PTP1B overexpression does not affect filopodial density and length. Dominant negative disruption of PTP1B function and gene deletion, lead to increased length of dendritic filopodia and reduction of mushroom-like spines, accompanied by a disorganization of pre- and post-synapsis, as judged by decreased clustering of synapsin-1 and PSD-95. Immunoprecipitation of N-cadherin from hippocampi of adult wild type (WT) and PTP1B knockout (KO) mice reveals a 30% reduction in the amount of associated B-catenin in KO mice. This is accompanied by a ~5-fold increase in the levels of phosphorylated tyrosine 654 on B-catenin. BDNF-dependent phosphorylation of B-catenin Tyr-654 is attenuated by PTP1B. Our results suggest that PTP1B is required for synapse maturation, likely by modulating N-cadherin-mediated adhesion through B-catenin dephosphorylation of Tyr-654.

Cellular and Molecular Neurobiology
Poster Number 35 | Session 2

"Opposite pre-and post-synaptic adaptations in the glutamatergic transmission in nucleus accumbens core, but not shell, underlies the long term sensitization to cocaine after a single restraint stress"

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Several evidences indicate that cocaine is able to induce behavioural and molecular sensitization in both, the dopaminergic (DA) and glutamatergic (Glu) mesocorticolimbic systems. The aim of this work was to study if the basal concentration of extracellular Glu as well as the AMPA receptor expression (AMPAR) participates in the long-term cross-sensitization between stress and cocaine.Wistar rats were restrained for two hours, while control animals were left undisturbed in their cages.Twenty-one days after this stress episode: I) Microdialysis: basal extracellular and saline- or cocaine (15 mg/kg ip) induced Glu levels were determinated by HPLC in NAc Core and Shell.II) AMPAR in Core after ip injection of saline or cocaine. Our results demonstrate that pre-stressed animals have raised basal extracellular concentrations of glutamate in the core, but not shell, and this could be related with the decrease in the cocaine-induced glutamate release in the core as compared with control values. We propose that the increase observed in the AMPAR in pre-stressed animals could be associated with the decrease in glutamate release after stimulus. These results are discussed in the context of disrupted glutamate homeostasis induced by acute stress 21 days before the cocaine challenge.

Cellular and Molecular Neurobiology
Poster Number 36 | Session 3

"Participation of BARS in membrane trafficking regulation in developing neurons"

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The ability of cells to produce highly compartmentalized membrane domains and hence to polarize is crucial for complex biological activities, such as the organization of the nervous system. Disruption of the endoplasmic reticulum (ER)-Golgi secretory pathway in developing neurons alters axon-dendritic formation. Therefore, detailed knowledge of the mechanisms underlying exiting from the Golgi is crucial for understanding neuronal polarity. In this study we have analyzed the role of Brefeldin A-Ribosylated Substrate (BARS) in the regulation of morphological polarization, the formation of Golgi outposts and the exit of membrane proteins from the TGN. The results obtained show that RNAi suppression of BARS inhibits axonal/dendritic elongation and branching, as well as the extension of Golgi-

outposts into dendrites. In addition, using a plasma membrane (PM) protein (e.g.transferrin receptor [TfR] fused to GFP) engineered with reversible/removable aggregation domains we observed that suppression or expression of DN-BARS delay the exit of TfR from the Golgi apparatus. Taken together, these data provide the first set of evidence suggesting a role for BARS in neuronal polarization by regulating membrane trafficking and organelle positioning.

Supported by ANPCyT y Agencia Córdoba Ciencia.

Cellular and Molecular Neurobiology
Poster Number 37 | Session 1

"Activation of a retinoid orphan receptor is required for docosahexaenoic acid protection of photoreceptors"

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Docosahexaenoic acid (DHA), the major omega-3 polyunsaturated fatty acid in the retina, promotes the survival of rat retina photoreceptors (PR) during early development in vitro and upon oxidative stress by activating the ERK/MAPK signaling pathway. We investigated if DHA activates this pathway by direct activation of tyrosine kinase receptors (TRK) or of retinoid nuclear receptors (RXR). Using retinal neuronal cultures we determined that DHA prevented PR apoptosis at early culture times in spite of the presence of a TRK inhibitor (K252a), implying TRK are not involved in its effects. On the contrary, RXR antagonists (HX531 or PA452) inhibited DHA protection during early development in vitro and upon paraquat and H2O2-induced apoptosis. Moreover, RXR agonists (HX630 or PA024) decreased ROS production in H2O2-treated neuronal cultures, as we previously showed for DHA. To evaluate whether DHA has to be released from phospholipids to exert its protective effect, DHA-supplemented cultures were treated with a phospholipase A2 inhibitor (BEL) prior to H2O2 treatment; BEL addition blocked DHA protection on PR upon oxidative stress. These results suggest a new pathway for DHA actions in PR: it is first released from phospholipids and then activates RXR to promote PR survival.

Cellular and Molecular Neurobiology
Poster Number 38 | Session 2

"Analysis of Amyotrophic Lateral Sclerosis Immunoglobulin-G interaction with pre-synaptic proteins"

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Amyotrophic Lateral Sclerosis (ALS) is a neurodegenerative disease characterized by a progressive loss of motoneurons. The majority of cases belong to a sporadic form with unknown etiology. Several pieces of evidence support autoimmunity as one of the possible mechanisms contributing to ALS. We detected a diminished interaction of ALS antibodies with neuromuscular junctions from CaV2.1-deficient mice, and a lack of effect on spontaneous synaptic activity. The aim of our research was to evaluate the interactions between ALS-IgGs and P/Q-type calcium channels, in addition to other pre-synaptic proteins. We used HEK cells transfected with the P/Q-type calcium channel and analyzed cell lysates by Western blotting or immunoprecipitation assays with ALS-IgGs. We also analyzed ALS-IgGs immunoprecipitates from synaptosomal proteins for the presence of synaptic markers. None of the evaluated ALS-IgG samples exhibited affinity towards the pore-forming subunit expressed in HEK cells, evaluated either by western blotting or immunoprecipitation assays. The analysis of synaptosomal proteins immunoprecipitated with ALS antibodies could not revealed the presence of the alpha1 subunit or other synaptic proteins. Further studies will investigate the interaction with other synaptic components.

Cellular and Molecular Neurobiology
Poster Number 39 | Session 3

"Lysosomal dysfunction is involved in apoptotic cell death in a Parkinsonism model"

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Lysosomal dysfunction has been widely related to neurodegeneration. Moreover, recent reports have linked abnormal protein accumulation in Parkinson¥s disease to ROS-mediated lysosomal permeabilization, degradation failures, proteases translocation to cytosol (e.g. Cathepsins B/D) and cleavage of ectopic targets and cell death. Manganism is a Parkinsonism induced by chronic manganese (Mn) overexposure. Considering that Manganism shares biochemical pathways with Parkinson's disease and several studies point out astrocytes as the first target of Mn- induced damage, the aim of the this work was to study the possible link between lysosomal disruption and apoptotic cell death in rat astrocytoma C6 cells. Using LysoTracker staining we detected large structures resembling lysosomal inclusion bodies. Moreover, Cathepsin B inhibitor (Ca-074 Me, 1μM), Cathepsin D inhibitor (Pepstatin A, 10μM) and the V-ATPase inhibitor (Bafilomycin A1, 0.1nM) prevented cell death, caspases-8 and -3 activation and the FasL protein levels increment. These novel data suggest that lysosomal pathway is a potential target for therapeutic intervention in Mn-induced Parkinsonism and probably in other neurodegenerative conditions.

Cellular and Molecular Neurobiology
Poster Number 40 | **Session I**

"Mutations in the Go-interacting domain of APP protects neurons for Amyloid beta toxicity"

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Amyloid beta (A*ß*) is a metabolic fragment of the Amyloid *ß* precursor protein (APP).Deposition of A*ß* in the brain and neuronal degeneration are characteristic hallmarks of Alzheimerís disease(AD).Our lab have previously showed that interaction of A*ß* with APP induce neurodegeneration by a mechanism that involves heterotrimeric Go protein activation,suggesting an A*ß*-receptor-like role of APP in neuronal degeneration.In this presentation,to further study the signaling mechanism involved in APP-dependent toxicity of A*ß*,we performed site directed mutagenesis in the APP-Go protein interaction domain.We found that these mutations abrogated the A*ß* toxicity.In addition,we found that overexpression of wild type G*α*o subunit abolished the A*ß* toxicity in a dose dependent manner,suggesting that APP/Go-dependent toxicity of A*ß* is not mediated by G*α*o subunit-signaling.Moreover,the overexpression of both,the c-terminal fragment of the *ß* adrenergic receptor-kinase(*ß*-ARK-CT)and a prenylation-deficient mutant form of G*ß*,inhibit APP-dependent toxicity of A*ß*.These results suggest that A*ß*

toxicity in neurons requires interaction of APP with Go protein,and signaling is mediated by Gβγ subunit of Go protein.

Cellular and Molecular Neurobiology
Poster Number 41 | **Session 2**

"Characterization of ena/VASP domains with dominant negative function"

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Neurodegeneration is a process known to occur in metazoans. In previous work we found that the gene enabled (ena) is implicated in progressive neurodegeneration in Drosophila. Silencing ena ortholog genes (the ena/VASP family) in mouse hippocampal neurons triggered neurite retraction and concominant neuronal cell death through an apoptotic pathway. Since our ultimate goal is to confirm these results in a mouse model of late onset progressive neurodegeneration through deregulation of ena/VASP family members, we characterized potential dominant negative (DN) versions to identify the most effective one. Ena/VASP proteins share three well-defined domains, the EVH1 domain, a central proline-rich region, and EVH2. Overexpression of the DN (EVH1-GFP or EVH2-GFP) in mouse hipocampal neurons led to neurite retraction reminiscent of what had been observed with the RNAi pools. Thus, to begin to dissect potential signaling pathways we seek to define if a specific domain of ENA is responsible for the degenerative phenotype in Drosophila; transgenic fly lines expressing the EVH1 and EVH2 domains were generated. Preliminary results suggest that the functional inhibition of the EVH2 domain could be responsible for the behavioral phenotype observed in the original mutant.

Cellular and Molecular Neurobiology
Poster Number 42 | **Session 3**

"Novel signaling mechanisms and biological functions induced by the transmembrane protein Lrig1"

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Since discovery of Leucine-rich repeat and Ig-like domain (Lrig) mammalian proteins, they have been characterized as potent inhibitors of many receptor tyrosine kinases, such as Ret, Met and EGFR among others. However, given their structural features, it has been proposed that Lrig transmembrane proteins could influence neuronal development functioning as a cell-type specific adhesion molecule. Here, we show that Lrig1 is able to associate in a homophilic and heterophilic manner, and the presence of the LRR domain is required for these interactions. In neuronal cells expressing endogenous Lrig proteins, the addition of purified Lrig1ECD stimulates axonal growth and promotes the rapid activation of cytoplasmic tyrosine kinases known to participate in cytoeskeletal rearrangements. Together, these results provide an insight into Lrig1 function and establish a new mechanism of intercellular communication with potential relevance for neuronal development.

Cellular and Molecular Neurobiology
Poster Number 43 | Session 1

"The NPAS3 Gene Locus Contains the Highest Number of Non-Coding Accelerated Elements in the Human Genome"

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Using available datasets of genomic elements that show evidence of rapid evolution in the human lineage termed human accelerated elements (HAE) we uncovered the most accelerated region in the human genome. This region is located in chromosome 14 and contains 11 accelerated elements and includes the gene NPAS3. This gene is a transcription factor of the bHLH-PAS family that is broadly expressed in the developing mouse nervous system playing an important role in normal brain development and neurosignaling pathways. In addition, its dysfunction has been associated with schizophrenia in humans. The finding that this gene shows a set of highly conserved putative regulatory regions that evolved faster in the human lineage suggests that it might have acquired a new expression pattern in the human brain and probably a novel function. Using a transposon-based transgenic assay in zebrafish we tested the ability of NPAS3 elements to function as developmental enhancers. Our results indicated that 9 out of the 11

HAE are developmental enhancers. Additionally, we performed a comparative expression analysis over selected NPAS3 elements in transgenic mice. So far our results show that ortholog human and mouse sequences of two HAE display differences in expression patterns in transgenic mouse.

Cellular and Molecular Neurobiology
Poster Number 44 | Session 2

"Molecular evolution of calcium permeability of alpha9alpha10 nAChRs"

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Nicotinic cholinergic receptors (nAChRs) can be classified based on their ability to permeate Ca2+. The Ca2+ permeability of the mammalian a9a10 nAChR is among the highest (PCa/PNa~10), in accordance with its known function as the source of Ca2+ entry to cochlear hair cells and modulation of sound amplification (Elgoyhen et al., 2001). Surprisingly, we now report that the calcium permeability of chicken a9a10 nAChRs (PCa/PNa<2) is much lower than that of its mammalian counterpart. This may follow from the differential evolutionary history recently described for vertebrate $\alpha 9\alpha 10$ nAChRs (Franchini and Elgoyhen, 2006) and provides the opportunity to analyze the determinants of Ca2+ permeability by searching for the differences in the amino acid sequence that natural selection (and not arbitrary experimental mutagenesis) has fixed. Through the generation of chimeric receptors and site directed mutagenesis we show that residues located in the extracellular vestibule, the TM1-TM2 loop and the intracellular domain are determinants of the Ca2+ permeability of a9a10 nAChRs. These results indicate that the transmembrane pore lining TM2 domain is not the sole determinant of calcium permeability and that ions are selected along the entire conduction pathway.

Cellular and Molecular Neurobiology
Poster Number 45 | Session 3

"Evaluating the role of luminal calcium on intracellular calcium signals mediated by IP3 receptors"

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Calcium signals are used by almost all cell types to initiate a large variety of processes. Calcium release through IP3 receptors (IP3Rís) is a key component in many of these signals. IP3Rís are located on the membrane of the endoplasmic reticulum, usually forming clusters. The channel becomes open upon binding IP3 and calcium to a cytosolic site. There are also calcium binding sites on the luminal side of the reticulum. However, the way that luminal calcium regulates the kinetics of IP3Rís or the role it plays on the dynamics of the signals are unknown. In this work we have taken the first steps towards an evaluation of this role. To this end, we have elaborated a protocol to load the reticulum of Xenopus Laevis oocytes with a calcium dye. We show the success of the protocol by observing simultaneously the lumen (marking calcium) and the membrane of the ER (using a specific marker). We then show IP3 mediated signals as observed from the lumen and from the cytosol simultaneously using two dyes that are differentially contained in both regions and that fluoresce with different wavelengths. This opens up the possibility of getting a quantitative understanding of luminal calcium on IP3 mediated calcium signals.

Cellular and Molecular Neurobiology
Poster Number 46 | Session 1

"Comparative analysis of Delta-like 1 regulatory elements during brain development using transgenic mice"

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The gene Delta-like 1 (Dll1) has been implicated in the proliferation/differentiation switch of neural precursors during brain development. Dll1 encodes a ligand of Notch1 that inhibits neuronal precursors differentiation allowing them to proliferate increasing the undifferentiated progenitor pool. The expression of Dll1 is thighly

controlled during brain development but the regions and mechanisms controlling its expression are not completely known. Previous works have shown that a region located 4.3 kb upstrof Dll1 (4.3-Dll1) drives the expression of the reporter gene lacZ in a subdomain of the endogenous expression pattern. In addition, in our laboratory we have identified an additional region located at 5.9 kb of Dll1 (5.9-Dll1) that controls the expression of a reporter gene in an overlapping manner to the endogenous gene pattern. Using transgenic mice expressing fluorescent proteins under the control of 4.3-Dll1 and/or 5.9-Dll1 we have performed a detailed temporo-spatial study of coexpression and colocalization during mouse development in order to identify the unique properties of each regulatory region. Our data brings new information into the understanding of the regulation of Dll1 expression during brain development.

Cellular and Molecular Neurobiology
Poster Number 47 | Session 2

"Impact of mu-opioid receptor polymorphisms on neuronal calcium channels activity"

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The human mu-opioid receptor (hMOR) gene Single Nucleotide Polymorphism A118G/N40D (hMOR-D) has a high frequency of occurrence (~18%). Presence of hMOR-D in chronic pain patients is associated with lower morphine dose requirements. The molecular basis of this association is unknown. MOR activation inhibits presynaptic CaV2.2 calcium channels in nociceptors and, as a consequence, impairs synaptic transmission, reducing pain. Nociceptors are enriched in a CaV2.2 splicing isoform, named CaV2.2e37a, which has a distinct voltage independent sensitivity to MOR activation. Here, we hypothesized that hMOR-N or hMOR-D could differentially regulate CaV2.2e37a activity. To test this, we measured calcium currents in a heterologous system expressing either hMOR-N or hMOR-D and CaV2.2e37a. In dose-response curves for the MOR-agonist DAMGO, we found that hMOR-D has 4-fold higher potency and slightly higher efficiency to inhibit CaV2.2e37a currents, as compared to hMOR-N. Of note, the proportion of voltage dependent and voltage independent inhibition of CaV2.2e37a mediated by hMOR-N or hMOR-D was the same. Thus, we propose that the higher potency of hMOR-D on CaV2.2e37a contributes the lower morphine requirement in chronic pain patients.

Cellular and Molecular Neurobiology
Poster Number 48 | Session 3

"Modulation of the local translation at the synapse by distinct mRNA silencing foci"

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XRN1 is a ubiquitous RNA binding protein with 5í- 3¥ exoribonuclease activity. XRN1 concentrates in discrete foci, termed Processing Bodies (PBs). We found that in neurons, XRN1 doesnít colocalize with PBs, but form discrete structures associated to the postsynapse, that we call NPSAXs (Non PB Synapse-associated XRN1). The NPSAXs are dynamic; they increase in number and size when mRNAs are released from polysomes, and dissolve upon treatment with drugs that stabilizes polysomes. Moreover, the NPSAXs respond to distinct synaptic stimuli. NMDAR activation provokes an enhancement in NPSAX, whereas mGluR stimulation provokes their dissolution. We found that the stability of the NPSAXs upon synaptic stimulation correlates inversely with polysome integrity. We have previously shown that other synaptic mRNA silencing foci, termed S-foci respond with a distinct pattern. We conclude that the NPSAXs harbour mRNAs that are silenced upon NMDAR stimulation, whereas they release mRNA to allow their translation upon mGluR stimulation. These observations highlight the selective use of distinct mRNA silencing foci for the fine tuning of local protein synthesis at the synapse.

Cellular and Molecular Neurobiology
Poster Number 49 | Session 1

"Structural interactions of Alpha-synuclein with mitochondria"

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Parkinsonís disease (PD) is caracterized by loss of dopaminergic neurons in the

Substantia Nigra. Alpha-Synuclein (AS) is a 15 KDa protein expressed in central nervous system and is the major component of the protein aggregates in PD. It was proposed that in early steps of AS aggregation, toxic oligomeric species are formed that may lead to mitochondrial malfunction, fragmentation and apoptosis. The aim of this work is to characterize the interaction of AS with mitochondrial membranes in intact organelles. Mitochondria were isolated from SH-SY5Y cells and incubated with fluorescent labeled AS (0, 1 and 10μM). Outer membrane was revealed by immunofluorescence of TOM20. A combined study using confocal and widefield microscopy with deconvolution and 3D reconstruction, reveals that there is a different localization of AS dependent on the concentration. At 1 μM of AS the protein colocalizes with TOM20 showing an outer membrane distribution. When mitochondria were incubated with 10μM AS, it localizes in the interior of the organelle. STED microscopy shows that AS is distributed in clusters in the outer membrane, not revealed by the other tecniques. This results exhibit features on AS interaction with mitochondria that may help to explain the malfunctions observed in PD.

Cellular and Molecular Neurobiology
Poster Number 50 | **Session 2**

"Control of synaptic morphology and function by the Smaug1-Nanos1 translation regulation pathway"

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Local translation at the synapse is an important mechanism for synaptic plasticity. Smaug is a translational repressor initially identified in Drosophila. Fly Smaug regulates the stability and/or translation of hundreds of maternal mRNAs that contain specific motifs termed SRE. We have previously shown that mammalian Smaug1 represses the translation of reporter mRNAs with SRE motifs (Baez and Boccaccio, JBC 2005). Smaug1 forms granules containing silenced mRNAs located at the post-synapse (Baez et al, submitted). Here, we show that mammalian Smaug1 has an important effect on synapse morphology. Smaug1-depleted neurons provokes smaller and more numerous PSD95 synaptic clusters. We found less mushroom-shaped and more thin spines upon Smaug1 knockdown. In addition, Smaug1-depleted neurons respond defectively to a repetitive depolarizing stimulus, as indicated by a reduced induction of ARC, an early gene marker of activity. The mRNA encoding the translational repressor Nanos 1 has SREs and we found that

Smaug1 knockdown increased Nanos1 protein level. Moreover, a Smaug1/Nanos1 double KD partially revert the Smaug1-KD phenotype. Our results suggest that the Smaug1-Nanos1 pathway is an important mechanism for local mRNA regulation that affects synapse morphology and plasticity.

Cellular and Molecular Neurobiology
Poster Number 51 | **Session 3**

"Role of Protein Kinase D1 in neurotransmitter receptorsí sorting"

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Most of neuronal dendritic and axonal membrane protein sorting, a key process for establishment and maintenance of neuron polarity, occurs at the Trans-Golgi Network (TGN). Additionally, accurate localization of membrane neurotransmitter receptors is essential for transmission of neural impulse and for proper neuronal maturity and development. Metabotropic and ionotropic glutamate receptors contribute to synaptic plasticity, hence in learning and memory processes, and their transport towards their final localization - dendrites or axons - is still a complex and not well-characterized process. Protein Kinase D1 (PKD1) is a major component in membrane trafficking events; in neurons, PKD1 participates in dendritic membrane proteinsí sorting; in non-polarized cells, such as HeLa, this kinase regulates TGN vesicle fission. All these previous observations lead us to hypothesize that PKD1 regulates glutamate receptorsí sorting. In order to test our hypothesis, we have analyzed the involvement of PKD1 in metabotropic glutamate receptor 1 (mGluR1) sorting. Expression of kinase inactive PKD1 or its depletion by short harping PKD1, alter the intracellular trafficking and membrane delivery of mGluR1, an effect coupled with a significant reduction in the length of dendrite branches.

Cellular and Molecular Neurobiology
Poster Number 52 | **Session I**

"Functional evidence of rapid evolution of amniot alpha9alpha10 nAChRs"

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The a9a10 nicotinic acetylcholine receptor (nAChR) mediates efferent inhibition of vertebrate hair cells. They were the last nAChR subunits to be identified, due to the fact that they present distinct pharmacological and biophysical properties that set them apart from the rest of the nicotinic subunits. Previous work has shown profound differences in calcium permeability and channel gating properties between rat and chick a9a10 receptors. Here, we show that this interspecies variability is even more profound when performing a comparison with the Xenopus tropicalis (frog) a9a10 receptor. Desensitization, modulation by extracellular calcium and current-voltage relationships of the frog receptor differ significantly from those of amniote a9a10 receptors and closely resemble those of a7 (or a8) neuronal nAChRs. Moreover, in order to evaluate whether these differences in biophysical properties correlate with differences in pharmacology, we are performing a detailed pharmacological profiling of chick and frog a9a10 nAChRs. We conclude that a process of rapid functional evolution shaped the peculiar properties of amniot a9a10 nAChRs, setting them apart from other members of the nicotinic receptors family.

Cellular and Molecular Neurobiology
Poster Number 53 | Session 2

"Epigenetic regulation induced by prenatal stress in rat hippocampus"

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Experiences during early development like prenatal stress alter gene transcription throughout the lifespan. These experience-induced changes could occur through epigenetic modifications such as DNA methylation, histone modifications and microRNAs-regulation. Epigenetic mechanisms regulate synaptic plasticity and its deregulation has been related to mood disorders. Our aim is to study prenatal stress-induced epigenetic changes in gene expression in the model gene gpm6a that encodes the neuronal glycoprotein M6a. Real Time-PCR measures of gpm6a mRNA levels in 60-days-offspring hippocampus from stressed and control rats showed an increase in prenatally stressed animals compared with control ones. Preliminary results from direct sequencing of bisulphite-converted genomic DNA

of the same samples showed a differential gpm6a methylation status between prenatal stressed and control animals. We also evaluated the microRNA 133a as a regulator for gpm6a mRNA expression in microRNA-treated primary culture of hippocampal neurons. Real Time-PCR showed a 40% decrease in the mRNA levels in treated cells, indicating that mir133 might regulate gpm6a expression. Altogether these results suggest that diverse epigenetic mechanisms may control gpm6a expression.

Cellular and Molecular Neurobiology
Poster Number 54 | **Session 3**

"Blocking BDNF inhibits TrkB receptor phosphorylation and induces an increase in neuronal death after Status Epilepticus"

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Brain-Derived Neurotrophic Factor (BDNF) has been implicated in several aspects of adult hippocampus physiology. BDNF is initially synthesized as proBDNF and its interaction with his receptor TrkB has been implicated in neuronal survival, while proBDNF with p75ntr/sortilin in apoptosis. We previously showed that cell death induced by Status Epilepticus (SE) produce a decrease in TrkB membrane expression and a switch BDNF/TrkB to BDNF/p75ntr binding. We hypothesize that this phenomenon has a key role in the development of neuronal death.To test this we administrated unilaterally TrkB-Fc (a BDNF scavenger) in the CA1 region of hippocampus immediately after SE. Animals were sacrificed 24h later and TrkB and pTrkB levels were analyzed by Western Blot. Neuronal damage was assessed by FJB. We found that TrkB-Fc only blocks BDNF and not proBDNF, and therefore could prevent the decrease in the levels of TrkB while not in pTrkB levels. These results indicate that BDNF release is able to produce the decrease of its own TrkB receptor and in this scenario facilitate its binding with p75ntr/sortilina. These results strongly suggest that BDNF induce cell death only in the absent of TrkB signaling.

Cellular and Molecular Neurobiology
Poster Number 55 | **Session 1**

"Amyloid precursor protein delays functional integration of adult-born

hippocampal neurons"

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The amyloid precursor protein (APP) has been largely implicated in impairment of synaptic transmission and plasticity in animal models of Alzheimer¥s disease. We took advantage of adult neurogenesis to investigate the effects of cell-autonomous expression of beta-CTF, a cleavage product of APP, on functional and structural plasticity of hippocampal circuits in an otherwise healthy background. B-CTF was expressed together with a fluorescent reporter in neural progenitor cells of the adult mouse dentate gyrus by retroviral delivery. Morphological and functional connectivity of the neuronal progeny were analyzed after several days post infection (dpi). Expression of b-CTF induced a substantial reduction of glutamatergic afferent connectivity that was observed at 21 dpi but it was normalized by 35 days. This transient reduction in functional connectivity was paralleled by a decrease in dendritic length (with no changes in spine density) expressed at 21 but not 35 dpi. Thus, b-CTF appears to delay dendritic growth without altering synapse formation. Finally, similar defects in neuronal development were observed by retroviral expression of the non-amyloidogenic alpha-CTF. These results indicate that APP elicits a protracted dendritic development that is independent of Ab production.

Cellular and Molecular Neurobiology

Poster Number 56 | Session 2

"Microtubule stability and polarized insertion of the IGF-1 receptor in neuronal polarization"

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Two particularly early events that occur in neurons that have not exhibited a discernible axon (stage 2 of differentiation) are i) The segregation of activatable, membrane inserted IGF-1 receptors (IGF-1r) in one neurite; and ii) An increased microtubule stability in a single neurite. The experiments shown in this communication were designed to study a possible relationship between these two phenomena. We treated hippocampal pyramidal neurons in culture (stage 2 of

differentiation) with two drugs that can alter microtubule stability: i) Taxol (increases microtubule stability) and ii) Nocodazole (decreases microtubule stability). These cells were then challenged with IGF-1 to trigger polarized insertion of the IGF-1r. Our results indicated that in the neurons treated with Taxol, activatable (membrane inserted) IGF-1r were found in all the neurites in contrast with control cells which exhibited polarization of activatable IGF-1r to one neurite. In contrast, the cells treated with nocodazole did not exhibit activatable IGF-1r in any of the neurites. These results indicate that polarized insertion of IGF-1 receptor at the growth cone and shaft of a minor neurite, essential for neuronal polarization, depends on increased microtubule stability in one (most probably the same) neurite.

Cellular and Molecular Neurobiology
Poster Number57 | Session 3

"Ubiquitin Proteasome System (UPS) inhibition induce retrograde axonal transport reductions by impairing the endosomal-lysosomal pathway (ELP)"

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Alzheimer's disease(AD),accounts for 70% of dementias. Accumulation of aberrant proteins in synapses in AD suggest that proteins degradation might be impared. The two major routes of protein degradation are the ubiquitin-proteasome system(UPS) and the endo-Lysosome pathway(ELP). Consequently itís important to understand how UPS is delivered to synapses and the role of UPS activity in synaptic ELP regulation. To test for the axonal transport of UPS we performed live imaging experiments of a YFP proteasome subunit(α4-YFP)revealing processive anterograde and retrograde particulated movement. To test for the role of UPS activity in transport regulation we inhibited the proteasome and observed a selective decreased in retrograde transport. Double movies of α4 and Lysotracker suggest that retrograde moving UPS is associated with lysosomes. Subcellular fractionation in sucrose gradients after UPS inhibition revealed impairments in endosomal membranes. Moreover,UPS inhibition induced a decrease in retrograde axonal transport and lysosome axonal densities. Our results revealed that defects in UPS activity impairs the formation of endo-lysosomal vesicles and reduce lysosome retrograde transport suggesting a relevant crosstalk between UPS and ELP with important implications for AD.

Cellular and Molecular Neurobiology
Poster Number 58 | Session 1

"Myelin-associated glycoprotein modulates programmed cell death of motoneurons during development"

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Myelin associated glycoprotein (MAG) is a lectin present in the periaxonal layer of myelin that engages several axonal receptors, including Nogo-receptors (NgRs), to mediate its biological functions. It has been proposed that NgRs have a modulatory rol on programmed cell death (PCD) of motoneurons (MNs) dependent of the neurotrophin receptor P75NTR.. The aim of this study was to analyze a possible modulatory role of MAG on PCD of MNs. A time course study showed that early after birth Mag-null mice have a reduction in MNs count. Also Mag-null mice exhibit increased susceptibility in an in vivo model of PCD induced by a sciatic nerve crush. Interestingly pre-treatment with a soluble form of MAG (MAG-Fc) prevented MN apoptosis in this model. Studies using an in vitro model of P75NTR-dependent PCD on spinal cord organotypic cultures confirmed the protective role of MAG. Similarly MAG-Fc exerted a protective effect against PCD when tested in a MN-derived cell line with, opening the opportunity to study the signaling pathway associated with this effect. Overall these results demostrate a new role for MAG as protecting MNs from PCD. These results expand our knowledge of the nurture role of myelination on axons and at the same time open new possibilities for therapeutic intervention.

Cellular and Molecular Neurobiology
Poster Number 59 | Session 2

"Late neurogenic events in the developing mouse spinal cord: identification of a novel subset of embryonic V2 neurons"

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Despite the progress made in understanding the mechanisms in early cell type specification in the developing neural tube, late embryonic neurogenic events have not been deeply studied. In order to identify the fate of late-born cell types in the ventral spinal cord, we performed genetic lineage tracing in the mouse. We found a novel population of cells marked by the expression of the transcription factors Gata2/3 that are distinct to previously characterized interneurons. Our morphological and molecular analyses show that these cells are cerebrospinal fluid-contacting neurons (CSF-cN) of the central canal. BrdU birthdating experiments indicate that CSF-cN progenitors actively divide until ~E14, contrasting to the earlier generation of V2 interneurons (~E10). Additionally, positional analysis with respect to dorso-ventral patterning genes in combination with the analysis of 5 mutant mouse lines, suggest that CSF-cN derived from p2 ventral progenitors. Finally, while the transcription factor Foxn4 is required for the development of early-born V2 neurons, we found that differentiation of CSF-cN is unaffected in Foxn4 mutants. These results suggest that distinct genetic mechanisms govern the genesis of subsets of V2 cells, including CSF-cN, a novel "late-born" V2 population.

Cellular and Molecular Neurobiology
Poster Number 60 | Session 3

"Effect of Shiga toxin-producing E. coli (STEC) and LPS in the microvasculature of striatal brain mice"

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The central nervous system (CNS) is usually affected in patients that suffer from hemolytic uremic syndrome by STEC infection. In addition to Stx2, LPS from STEC secretion may be involved, and the neurovascular unit alteration in this pathology is poorly understood. The aim of this study was to determine whether Stx2 alters striatal brain microvasculature, and whether LPS exacerbates it. 30g NIH mice were subjected to endo venous (e.v.) sublethal administration of Stx2, Stx2+LPS, LPS or vehicle. The brains were processed and analyzed until day 7 by confocal microscope. Treated brains were subjected to Lectin histofluorescence from Lycopersicon esculentum to study endothelial profile. Confocal micrographs were analyzed by Image J. An increase in the number, area and histofluorescence intensity of microvesels were found in striatal brains of e.v. Stx2+LPS administration after 7

days, compared to Stx2, LPS or vehicle ($p<0,05$). Stx2 changed the microvasculature profile; LPS exacerbated the action of Stx2 and may play an important role at the blood-brain-barrier permeability in STEC pathologic events.

Cellular and Molecular Neurobiology
Poster Number 61 | **Session 1**

"Autofluorescent nanocrystals of chalcedony and silica polymorphs in human cerebellum from elderly patients"

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Chalcedony is a microcrystalline fibrous form of SiO2 and it is a product of biomineralization. This paper introduces the recent research advances on silica biomineralization of human cerebellum from elderly people, which mainly focus on analysis of the nano-structure and components of chalcedony. Chalcedony was identified by us, in the human cerebellum by using a polarized light microscope. Now, the identification of the chalcedony is documented, by using a Leica TCS - SP2 Laser Scanning Confocal Microscope. Autofluorescent chalcedony crystals were obtained and a topographic study is shown. In the molecular layer of the cerebellum, chalcedony occurred as a rhombohedral crystal. Chalcedony consists of nanoscale intergrowths of polymorphs: quartz and moganite. Polymorphs were detected in the 3-D image of the crystals, by using different ion lasers and a Leica software. Quartz crystal is rhombohedral in shape (trigonal) 2 micron in size and is in large proportion to moganite. Moganite crystal is pinacoid in shape (monoclinic) of 1 micron in size, and is in small quantity. This is the first time that chalcedony and polymorphs have been identified as 3-D auto-fluorescent crystals in the human cerebellar tissue.

Cellular and Molecular Neurobiology
Poster Number 62 | **Session 2**

"Optic tectum morphogenesis: A step-by-step model based on the temporal-spatial organization of the cell proliferation. Significance of deterministic and stochastic components subsumed in the spatial

organization"

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The present work is a computer-assisted analysis of the morphogenetic role of cell proliferation in the developing chick optic tectum (OT). Records of mitotic cells spatial co-ordinates are analyzed as stochastic point process by means of standardized methods of signal analyses. Signals derived from mitotic cells records subsume a) a deterministic component indicative of a long-range influence that install asymmetric distribution of mitotic cells and b) a stochastic component indicative of short-range anti-correlations between neighboring cells. The existence of zones of high mitotic density (ZHMD) with typical positions results in a typical pattern of differential planar expansion. A simple model explaining how the spatial and temporal organization of the neuroepithelial cells proliferation contributes to the optic tectum morphogenesis is presented: 1. A medial ZHMD (mZHMD) appears at the caudal zone. 2. The mZHMD expands cephalically forming the dorsal curvature and then duplicates into two bilateral ZHMDs (bZHMD). 3. The bZHMDs move towards the central region of each hemitectum. 4. The planar expansion of both bZHMD and the slow growth of dorsal midline produce a medial groove. 5. A relative caudal displacement of the bZHMDs produces a caudal curvature, the OT caudal pole.

Cellular and Molecular Neurobiology
Poster Number 63 | **Session 3**

"The role of Disialic acids in the evaluation of procedural memory"

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Alpha2,8-linked disialic acid (diSia) residues occur in several glycoproteins of mammalian brains. We previously demonstrated a constant expression of diSia in hippocampus, olfactory bulb and cortex, but, a gradual decrease in cerebellum from neonate to senile mice. Based on this and on the relationship of the cerebellum

with procedural learning, we decided to study if a relation exists between the expression of diSia and that process. Through RNA interference (RNAi) we inhibited in the brain the expression of ST8SiaIII, the enzyme responsible for diSia formation. We then evaluated the procedural memory on treated animals trough a T-Maze test. For this purpose, C57BL/6 neonate mice were treated with RNAi for ST8SiaIII into the ventricular cavity. Real-time PCR was used to confirm the reduction of the ST8SiaIII mRNA. Pst8siaIII-treated mice had low levels of ST8SiaIII mRNA 8 days after treatment, and showed a reversal to normal levels at older ages. On the T-Maze, treated mice presented a reduction of arm alternation in all ages compared to control being more evident in older ages. We can conclude that ST8SiaIII mRNA is necessary at early postnatal ages, as lower levels induce loss of procedural memory at premature ages.

Cellular and Molecular Neurobiology
Poster Number 64 | **Session 1**

"Characterization of a hypoxic-ischemic rat model by studying thioredoxins protein family and behavior"

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Brain damage resulting from perinatal cerebral hypoxia-ischemia is a major cause of acute mortality and chronic disability in infants and children. The production of reactive oxygen species (ROS) has been proposed as an important cause of neuronal death and consequently brain damage after ischemia-reperfusion. In the present work, we study the differential expression of the oxidoreductases of the thioredoxin family of proteins, and their role in the cellular response to oxidative stress in a modified model of perinatal hypoxic-ischemia developed by Vannucci (1981) and Levine (1960). In addition, to further characterize the model, we conducted a behavioral test battery including elevated plus maze (EPM), open field (OF), and Morris water maze (MWM). Rats subjected to hypoxia-ischemia showed reduced locomotion when exposed to OF and a deficit in the extinction of a spatial memory in the MWM. At a molecular level we have observed several remarkable

differences in both abundance and regional distribution of this family of proteins at different times. By means of Western Blot and light microscopy analysis, we have observed some remarkable differential expression of these proteins in the most vulnerable areas of the brain to hypoxia-ischemia.

Cellular and Molecular Neurobiology
Poster Number | 65 Session 2

"Sonic hedgehog response in photoreceptors in a model of spectral deprivation"

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Photoreceptor cells have a specialized primary cilium called the outer segment (OS). Mutations that affect OS generally lead to ciliopathies, which include retinal photoreceptor degeneration. Furthermore, macular degeneration is associated to "blue" light exposure, since blue light wavelengths impart the greatest risk of photochemical damage. The morphogen Sonic hedgehog (Shh) is essential for photoreceptor differentiation and retinal cell survival during embryonic development. Shh is expressed in the retina during postnatal growth but its function is unclear. With the aim to analyze the response of Shh by light and its function in the OS, we applied a spectral deprivation model. We found a gradual decrease of Shh protein when chickens were reared with long, medium or short wavelength lights compared with a white light group. Our results could indicate progressive retinal cell modifications both structural and functional in these cells. We suggest that Shh signaling may be required to maintain photoreceptors viability throughout life and that their levels are in part regulated by light.
UBACyT 20020090200198.

Cellular and Molecular Neurobiology
Poster Number 66 | Session 3

"An in vitro model to study inhibition of sensory and motor axon regeneration mediated by Guillain BarrË Syndrome-associated anti-glycan antibodies"

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Axon regeneration is a response of injured nerve cells that is critical for the restoration of structure and function after PNS or CNS injuries; this response is key to recover from numerous neurological disorders like acute immune neuropathy called Guillain Barré Syndrome (GBS). Some clinical studies associate the presence of anti- ganglioside antibodies (anti-Gg abs) with poor recovery in GBS. Patients with incomplete recovery have failure of nerve repair. We recently demonstrated in a passive transfer animal model that anti-Gg abs can halt axon regeneration. Defining the signaling pathways that prevent regeneration of injured axons can provide key insights to allow development of therapeutic approaches to enhance axon growth. For this reason, we developed an in vitro model of axon regeneration using organotypic co-cultures of spinal cord or dorsal root ganglion explants with peripheral nerve. Inhibition was produced by treating cultures with an anti-Gg mAb (GD1a/GT1b). Also, cultures were infected with VSV-G-pseudotyped lentivirus vector carrying the GFP sequence taking advantage of its greater trofism for neurons to keep track of regenerating axons on nerves. This model will be a useful tool to study the effect of anti-Gg Abs on the cytoskeleton of dystrophic growth cones.

Cellular and Molecular Neurobiology
Poster Number 67 | Session 1

"Unraveling synaptic NF-kappa B function. Localization during memory consolidation"

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NF-kappa B activation has recently been shown to be necessary for long-term memory consolidation and reconsolidation in the inhibitory avoidance learning in mouse. NF-kappa B is activated in the nucleus of hippocampal cells in a specific temporal window both in consolidation and reconsolidation. Previous results showed that the transcription factor is also present in synaptic terminals. More importantly we found NF-kappa B strongly bound to membranes of these terminals.

Some evidence has been found that, at this site, the transcription factor interacts with mRNA. Whether this affects the RNA¥s capability of being translated and if so, in what way, is yet to be discovered. This work focuses in the dynamics of its synaptic activation during consolidation and reconsolidation; discussing the novel localization of NF-kappa B in membranes of synaptic terminals and the possible role in local protein synthesis.

Cellular and Molecular Neurobiology
Poster Number 68 | Session 2

"Astrocytic subtype specification? Evidence from developmental genetics in the embryonic neural tube"

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In spite of the great progress made in the understanding of the ontogenetic mechanisms that control neuronal specification, much less is known about the origin of astrocytic subtype diversity. In this work, we begun to test the hypothesis that astrocytic subtypes specification follows similar logic that the acquisition of neuronal cell identity (dorso-ventral organization). We have focused on a small group of neural tube progenitors -termed p0- that express the transcription factor Dbx1, which are known to give rise to V0 neurons. In performing genetic cell fate mappings, we identified that these neuroepithelial cells contribute also to a group of laterally migrating cells. Marker expression analysis (NFIA, Sox9/2, GFAP) revealed that they are a subset of astrocytes that restrictively migrate along the spinal cord sulcus limitans. Interestingly, we found that in Dbx1 mutant mice, there is an increased number of p0-derived astrocytes, suggesting that Dbx1 directly or indirectly controls gliogenesis. We provide evidence that the genetic mechanism of Dbx1 involves modulation of Notch signaling pathway, by determining the class of Notch ligand (Delta/Jagged) that mediates cell-cell interactions controlling differentiation.

Cellular and Molecular Neurobiology
Poster Number 69 | Session 3

"Interaction of Estradiol and Notch/Neurogenin3 Signaling Pathway on Hypothalamic Neurons"

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Sexually segregated hypothalamic neurons respond differentially to the hormonal environment before the masculinizing actions of gonadal steroids. At embryonic stage 16, 17b-estradiol (E2) induces axonal growth only in males hypothalamic neurons. Recent studies have shown that E2 and Notch signaling converge to control neuritogenesis in hippocampal neurons. Activation of Notch is associated with an enhancement of the expression of the transcription factor Hes1 and Hes5, which negatively regulates the proneural gen Ngn3 that is involved in neuritic growth. Using qPCR we have detected that E14 hypothalamic neurons express Ngn3 in a sexually dimorphic pattern: females neuronal cultures had significantly higher Ngn3 mRNA levels than males. E2 significantly increased Ngn3 mRNA in males but not in females. This effect was blocked by the ERa/b mediated transcription antagonist ICI. In addition male neuronal cultures had significantly higher Hes1 mRNA levels than females and E2 significantly reduced Hes1 mRNA levels in males, but not in females. The effect of E2 on Hes1 was also blocked by ICI. These results suggest that E2 and Notch signalling pathway interact in hypothalamic neurons with a sexually simorphic pattern even before the organizational effect of gonadal steroids.

Cellular and Molecular Neurobiology
Poster Number 70 | Session 1

"Predicted nes and nls motifs affects subcellular localization of the ionic channel TRPM1"

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TRPM1, the founding member of the TRPM subfamily of Transient Receptor Potential (TRP) ion channels, was identified as a protein downregulated in highly metastatic melanoma cells. In alignment analysis of TRPM channels members we identified a sequence present in TRPM1 channel with no corresponding homologous region in any other family member. This sequence has been described as exon11. We thought that exon11 may be involved in channelís functionality. In silico analysis of this exon suggest the present of a nuclear export signal (NES). Other prediction analysis performed in TRPM1 show a sequence that could function as a nuclear localization sequence (NLS). We constructed TRPM1Δex11-GFP lacking exon11. When overexpressing TRPM1Δex11-GFP in HEK293T cells the amount of channel in the nuclear periphery is enhanced in contrast with TRPM1-GFP. To probe if NLS is an active signal in the channel we treated transfected HEK293T cells with TRPM1-GFP with actinomycin D, known for retard protein nuclear import. After the treatment we observed a typical distribution of GFP-tagged channel in the nucleus. These results suggest that TRPM1 possess functional NES and NLS motifs, how this motifs regulates the function of TRPM1 remain unknown

Cellular and Molecular Neurobiology
Poster Number 71 | **Session 2**

"Biomineralization by silica in subcellular fractions of electric tissue detected by using epifluorescence microscopy"

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Electric organs (EO) have been used for the study of cholinergic synapses. Chalcedony is a microcrystalline fibrous form of silica (SiO2) and it is a product of biomineralization. Chalcedony has been identified by us, in the electric organs from living fish by using a polarized light microscope (DMLP). In plane-polarized light, chalcedony is rounded in shape, 12ñ15 μm in size, translucent, with a low refraction index. The crossed-polarizer image shows first order birefringence color (grey white) and radial extinction. In this study, we will document the visualization and identification of the crystals of chalcedony in subcellular fractions obtained for differential centrifugation of the electric organs from Rajidae fish. Autofluorescent chalcedony was detected by using a Nikon Epifluorescent Microscope. The autofluorescent character of chalcedony (a mineral) allowed us to obtain images of the crystals, and a topographic study was recorded. All subcellular fractions

have autofluorescent chalcedony with different shape, size and intensity. These observations are in relation with neuronal cell death and synaptic terminal degeneration in Rajidae electric tissue.

Supported by grants to MPF from the SGCyT, UNS, Bahía Blanca, Argentina

Cellular and Molecular Neurobiology
Poster Number 72 | Session 3

"Delayed maturation of granule cells generated in the aging hippocampus"

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Neural progenitor cells of the adult hippocampus can differentiate and develop into fully functional neurons. Adult neurogenesis is tightly regulated by several physiological conditions, with aging being one important factor associated to decreased rates of neuronal production. In recent studies we have observed that the timing of neuronal maturation is regulated by the activity of the surrounding networks. In the present work we hypothesize that the aged dentate gyrus may present an environment that supports a slow rate of neuronal maturation. To test this hypothesis we injected a retrovirus expressing GFP in the dentate gyrus of five-month-old mice and analyzed the morphology and expression of neuronal markers at different ages of the newly born cells. Preliminary observations indicate that 21-day-old neurons in aged mice display very immature features when compared to neurons of the same age in young adult mice. We are now in the process of investigating whether such delayed maturation in the aged hippocampus is due to reduced levels of network activity, lack of secreted trophic factors, or cell intrinsic properties of aging neural stem cells.

Cellular and Molecular Neurobiology
Poster Number 73 | Session I

"Bone marrow mononuclear cells may provide an endogenous repair mechanism in the demyelinated sciatic nerve"

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In previous work, we have demonstrated the reorganization of major myelin proteins P0 and MBP and axonal protein PGP 9.5 in the demyelinated area, as well as the migration of CD34+ bone marrow mononuclear cells (BMMC) during Wallerian degeneration in ratsí sciatic nerves. Once in the demyelinated area, some BMMC conserve their phenotype while others change it to S100*ß*+, MBP+ and PGP9.5+. In the present work we evaluated prostaglandin (PG) participation in cell recruitment through Western blot and BMMC possible effect on remyelination through electron microscopy and immunohistochemistry. As regards PG analysis, the expression of Cox1 showed an increase 6 h after lesion, while Cox2 expression was induced 6 h after nerve injury and continued until 24 h, which suggests PG as one of the biological signals involved in BMMC recruitment. On the other hand, a reduction was observed in MBP and P0 clusters, as well as the presence of more myelinated axons in the crush area in BMMC-transplanted rats, although their myelin structure was still different from control axons. These data suggest BMMC beneficial effect on myelination either by preventing demyelination or stimulating remyelination. Yet, further experiments will be necessary to elucidate the mechanisms involved in BMMC migration.

Cellular and Molecular Neurobiology
Poster Number 74 | Session 2

"Acute effects of Pregabalin on the distribution and electrophysiological properties of the Cav2.1 calcium channels expressed in the HEK293t cell line"

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We established a cell model to study the acute effects of Pregabalin (PGB), a drug widely used in epilepsy and neuropathic pain, on calcium channels at the cellular level. HEK293t cells were transfected with plasmids coding for all subunits of the Cav2.1 channel. We used aα1 fused to eGFP tag, whereas in parallel experiments α2δ subunits were visualized through a pHluorin tag to follow the distribution

of the different subunits in time and at different experimental conditions. The expressed channel was functional as shown by increases in calcium currents of transfected cells measured by whole cell patch-clamp recordings, showing a maximum barium currents peak in the I-V curve at ~20 mV. The fluorescent signal showed a homogenous distribution; and acidification of the medium reduced the fluorescence when pHluorin- $\alpha 2\delta$ was expressed, which recovered after basification. Incubation of cells with 500 uM PGB for 30 minutes induced changes in localization of the subunits as measured by fluorescent time lapse microscopy, whereas no changes were visualized for cells expressing a control GFP protein. Together these results show strong evidence for an acute effect of PGB on calcium channels¥distribution and electrophysiological properties of the channel.

Cellular and Molecular Neurobiology
Poster Number 75 | **Session 3**

"A FRET analysis of Rho-A activity during the establishment of polarity in cultured hippocampal neurons"

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Studies over the last decade on the establishment of neuronal polarity have implicated as a key regulator of this process the small GTPase RhoA. In particular it has been shown that over-expression of RhoA halts neuronal polarization on hippocampal neurons whereas expression of dominant-negative RhoA results in multiple axon formation (Da Silva et al,2003) Stressing the importance of this protein, our laboratory has recently shown that the RhoA activator Lfc has a negative impact on the development of polarity (Conde et al, 2010) These observations clearly suggest that inhibition of RhoA activity is required for axon formation and neuronal polarization. All these studies, have assumed that RhoA inhibition should take place at the growth cone of the future axon, but this has never been proved so far. Using a RhoA biosensor and a FRET approach we have now obtained evidence about the spatial regulation of RhoA activity during polarization. These results provide evidence suggesting that decreased RhoA activity, spatially localized to the axon, parallels neuronal polarization. This event appears to be related with increased microtubule stabilization since low taxol doses (inducing multiple axon formation) are paralleled by decreased RhoA activity along axons.

• CHRONOBIOLOGY

Poster Number 76 | **Session 1**

"A novel nitrosomelatonin drug resynchronizes the circadian clock"

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Circadian rhythms modulate most physiological processes. The mammalian circadian clock is mainly synchronized by light through the retino-hypothalamic tract. Downstream, signal transduction is mediated through a calcium-dependent kinase- neuronal nitric oxide (NO) synthase-guanilyl cyclase pathway, in particular for the phase-advancing effects of light. We have employed a novel NO donor, nitroso melatonin (NOMel) to pharmacologically increase NO levels and therefore enhance photic effects on hamster locomotor activity circadian rhythms. Intraperitoneal administration of NOMel during a 6-hour phase advance of the 24 hour LD cycle significantly accelerated resynchronization to the new LD phase. In addition, NOMel generated a 1.5 fold increase in a subsaturating light pulse (LP)-induced phase advance during the late subjective night (circadian time 18), but did not interact with phase- delaying LP during the early subjective night. Also, preliminary evidences indicate an increase in nitrite levels after the administration of the drug followed by an advancing LP at circadian time 18. In conclusion, we show chronobiotic properties of the NOMel compound, and this work supports the importance of NO-mediated neurotransmission for circadian phase advances.

Chronobiology

Poster Number 77 | **Session 2**

"GAD1 and its potential relationship with NeuroD1 in the rat pineal gland"

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There much controversy regarding the existence and role of an intrinsic GABAergic system in the pineal gland. Microarray analysis of pineal glands from neonatal NeuroD1 KO mice identified Gad1 as the most up-regulated gene. Its product, the enzyme glutamic acid decarboxylase 1 (GAD1), converts glutamic acid into gamma-aminobutyric acid (GABA). In our laboratory, we are characterizing the ontogeny of this and other components of the gabaergic system and their relationship with the differentiation factor NeuroD1 in the rat pineal gland. The 67 kDa GAD1 protein was identified as a unique band in cytoplasmic pineal extracts via Western blot using a specific monoclonal antibody. GAD1 immunoreactivity was mainly observed in a few NeuroD1-negative astrocyte-like interstitial cells. The expression of GAD1 in pinealocytes is still under investigation, as its levels appear to be below the detection limits of standard IHC. The bilateral removal of the superior cervical ganglia did affect pineal GAD1 expression, and did increase the vimentin-positive cell population. Interestingly, a subpopulation of these glia-like cells was negative for nuclear NeuroD1. Further studies are being performed to elucidate the role of GABAergic innervation and intrinsic GABAergic cells in the pineal ontogeny.

Chronobiology
Poster Number 78 | Session 3

"Time waits for nobody: interaction between the circadian clock, dopamine and interval timing"

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Temporal perception is fundamental for environmental adaptation. Time estimation in the second-to-minutes range - known as interval timing - involves the interaction of the basal ganglia and the prefrontal cortex. In this work we tested the hypothesis that dopamine signaling is involved in the interaction between circadian and interval timing. Animals were trained following the peak-interval (PI) procedure. Results show significant differences in the estimation of 24-second intervals at different times of day, with higher accuracy at night. Interval timing was also studied in animals under constant light (LL) conditions, which abolish circadian activity and temperature rhythms. Mice under LL conditions were unable to acquire temporal control in the peak interval procedure. However, daily injections of L-DOPA before the experiment improved timing performance in LL mice, suggesting that an increase of dopamine is necessary for the interval to be

timed. We are currently studying circadian rhythms in clock gene expression in the mouse substancia nigra (SN), a target area for interval timing. Taken together, our results indicate that short-time estimation is modulated by the circadian clock, involving dopaminergic neurotransmission.

Chronobiology
Poster Number 79 | **Session 1**

"A circadian NeuroD1 protein as a potential Aanat gene regulator in the adult rat pineal gland"

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NeuroD1 is a transcription factor responsible for the establishment and maintenance of endocrine phenotypes. In the pineal gland, the Aanat gene encodes a key enzyme in melatonin synthesis, the arylalkylamine N-acetyltransferase. E-Box motifs identified in the Aanat gene promoter have been associated with its highly tissue-specific expression. In this work, we characterized NeuroD1 protein expression in adult rat pineal glands via IHC and WB. We also determined, via EMSA, whether an E-Box motif located upstream of the CRE-inverted CCAAT cassette in the Aanat promoter might recruit NeuroD1. NeuroD1-immunoreactivity was observed in pinealocytes and in a subpopulation of vimentin-positive astrocytes. Whereas at ZT14 NeuroD1 was mainly nuclear, at ZT6 there was a weaker signal in the cytoplasm. After SCGx, a heterogeneous pattern of subcellular NeuroD1-immunostaining was observed among pinealocytes at ZT14. These data were confirmed by WB. Aanat E-Box-containing probe was able to bind proteins from pineal nuclear extracts; NeuroD1 may form part of these complexes, according to supershift assays with an anti-NeuroD1 antibody. These results suggest that NeuroD1 protein dynamics are under circadian control and that NeuroD1 modulates Aanat gene expression in the adult rat pineal gland.

Chronobiology
Poster Number 80 | **Session 2**

"Circadian structural remodeling of the PDF circuit modulates locomotor activity in Drosophila"

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In our laboratory we are studying the relevance of specific neurons in the circadian control of behavior. The small ventral lateral neurons transmit time of day information releasing the neuropeptide pigment dispersing factor (PDF). Also, we demonstrated daily structural changes of axonal terminals of this circuit. Axonal arborizations show higher complexity during the day and a closed configuration during the night. Currently, we are analyzing which molecular mechanisms are responsible for this particular type of plasticity. Until now we have demonstrated the relevance of matrix metalloproteinases in the control of this form of plasticity. Moreover down-regulation of MMP1 in the PDF circuit alters circadian locomotor activity. On the other hand we are currently exploring the GRASP (GFP Reconstitution across Synaptic Partners) technology to map the synaptic contacts of the PDF circuit and have found some potential contacts which are going to be confirmed employing markers of pre and post-synaptic compartments. In addition, we are exploring if structural remodeling implies different synaptic partners throughout the day. In conclusion, MMPs are in part responsible for the daily changes in the axonal arborizations and this structural plasticity affects circadian behavior.

Chronobiology
Poster Number 81 | **Session 3**

"Stars of the show: Role of SCN astrocytes on immune-circadian interactions"

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Circadian rhythms in mammals are orchestrated by a master oscillator that resides in the hypothalamic suprachiasmatic nuclei (SCN) and drives a number of physiological and behavioral rhythms. Over the last years, several evidences have

emerged pointing towards a bidirectional communication between the immune and circadian systems. Astrocyte cells have a functional circadian clock, are capable of regulating neuronal physiology and also accomplish several immune functions in the central nervous system, which is why they appear as good candidates as mediators for immune-circadian communication. In this work, we investigated the response of SCN astrocytes to immune stimuli. We show that TNF-alpha can alter the rhythms of PER2 expression in SCN astrocytes, both in amplitude and phase. Furthermore, we explored the effects of conditioned media from immune-challenged SCN astrocytes on clock gene expression and found that these cells are able to modulate the clock trough the action of TNF-alpha. Our results support the idea of an important role of SCN astroglia in the interactions between the immune system and the circadian clock as well as giving further evidence to the participation of TNF-alpha as key component in the neuroimmune interactions occurring in the SCN.

Chronobiology
Poster Number 82 | Session 1

"Identification of fast neurotransmitter(s) in the central pacemaker of the Drosophila brain"

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The circadian clock is a temporal filter. Based on self-sustaining transcriptional negative feedback loops, it ultimately gives rise to circadian rhythms. Molecular oscillators are hierarchically organized as pacemakers and slaves. Circadian control of clock outputs ranges from the most immediate ones within pacemaker neurons to behaviors. In the Drosophila adult brain, the small ventral lateral neurons (sLNvs) constitute the central pacemaker. It comprises five cells, of which four rhythmically release the neuropeptide pigment dispersing factor (PDF). PDF triggers specific (i.e., resetting) signaling events in downstream targets. Aside from the PDF-containing dense core vesicles, the presence of small clear vesicles near synaptic output sites in the axonal terminals of the sLNvs suggests that additional fast neurotransmitter(s) (NT) could take part in this process. Still, there is no candidate NT in the sLNv cells as they are not immunoreactive either to several biogenic amines or to GABA. The goal of this work (which is still in progress) is to evidence the nature of this NT by downregulating the expression of NT re-uptakers in the PDF+ neurons and screening for changes in two clock outputs: daily structural

remodeling of PDF axonal projections and rhythmic locomotor activity

Chronobiology
Poster Number 83 | **Session 2**

"PDF activates a BMP retrograde signal to shape the architecture of a key circadian pacemaker circuit"

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The neuropeptide pigment-dispersing factor (PDF) is key to synchronize and support molecular oscillations within the circadian pacemaker groups in Drosophila. It is expressed in the lateral ventral neurons (LNvs), including the small-LNvs (sLNvs) and the large-LNvs (lLNvs). The sLNvs project to the dorsal protocerebrum, where other circadian clusters are located, and are indispensable for maintaining behavioral rhythmicity under constant conditions. PDF immunoreactivity cycles in these dorsal terminals. We have shown that the complexity of these arborizations change throughout the day in a circadian fashion. To understand this phenomenon and its relationship with PDF we evaluated the structural plasticity in a PDF null. Surprisingly, the projections of sLNvs showed a miss-routing phenotype. When we reduced PDF levels acutely at specific stages we discovered that PDF is necessary during larval stage 1 for the correct development of the sLNv. This phenotype is also present in PDF-receptor (PDFR) mutant and loss of PDFR in Tim+PDF- neurons is sufficient to cause the defect. Finally, we investigated the signaling pathway involved in this phenomenon. Expression of constitutively active receptors of the BMP signaling pathway in a PDFR-mutant context prevents the miss-routing phenotype.

Chronobiology
Poster Number 84 | **Session 3**

"P-bodies and stress granules are regulated by circadian clocks"

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Gene expression is modulated by clocks at different points, includding the posttranscriptional level. 5-20% of the mRNAs expressed in a given tissue show daily changes in their levels. Whereas transcriptional oscillations have been extensivelely analyzed, the circadian regulation of posttranscriptional events has been less studied. In the cytoplasm, messenger ribonucleoprotein (mRNP) complexes can assemble into cytoplasmic mRNP granules as P-bodies and stress granules (SG). These foci are involved in the regulation of mRNA decay and storage, as well as translation. We have analyzed whether these foci are circandianly regulated. NIH3T3 cells were synchronized by serum shock. SG and P-bodies were detected at different times by ICC with antibodies against eIF3 and GE-1 respectively. P-body per cell (number) showed fluctuations peaking 28 h after synchronization and reaching highest areas 14 h after the shock. The arsenite-induced SG also showed temporal changes through time. We have also analyzed the temporal expression of factors that could be responsible for the generation of these rhythms. We found some RNA-binding proteins whose transcripts exhibit temporal changes. Our results suggest that P-bodies and SGs, or a subpopulation of them, are regulated by circadian clocks.

Chronobiology
Poster Number 85 | **Session 1**

"Electrophysiological analysis of clock neurons in Drosophila melanogaster"

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The rotation of our planet generates cyclic environmental conditions to which organisms have adapted by developing an endogenous clock. This allows them to anticipate the daily changes in light and temperature to adjust their behaviors and physiology accordingly. These circadian rhythms have been extensively studied in the fruit fly where many clock genes that interlock through negative feedback loops and generate daily oscillations have been described. Clock genes are expressed in approximately 150 clock neurons of which a particular subset, the pigment dispersing factor expressing lateral neurons have been found to play a central role. Although the mechanisms that generate the molecular clock have been studied for decades, it is the electrical properties of neurons what dictates their role within a circuit. These properties depend on the type and quantity of ion channels. The recent development of brain preparations to perform recordings of lateral neurons

is starting to close a gap between the molecular and the electrophysiological properties of clock neurons. We will present preliminary electrophysiological data that reveals novel inputs to lateral neurons together with initial results of an ion channel RNA interference screen that impacts on circadian locomotor behavior.

Chronobiology
Poster Number 86 | Session 2

"TNF-alpha mediates the circadian response to lipopolysaccharide in the central nervous system"

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Although there is substantial information regarding the circadian modulation of immunological variables, little is known about the circadian effect of immune factors. Systemic low doses of endotoxin lipopolysaccharide (LPS) delivered at CT15 (Circadian Time 12: locomotor activity onset) induce phase-delays of locomotor rhythm in mice. Intracerebroventricular (icv) injections of interleukin (IL)-1b and tumor necrosis factor (TNF)-a, which are strongly stimulated by LPS, also induced phase-delays at CT15. Moreover, the icv inoculation of TNF-a, but not IL-1, receptor antagonists abolished the LPS circadian effect. TNF receptor I/p55 KO mice exhibited a longer free running period, but a similar activity pattern and light responses than controls. In addition, there was no phase shifting effect of LPS in TNFRKO mice (p=0.006), although in KO and WT mice LPS inhibited wheel-running activity. LPS-induced Per-1, but not c-Fos, immunoreactivity in the paraventricular hypothalamic nucleous (PVN) decreased in TNFRKO mice in comparison with WT mice (p=0.025). In conclusion, we found evidence of TNF-a dependence, through the TNFRI, in LPS-mediated circadian responses. Moreover, our result suggest a local, rather than peripheral, effect of TNF-a in the brain, acting on the PVN or SCN nuclei.

Chronobiology
Poster Number 87 | Session 3

"NO-cGMP-PKG pathway in the SCN: The leftovers"

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The NO/GC/cGMP/PKG pathway is essential for light sincronization of the circadian clock. PDEs are key regulators of intracellular cGMP concentrations. To study the regulation of cGMP levels in the hamster SCN, we have determined by RT-PCR the presence of several PDEs isoforms in this model. In hamsters receiving specific PDE5 inhibitors, reentrainment to a 6h phase-advance of the LD cycle took significantly shorter than controls, also elicited an increase in light-induced phase advances when injected 45 min before light stimulation at CT18, and increase the number of light-induced PER1 ir-cells at CT18. We have studied the role of nitric oxide (NO) in the intercellular communication within the SCN. Administration of the NO scavenger PTIO blocked photic phase advances and inhibited light-induced cFos and PER1-ir. In addition we found an inhibition in the non-parametric entrainment to 23h cycle in hamsters receiving a single dose of PTIO before light stimulation. Pharmacological inhibition of PDE5 affects photic entrainment,indicating a potential benefit for circadian disorders which require an increase in light signaling to the clock. These findings could serve as a basis for pharmacological treatment for optimizing circadian adaptation to environmental changes,including jet-lag.

• COGNITION, BEHAVIOR, AND MEMORY

Poster Number 88 | Session 1

"Introspective reports reveal explicit conscious strategies in visual processing in Autism Spectrum Disorders"

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Subjective confidence beliefs are ubiquitous in decision-making. Through a Partial Report Paradigm (PR) experiment, we explored whether subjects with Autism Spectrum Disorders (ASD) have an altered conscious access to visual stimuli. Subjects had to identify a cued letter in a crowded context, and had to quantify their degree of confidence in their decision on a continuous scale from 0% confidence (subject reporting a random decision) to 100% confidence. Results showed that while having a slightly worse objective performance than controls, the subjective beliefs of ASD subjects reflected their objective performance better. Specifically, ASD subjects presented more sensitivity (less "high confidence errors"). Control subjects, on the contrary, frequently reported high confidence when making errors, and vice-versa, low confidence when answering correctly , a hallmark of unconcious processing. This suggests that ASD subjects have an increased conscious access to visual stimuli, an idea in line with theories of enhanced visual perception of autism.

Cognition, Behavior, and Memory
Poster Number 89 | **Session 2**

"Towards a behavioral characterization of a transgenic model of TDP-43 proteionopathies"

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TAR DNA binding protein 43 (TDP-43) is a predominantly nuclear DNA/RNA binding protein that has been reported to regulate transcription, alternative splicing and RNA stability. It has recently been identified as a pathological hallmark of the neurodegenerative disorders amyotrophic lateral sclerosis (ALS) and fronto-temporal lobar degeneration (FTLD). We recently generated and characterized new animal models based on the conditional overexpression in the mouse forebrain of human wild-type TDP-43 protein or a cytoplasmically-localized form, and showed that these mice recapitulate key aspects of FTLD and ALS. However, the physiological role of TDP-43 in behavioral responses has not been investigated. Within this framework, we propose: 1) To evaluate if TDP-43 overexpression leads to cognitive deficits, studying the learning and memory performance of these transgenic mice using the inhibitory avoidance task. 2) To study other behavioral responses in these animals, including locomotor activity, anxiety, hyperactivity, and exploratory behaviors (performing rotarod and open field tests). The results from this project will allow us to shed light onto the physiological roles of TDP-

43 in the nervous system, and to address the pathogenic mechanisms underlying TDP-43 proteinopathies.

Cognition, Behavior, and Memory
Poster Number 90 | **Session 3**

"Cognitive training and neuronal and glial markers in the brain of non human primates (Macaca fascicularis)"

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We present initial results regarding the impact of intensive cognitive training on specific brain areas in a non human primate (Macaca fascicularis). Under the assumption that cognitive training produces preferential changes at the participating neural circuits, we are in process of evaluating the expression of molecular markers in selected brain regions. Immunohistochemical analysis is being performed in a group of monkeys that received cognitive training twice daily during three months, as detailed in a previous communication. We began a doble-blind analysis of the hippocampus of each animal using cell proliferation- and new neuronal markers (BrdU, doublecortin-DCX), as well as non neuronal markers (aquaporin 4, Ezrin, EAAT1, GFAP). We aimed at the possible association of neuronal and glial changes. We report here the quantification of the number of BrdU- and DCX labelled cells from the right hippocampus. Since double-blind condition has not been broken up yet, this report aims at analyzing correspondence between BrdU and DCX. Present results show a clear and opposite tendency between both markers. Support : Lic.B. Stuto (CONICET), Mrs.C. Juárez (CONICET). Fund. Quirno, Fund. Conectar, FONCYT, CONICET.

Cognition, Behavior, and Memory
Poster Number 91 | **Session 1**

"Early olfactory experiences within the honeybee hive have a long-lasting effect on recruitment"

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Honeybees are suitable models to study learning and communication interactions since olfactory cues predicting food can be learned while forager transfer spatial information via dance. Floral scent learning within this context can be an important component during recruitment and might even have long-term consequences on foraging decisions. However, bees can learn food scents even shortly after emergence and remember at foraging ages, for instance while following dances to search for information about feeding locations. We determined that dancers, which brought back the same scent that had circulated inside the hive 8 days ago are more followed by the early experienced bees than dancers carrying novel scents. Also, a higher proportion of early experienced bees arrived at the feeder scented with the familiar odor. Thus, olfactory experiences acquired as early as the first week of the adult lifespan are efficiently retained to bias later foraging-related behaviors at a long-term scale within the honeybee colonies.

Cognition, Behavior, and Memory
Poster Number 92 | Session 2

"Neural correlates of sensorimotor synchronization"

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Sensorimotor synchronization (SMS) is a form of referential behavior, restricted to human, in which an action is coordinated with a predictable external stimulus (visual or auditory). Finger tapping to a metronome is a paradigmatic task in SMS studies. In this task the subject is instructed to tap in synchrony with a periodic sequence of brief tones, and the time difference between each stimulus and its corresponding response (asynchrony) is recorded. Despite being a very simple task, finger tapping involves several systems like auditory and time perception, time comparison, error correction and motor execution. We make a first step towards the identification of the neural correlates of the SMS by recording high-density EEG event-related potentials and the concurrent behavioral asynchronies during a finger tapping task. Through Principal Component Analysis, we found components whose activations are locked to either stimulus or response occurrence. Interestingly, we found some components carrying information about the higher-level percept of asynchrony 70ms after the current stimulus-response pair, and the error correction process 200ms before the following stimulus-response pair. We

also found an asymmetry between early and late responses.

Cognition, Behavior, and Memory
Poster Number 93 | **Session 3**

"Pharmacological study of serotonin receptors in reward-directed behaviors"

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The medial Prefrontal Cortex (mPFC), one of the brain structures that controls reward-dependant behaviors, expresses the serotonin (5-HT) receptors 5-HT1A, 5-HT2A and 5-HT3. Here we studied the effects on the performance of rats in a operant conditioning task of: a) 5-HTP, a 5-HT precursor; b) Buspirone, 5-HT1A agonist; c) Risperidone, 5-HT2A antagonist; and d) Ondansetron, an 5-HT3 antagonist. The administration of 5-HTP (50 mg/kg) did not affect the percentage of responses but increased the latency time to respond. Buspirone (10 mg/kg) had a similar effect, i.e., no differences in responses and highest latency times. Risperidone (1 mg/kg) completely blocked the learning of the operant conditioning task, evidenced in both parameters. Finally, Ondansetron (2 mg/kg) significantly reduced the number of responses, as well as the latency times. The 5-HT exerts a mainly inhibitory effect on the activity of the pyramidal neurons of the mPFC, given to the presence of 5-HT1A receptors in the axon hillock. 5-HT can also inhibit pyramidal neurons indirectly through the activation of 5-HT2A and 5-HT3 receptors localized in GABAergic interneurons. Therefore, these results indicate that the serotonergic circuit comprising the mPFC is involved in the learning of a reward-dependent task.

Cognition, Behavior, and Memory
Poster Number 94 | **Session 1**

"Social Isolation during adolescence enhanced cocaine stimulant effects in youth rats"

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Functional brain differences between adolescents and adults, in a variety of species, may explain in part adolescentsí typical behaviors such as novelty seeking and their higher vulnerability to stressful situations. Adolescent rats show a higher level of social interaction with their kind around post natal day (PND) 28-35, and it has been shown that 5 days of social isolation during this period induced molecular changes in the prefrontal cortex that last until adulthood. In adult rats stress enhances cocaine stimulants effects; however it is not known if isolation during adolescence has long term effects on cocaine stimulant properties. Therefore, our main goal was to evaluate whether isolation during adolescence increases cocaine locomotor response in young rats. Male Wistar rats on PND 30 were individually housed for 5 days, while control rats remained group housed. Between PND 60 and 67 all rats received 4 cocaine injections and locomotor response was measured for 20 min. A dose response curve (0, 1, 5, 10 mg/kg ip) showed that social isolation enhances the induction of sensitization with the lower dose without altering the acute response. The data presented here supports the idea that isolation stress during adolescence increases cocaine vulnerability in youth rats.

Cognition, Behavior, and Memory
Poster Number 95 | Session 2

"A new vision on the retrieval and storage deficit interpretations of experimental amnesias: A deficit in the long-term memory expression to understand "amnesic" scopolamine effects"

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Traditionally, experimental amnesia was explained by storage or retrieval deficits. We present an experimental approach that offers a different perspective: positive modulation of memory expression during reconsolidation could be used to distinguish between reactivated but unexpressed memories and obliterated memories. We refer to the possibility that when a memory is retrieved, the respective neuronal trace is reactivated although this trace could not take control over behavior. In Chasmagnathus, it was described that the scopolamine (SCP) has an amnesic effect when is administered either immediately before or after training: the traditional interpretation is that SCP disrupt consolidation and thus

long-term memory (LTM) storage. Here, we tested if the SCP amnesic effect can be explained as a LTM-storage deficit or as a decrease in LTM-expression. Our results show that the amnesic effect induced by SCP is the consequence of a memory trace that is not expressed but it has indeed the potential of being reactivated and labilized. This study could be relevant to understand the role of muscarinic mechanisms in memory.

Cognition, Behavior, and Memory
Poster Number 96 | **Session 3**

"Exploring teaching skills in young children"

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While current neuroscience and psychology has provided a vast corpus of information about the learning brain, very little is known about the teaching brain and the psychological organization, development, and phenomenology of the ability to teach. Paradoxically, teaching (Caro, 1992) has been proposed as a cognitive skill unique to human culture (Gegerly, 2005). We propose that teaching is a natural ability that matures in the first years of life, without the need for formal education. In the present work, we began to investigate the spontaneous progression of the teaching capacities in young children and its relation to their own learning experience. We examined kidsí understanding of the correspondence between teaching and learning in games in which rule are learned based on inference and then explained to an adult. We also investigate meta-cognition, assessing the degree of awareness that they have of these mechanisms during development. Preliminary results suggest that children as young as three year old, capable to use screening-off information to learn the causal structure of biological events (Schulz, 2004), spontaneously transmit this knowledge to others, using well known ostensive cues which denote a pedagogical intention.

Cognition, Behavior, and Memory
Poster Number 97 | **Session 1**

"Role of SPARC on hippocampal function"

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It is estimated that about 10% of the world population suffers from depression at some point in their lives. However, genetic and environmental factors that contribute to this disorder are only beginning to be found. SPARC (secreted protein acidic and rich in cysteine) is an extracellular matrix protein highly expressed in the developing brain and in the adult hippocampus after deafferentation. However, its expression and function in the brain has only begun to be understood. We observed that SPARC KO mice present high levels of anxiety-related behaviors, while they show low levels of depression-related behaviors. By injecting an adenoviral vector that expresses SPARC in the dentate gyrus (DG) of SPARC KO mice, we rescued the depression-related, but not the anxiety-related, phenotype. We hypothesized that alterations in neuronal activity during development in SPARC KO mice results in long-term alterations in the structure and function of the DG, which determine the alterations observed in behavior. We evaluated whether neuronal activity was altered in the KO DG by studying c-fos expression. Moreover, we studied whether SPARC levels in the DG can modulate neurogenesis. Our results show a role of hippocampal SPARC in the expression of depression-related behaviors.

Cognition, Behavior, and Memory
Poster Number 98 | **Session 2**

"Study on the Influence of Body Schema in Reaching Decisions Using Proprioceptive Illusions"

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When trying to decide whether an object is reachable or not, the question of how the brain computes distances arises. Previous studies indicate that cortico-motor representations are used while solving this task (Coello, 2008). Furthermore, during the process of decision making, response times scale with manual rotations needed to perform the manipulation (Frak, 2001), suggesting that the cognitive process involved in this decision is motor imagery. We propose that humans categorize objects using dynamic sensory information of the body schema to simulate a reaching movement. In order to test this hypothesis, we employed a classic technique of vibration-induced proprioceptive illusions (Lackner, 1988) to create the perception of the index finger being elongated or shrunk. During the first stage of our project we studied illusions perceived through mechanical

vibration of muscles. Our results show good response to stimulation in about 45% of trials. In future experiments, participants will be shown dots in a horizontal screen while perceiving illusions of body deformation, and will be asked to respond if the dot is reachable without performing any movement. We predict that the distance threshold will change accordingly with the perceived finger length.

Cognition, Behavior, and Memory
Poster Number 99 | Session 3

"Reconsolidation or extinction? Thinking of GABA"

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Here we investigate the role of the GABAergic system over the different memory phases in an invertebrate memory model. We have found that post-training administration of muscimol disrupts, while post-weak-training administration of bicuculline facilitate the formation of fear memory. Likewise, the same effects are found if the drugs are administrated pre- or post-memory reactivation. These results suggest a role of GABA in the consolidation and reconsolidation of fear memory. The same pattern is found when the drugs are administered at the end of the extinction memory training. On the contrary, opposite effects were found when the same drugs were administrated before extinction memory training. That is, facilitative effects when administrating muscimol, and detrimental effects with bicuculline. It seems that the GABAergic system plays different roles on the acquisition and consolidation of extinction memory. We propose that an activation of the GABAergic neurotransmission is necessary for the acquisition, whereas a reduce neurotransmission is required for the consolidation of the extinction memory.

Poster Number 100 | **Session 1**

"Acute ghrelin administration reverses depressive-like behavior induced by bilateral olfactory bulbectomy in mice"

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Ghrelin (Ghr) is an orexigenic peptide produced in the stomach. Recent studies seem to indicate that metabolic signals as Ghr may interact with central nervous circuits regulating reward and mood. Thus, Lutter & col have suggested that Ghr could defend against depressive-like symptoms of chronic stress in a restricted food intake model(2008). The bilateral olfactory bulbectomy(OB) in rodents is an animal model that appears to fulfill many of the criteria necessary for depression study. Take in mind the possibility that Ghr could be an alternative therapeutic tool for the depression treatment, we study if the acute intracerebroventricular Ghr infusion (0.3 & 3.0 nmol/µl) reverts some of the depressive-like behaviors induced by OB in female mice. We have evaluated the immobility time in the tail suspension test, a test predictive of antidepressant activity. The OB animals treated with saline (OB-saline) presented an increase on immobility time in relation to sham (OB-saline:93.22 $\pm$ 8.92 vs Sham-saline:59.02 $\pm$ 6.31; $p \leq 0.05$), but the acute Ghr 3.0 nmol/µl infusion induced an decrease on immobility time in relation to OB-saline (OB-Ghr 3.0 nmol/µl:6.95 $\pm$ 1.25 vs OB-saline:93.22 $\pm$ 8.92; $p \leq 0.05$), indicating that Ghr could reverts the immobility response in OB.

Cognition, Behavior, and Memory
Poster Number 101 | **Session 2**

"Role of 5-HT2a serotonin receptor in processes of cognitive flexibility"

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Our ability to adjust our behavior to the dynamic changes of the environment is called cognitive flexibility. It is known that this mechanism, which allows an organism to adapt, requires an executive control system and is altered in certain mental disorders. Human and animal models suggest that different regions of the Pre Frontal Cortex play key roles in cognitive flexibility since they are main areas of integration of cognitive, motivational and emotional information. Deficits in cognitive flexibility are characteristic of many psychiatric disorders including schizophrenia, depression and obsessive-compulsive disorder. As some other executive functions that are affected in these cases, atypical antipsychotics tend to ameliorate the deficit. Together with the clinical data, there are pre clinical data that support a role of the serotonergic system in cognitive flexibility. 5-HT2aR has been the focus of attention since they are one of the most important post-synaptic receptor of serotonergic system and highly expressed in limbic system. One of the main problems addressing the role of 5-HT2aR in cognitive flexibility is the lack of specific drugs. We propose then, to study the role of 5-HT2AR in processes involving cognitive flexibility using a genetically modified mouse model.

Cognition, Behavior, and Memory
Poster Number 102 | Session 3

"Reactivate, labilize and strengthen memories without the need of conscious recollection: Study of the temporal dynamics of reconsolidation of human declarative memory"

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The idea that consolidated memories become immutable has been challenged. A reactivated memory may become labile and then susceptible to amnesic or strengthening agents. This process, known as reconsolidation, has become the inescapable evidence that memories are dynamic. Our recent results showed for the first time that during reconsolidation human declarative memory could be improved by a naturalistic mild stressor (cold pressor stress, CPS): we found that is possible to reactivate, labilize and strengthen unexpressed memories. We used a declarative memory paradigm consisting of paired associated cue-response syllables. Here, long term memory is reactivated either 6 or 21 days after training and tested one day later. Two different tests, cued recall and recognition, and

two types of modulators (CPS or glucose), were used to evaluate the temporal dynamics of this declarative memory. We found that, three weeks after learning, once memories have been forgotten (i.e. unexpressed), memory improvements can only be obtained by glucose as the modulator, but not CPS. Thus, we propose that the non conscious reactivation of apparently forgotten memories, during reconsolidation, can lead to the recovery of long-term memory expression.

Cognition, Behavior, and Memory
Poster Number 103 | Session 1

"Amnesia by muscarinic receptors blockade is prevented by previous exploration of an open field"

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Muscarinic cholinergic (MAChR) blockade, either extended or restricted to the hippocampus, before or after acquisition of inhibitory avoidance (IA) in the rat, causes anterograde or retrograde amnesia respectively, since there is no long term memory (LTM) expression 24 h later. Rats exposed to 1 or 2 open field (OF) sessions for 3 min, behaved as control animals after training with a mild footshock in a step-down IA task. Two OF sessions led to IA-LTM formation in spite of an extensive or restricted to the hippocampus MAChR blockade. It was reported that during and after OF exposure and reexposure, there was an increase in ACh release from both cerebral cortex and hippocampus; this could contribute to "prime the substrate" by lowering the synaptic threshold for synaptic plasticity, leading to LTM. In the frame of the "synaptic tagging and capture" hypothesis, plasticity-related proteins synthesized during the previous OF would facilitate synaptic plasticity for IA in the same structure. However, hippocampal protein synthesis inhibition by anysomicin, which induced IA anterograde amnesia (see Snitcofsky et al.), was also prevented by 2 OF exposures, strongly suggesting that other structure should be involved.

Cognition, Behavior, and Memory

Poster Number 104 | **Session 2**

"Response to reward change in a model of schizophrenia like behavior produced by postweaning isolation"

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Isolation rearing from weaning produces long-term changes in behaviour including neophobia, impaired sensorimotor gating, social withdrawal and cognitive inflexibility, which cover the domains affected in schizophrenia. Our aim was to investigate the effect of early isolation (EI) has on the response to the reward change in adult rats. Animals had access to different concentrations of sucrose solutions, and goal tracking time was measured. In the Exp 1 we studied the impact of the EI (post natal day 21-36) on the partial reinforcement effect on the reinforcement devaluation. The EI altered neither the response to incentive devaluation, nor the partial reinforcement effect on the consummatory suppression, but EI animals showed an increase in consumption when the reinforcement was suddenly increased. In Exp 2 we evaluated animals in consummatory successive positive contrast and consummtory extinction (cE). Isolated subjects showed a more lasting positive contrast effect, but animals of both conditions did not differ in cE. These results suggest that EI do not produce major frustration reactions but express an increased euphoria, which is consistent with evidences of a major sensitivity to apetitive rewards and dopaminergic hyperactivity in mesolimbic system of isolated rats.

Cognition, Behavior, and Memory
Poster Number 105 | **Session 3**

"Social Isolation during adolescence enhanced cocaine rewarding properties in youth rats"

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Functional brain differences between adolescents and adults, in a variety of species, may explain in part adolescentsí typical behaviors such as novelty seeking and their higher vulnerability to stressful situations. Adolescent rats show a higher level of social interaction with their kind around post natal day (PND) 28-35, and it

has been shown that 5 days of social isolation during this period induced molecular changes in the prefrontal cortex that lasted until adulthood. In adult rats stress enhances psychostimulants rewarding effects; however it is not known if isolation during adolescence has long term effects on cocaine rewarding properties. Therefore, our main goal was to evaluate whether isolation during adolescence increases cocaine rewarding properties in young rats and cocaine relapse in adult rats. Male Wistar rats on PND 30 were individually housed for 5 days, while control rats remained group housed. Starting at PND 59 all rats underwent Conditioned Place Preference (CPP), and cocaine induced relapse (CR) 30 days later. A dose response curve (0, 1, 5, 10 mg/kg ip) showed that social isolation enhances cocaine CPP at 1 mg/kg and CR at 5mg/kg. These data support the idea that isolation stress in adolescence has long term effects on cocaine rewarding properties.

Cognition, Behavior, and Memory
Poster Number 106 | Session 1

"Participation of the mirror and mentalizing systems during the brain processing of different intentional actions"

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Several studies have suggested that intentions are recognized by the mirror-neuron system (MNS). Others emphasize that it is a "theory of mind" process, which activates areas outside the motor system (mentalizing). In the present study we examined brain activation of 20 healthy subjects by functional magnetic resonance imaging while both watching actions from videos and selecting the right goal from a list. The videos showed actions with simple direct goals, indirect or social interaction goals. We found that activity in the medial frontal cortex increased with the complexity level of the action during both observation and selection tasks. Activity in the temporoparietal junction was similar for all intentions during observation and greater for the complex goals during selection. Activity in the inferior frontal gyrus was similar for all tasks and videos, while inferior parietal lobule was greater for simple goals. All conditions and tasks activated MNS areas differently according to the intention category, while mentalizing areas were activated mainly in the last two categories. Activity was different according to

whether subjects were passively watching or explicitly thinking in the intention.

Cognition, Behavior, and Memory
Poster Number 107 | Session 2

"Ultrasonic Vocalization (USV) emission in infant rats, as a function of pre- and postnatal ethanol exposure"

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Previous studies indicate that infant rats emit higher levels of USVs after interacting with an ethanol-intoxicated dam. Prenatal experience with the drug also affects sensory recognition of ethanol-related cues which are likely to be present when the intoxicated dam interacts with its pups. In this study pregnant females received either 0 or 2 g/kg ethanol during gestational days 17-20. During postnatal day 3 (PD3), females were treated with either 0 or 2.5 g/kg ethanol and were later allowed to interact with their litter. Pups USVs were recorded 13 days later under the presence of alcohol odor or no odor. Ethanol olfactory cues depressed USVs. Nevertheless, pups that experienced alcohol in utero followed by an interaction with an intoxicated mother, showed very high levels of vocalizations which have been related with a negative emotional state. According to prior studies it appears that prenatal ethanol exposure sensitizes infantile perception of ethanol odor. In turn, and in accordance with the present results, this sensitization allows the establishment of stress-related memories when pups perceive ethanol-sensory cues derived from the intoxicated mother in conjunction with alterations in maternal care or nutrition.

Cognition, Behavior, and Memory
Poster Number 108 | Session 3

"Use of extra-maze cues for spatial learning in the toad Rhinella arenarum"

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Spatial orientation ability is a limiting factor for survival. Finding the right place to hide or where to find food can be the difference between living and dying. We, as mammals, orientate ourselves in space using multiple environmental cues of the surrounding world, and we set spatial relationships between them. Amphibians also have the capacity of spatial orientation, but it is still unknown how they perform this behavior. Ours previous studies showed that toads can use intra-maze visual cues to orientate. The goal of the present work is to determine if they can use extra-maze cues and what is the relevance they have in the orientation process. We trained toads (Rhinella arenarum) in a water finding orientation task using a transparent open field (to provide access to the context visual cues of the training room). After acquisition it was tested the relevance of the visual cues by hiding them with an opaque curtain. Tests revealed that animals use extra-maze visual cues to find the reinforcer inside the open field and that cues near the reinforcer are more relevant for orientation than those located far. Ours results show that amphibians are capable of using visual environmental cues for orientation, being cues near the goal more relevant to reach the spatial goal.

Cognition, Behavior, and Memory
Poster Number 109 | Session 1

"Effects of MK 801 on plasma corticosterone levels in stress-induced reinstatement of cocaine-conditioned animals"

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Previous results from our lab showed that MK 801 blocked the stress-induced reinstatement in an extinguished cocaine-induced conditioned place preference (CPP) in rats. In the present experiments, our goal was to determine if corticosterone, the principal hormone released in response to stress, could influence MK 801 effect on 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. On the reinstatement day, animals were injected with MK 801 (0.1 mg/kg i.p.) or vehicle, and subsequently assigned to two groups according to the following treatments: 1) Stressed animals (SA): 30 min-restraint exposure, and 2) Control animals (CA): left undisturbed in their home cages. All groups were then tested in the CPP,and their blood collected for plasma corticosterone determination. Results demonstrate that, although

MK 801 enhanced plasma corticosterone levels in SA and CA, non-significant differences were evident between both groups. These results show that neither the stress-induced reinstatement nor its blockage by MK 801 are dependent on the corticosterone response.

Financial Support: FONCYT, MinCyT, CONICET, SECyT

Cognition, Behavior, and Memory
Poster Number 110 | Session 2

"Differential changes in salivary markers of autonomic activity in response to elite competition"

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We investigated the response of salivary total protein (TP), alpha-amylase (sAA) and chromogranin A (CgA) to sporting competition and their relation with positive and negative affect. Eleven professional swimmers were examined during the first day of a national contest and on a recreated event that matched time-of-the-day and day-of-the-week assessments two weeks later. TP was determined by the Bradford method and sAA and CgA by western blotting upon awakening, 30 and 60 min post awakening, immediately before warming up for competition and 5, 20 and 60 min after competition. Psychometric instruments included the Positive Affect and Negative Affect Schedule - X. The concentrations of TP, sAA and CgA differed from controls only prior to and 5 min after the event. We observed associations between higher negative affect scores with higher levels of TP, sAA and CgA prior to the event on the competition day. Areas under the curve did not differ from controls for TP, sAA or CgA. TP and CgA showed a similar reactivity to sporting competition than sAA, which may be attributed to the mechanisms responsible for protein secretion into saliva. Strong adverse psychological stimuli only overrides the regular rhythm of salivary proteins moments before and after stressful situations.

Cognition, Behavior, and Memory

Poster Number 111 | Session 3

"Development of an Adeno-Associated Virus (AAV)-based model to study TDP-43 pathophysiology in rodents"

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TAR DNA-binding protein 43 (TDP-43) is a protein involved in diverse cellular functions and was recently identified as a major disease protein in frontotemporal lobar degeneration with ubiquitin-positive inclusions and amyotrophic lateral sclerosis. To study the pathophysiological role of TDP-43 in vivo, we developed a novel model using Adeno-associated virus (AAV)-based vectors to express wild-type TDP-43, a carboxyl-terminal fragment or TDP-43 variants with cytoplasmic or nuclear localization. This project aims to evaluate in the rodent brain whether overexpression of different TDP-43 species leads to disease-related pathological features (including TDP-43 aggregation, ubiquitination, hyperphosphorylation and cleavage). Our goal is to analyze if expression of any of these TDP-43 species results in neurodegeneration, and to study the potential effect of hippocampal or cortical TDP-43 overexpression in cognitive function (i.e. learning and memory) by behavioral analysis but also at biochemical level by assessing changes in plasticity-related pathways. The results of this project might provide more knowledge regarding the pathogenic mechanisms underlying TDP-43 proteinopathies, which hopefully will help the development of new and more effective therapies for these disorders.

Cognition, Behavior, and Memory

Poster Number 112 | Session 1

"The role of retrosplenial cortex and the molecular mechanisms involved in memory persistence"

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The persistence is a key characteristic of memory storage, but little is known about

the mechanisms and structures involved in maintenance of the memory trace. Although the hippocampus is crucial in the formation of new declarative memories, other brain regions probably mediate permanent storage of remote memories. It is known that the retrosplenial cortex is involved in spatial tasks and it has direct and indirect connections with the hippocampus; for this reason, we decided to investigate the role of retrosplenial cortex and the molecular mechanisms involved in memory processing. In this work, we show that inhibition of transcription in the retrosplenial cortex impairs memory formation around and a few hours later after inhibitory avoidance (IA) training. Moreover, translation is required in the retrosplenial cortex around training for memory formation and, late after training for memory persistence. In addition, long-lasting but not short-lived IA long-term memory depends on a delayed expression of c-Fos in the retrosplenial cortex. Our results support the hypothesis that recurrent rounds of consolidation-like events (i.e. transcription and translation) take place late after learning in the dorsal retrosplenial cortex to maintain memories.

Cognition, Behavior, and Memory
Poster Number 113 | Session 2

"Sensory Stimulus Categorization in Autistic Children"

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The Autistic Spectrum Disorder is a Neurodevelopmental Disorder that affects the domains of socializing, communication and behavior of those who suffer it. A frequent hypothesis about Autism is that people with this disorder present a diminished categorization capability, understood as the process by which different beings can be grouped within a given category and be considered as equals in a given context.In this work the perceptual characteristics and, in particular, the differences in categorization capability of children within the Autistic Spectrum Disorder are studied. To that purpose, a computer game consisting of a mono-parametric visual memory task was designed. This game was presented to three groups of players (Autistic children, children with Asperger's Syndrome, and a control group) in the range of 8- to 16-year olds in Bariloche, Rosario and Bahía Blanca. From each child an error distribution and a set of parameters were obtained that then allow to statistically infer if the child has used a categorical strategy to solve the task.

Cognition, Behavior, and Memory

Poster Number 114 | Session 3

"A study on rat vocal interactions and syntax"

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Rats and mice produce vocalizations that span the frequency range from the sonic to the ultrasonic. Above 40 kHz, rats produce a rich repertoire of vocalizations spanning a wide range of frequencies and duration. Semantic studies on these ultrasonic vocalizations (USVs) have shown a correlation with positive situations although the fine semantics of call types remains elusive. Syntax remains unexplored in rats. We have developed a setup for recording USVs from pairs of rats. Individual calls are recorded from two directional microphones hanging above and can be assigned unambiguously to each rat by their difference in power. We have developed a full set of algorithms to automatically detect, assign and classify the calls. Video tracking is also implemented to correlate vocal to other behaviors. This simple behavioral arena promotes high rates of USV production, averaging up to 2 calls per second per rat. We present analysis on both the sequencing of calls within single rat speeches and dynamics that emerge from their vocal interactions. The rat is a potentially powerful animal model to study the neurobiology of speech production and perception given their highly social ethology and amenability to physiological and behavioral studies in the lab.

Cognition, Behavior, and Memory
Poster Number 115 | Session 1

"Allopregnanolone prevents memory impairment through hippocampal serotonergic system"

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Learning and memory processes may be influenced by estrogen (E) and progesterone (P) and neurosteriods, such as Allopregnanolone (Allo). In previous work, we demonstrated that Allo improves memory in OVX female rats infused (s.c) with E and P on inhibitory avoidance task (IA task). Serotonine (5HT) is a neurotransmitter related with memory processes. Here we investigated the effect

of Allo over 5HT release in hippocampus. K+ evoked [3H]-5HT release of dorsal hippocampus slices of the different groups was carried out by superfusion method. Different treatments were: control (OVX-EP-Vehicle), OVX-EP-Allo and OVX-EP-Allo-8OHDPAT (5TH agonist). All groups were trained in IA task and, after 24-h intertrial delay; SD (step down) latency was tested. Results were analyzed by ANOVA 1 and Turkeyís post-test (p less than 0.05 was significant). In OVX-EP-Allo group the increased on latencies were related with a lower 5HT release compared with control. Intrahippocampal infusion of 8OH-DPAT prior to Allo reversed the increase of latencies and the decrease on 5TH release observed before. These data suggest that Allo would have a negative neuromodulatory action on hippocampal 5HT system that could explain the positive mnemonic effects on E-P primed rats.

Cognition, Behavior, and Memory
Poster Number 116 | Session 2

"Brain electrical activity in stroke patients under transcraneal magnetic stimulation therapy"

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Over the past years the repetitiverTranscraneal magnetic stimulation (rTMS) has been used for the investigation and therapy in several pathologies, including stroke. The effects of 1 Hz (rTMS) sessions on electroencephalographic activity are not fully studied. A double-blind prospective and longitudinal study was carried out to assess the electrical brain activity and to evaluate the clinical evolution in a sample of 9 subjects with chronic stroke after rehabilitation and the application of low frequencies rTMS (1Hz). The sample in study was randomly divided into two groups: 5 patients received sham rTMS stimulation (group I) and 4 patients received real rTMS (1Hz) (group II) both with daily sessions for 20 days. Electroencephalograms (EEG) were recorded before and after rTMS. The neurophysiological measures used were the resting EEG power spectrum (EEGPS), the Delta/Alfa ratio (DAR), the spike-frecuency and the spike- mplitude. Clinical characterization was assessed using Scandinavian (SS) and Barthel Index scales (BI). Sham rTMS group showed a significant overall stroke hemisphere increase ($p<0.05$) in Alpha power spectra, whereas effects on the power of the theta EEG bands significantly decreased ($p <0.05$). 1 Hz rTMS caused a tendency toward increase ($p=0.06$) in the Alpha band

power spectra in both brain hemispheres. There was also a decreasing tendency of the Delta band power spectra in both brain hemispheres. DAR diminished 23 % more in the 1 Hz rTMS group than in the sham rTMS group, and the spike-frecuency also increased in 1 Hz rTMS group after stimulations. When evaluating the clinical Scales after the rTMS, a tendency toward the increase of punctuations in the SS (p=0.06) was present in both groups of patients, being higher in the 1 Hz rTMS group than in the sham rTMS group. Stroke patients who received 1 Hz rTMS sessions experienced modifications on resting EEGPS, suggesting a propensity to the cortical activation in both brain hemispheres. 1Hz rTMS sessions on stroke patients cause an increment tendency of cortical excitability. The 1 Hz rTMS group had a better clinical recovery and of the brain electrical activity, both reflected in the modifications of the SS and in the DAR.

Cognition, Behavior, and Memory
Poster Number 117 | Session 3

"Serotonin and feeding regulation in ants: an immunohistochemical analysis"

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Serotonin (5-HT) plays an important role in the control and integration processes involved in feeding modulation in both vertebrates and invertebrates. Pharmacological studies have demonstrated that this amine promotes anorexic effects on different insects. In the nectar-feeding ant Camponotus mus, 5-HT depresses feeding by reducing the efficiency per contraction of the sucking-pump muscles. In order to study the neuronal pathway relating 5-HT, feeding structures and its modulation, we analysed the presence of serotonergic neurons in the main ganglia involved in feeding. Furthermore, we studied 5-HT association with the sucking-pump muscles and the alimentary canal of these ants. Immunohistochemical studies all along the alimentary canal revealed 5-HT-like immunoreactive processes on the foregut (esophagus, crop and proventriculus), while the midgut and hindgut lacked 5-HT innervation. Although both the frontal and the subesophageal ganglion contained 5-HT immunoreactive cell bodies, direct serotonergic innervation in the sucking-pump muscles was absent. Altogether, the results indicate that 5-HT plays an important role in the regulation of food transport throughout the gut in ants and acts most likely via indirect regulation of

the sucking pump activity.

Cognition, Behavior, and Memory
Poster Number 118 | Session 1

"Epigenetic mechanisms in object recognition memory"

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Long-term memory (LTM) consolidation requires mRNA and de novo protein synthesis. Transcription is controlled by transcription factors, their cofactors and repressors. Some cofactors can regulate gene expression by chemically modifying histones. Acetylation is the most studied histone modification related to gene expression regulation. This process is regulated by lysine acetylases (HATs) and deacetylases (HDACs). Here, we found evidence supporting that histone acetylation is involved in consolidation of novel object recognition memory in mice. We showed that hippocampal HDACs inhibition enhanced recognition memory. We found that standard and strong trainings produced LTM at 24 hours testing. However, strong trained mice showed significant higher levels of object discrimination than standard trained mice when we tested one week after training. Accordingly, only a strong training induced a general increase of hippocampal H3 acetylation 1 h after training. In order to evaluate the level of H3 acetylation in a particular gene, we are currently performing ChIP assay on the promoter regions of zif268 and BDNF gene. Based on our results, we hypothesize that histone acetylation is a key mechanism in LTM persistence, functioning as a molecular feature of stronger memories.

Cognition, Behavior, and Memory
Poster Number 119 | Session 2

"Molecular basis of ß-amyloid efect on memory formation in a triple transgenic mouse model"

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Beta-amyloid peptide (*ß*A) is the main component of neuritic plaques found in brains from Alzheimer Disease (AD) patients, clinically featured by cognitive impairment and progressive memory loss. *ß*A activates memory-involved signaling pathways, such as ERK/MAPK, among others. These effects have shown to be dependent on the type of peptide, its concentration, time of exposition and its aggregation state. However the nature of the mnesic effect is still unclear. To study neurobiological mechanisms related to initial states of AD, in which a subtle deregulation of the physiologic function of *ß*A can be inferred, we evaluated triple-transgenic (3xTg) mice memory in novel object recognition (NOR) task. 3xTg mice developed memory deficits between 3 and 6 months of age in correlation with ERK/MAPK activation specifically in prefrontal cortex (PFC). Here, we studied the correlation between MAPKs activation, *ß*A effect on memory formation and the aggregation level of the peptide. We found extra-nuclear ERK/MAPK activation at 3 months and nuclear activation at 6 months of age in PFC, but no activation in hippocampus. The results support that the memory deficit found in 3xTg mice is dependent, at least in part, on *ß*A accumulation and aggregation in prefrontal cortex.

Cognition, Behavior, and Memory
Poster Number 120 | Session 3

"Do psychostimulant drugs really have aversive properties?"

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Psychostimulant drugs induce appetitive and aversive learning in rats. According to some authors the appetitive effects are more likely to be associated with contextual cues, while the aversive effects have been consistently found in taste aversion learning. Parker claimed that rats avoid any taste that predicts a change in their homeostasis because this specie cannot vomit. The goal of the present study was to assess Parkerís hypothesis employing an odor learning preparation in infant rats (PD15-17). Two conditioning trials were employed in which subjects received contingent presentations of a novel odor (almond) and an emetic (LiCl) or psychostimulant (amphetamine) drug. The hedonic value of the almond odor was tested by means of a consumption test or in terms of locomotor avoidance. Rats showed odor avoidance in both tests when the almond odor was previously paired

with LiCl. However, subjects trained with amphetamine showed odor avoidance when they were evaluated in the consumption test, but odor preference in the locomotor activity test. These results represent an additional evidence for Parker`s hypothesis, showing that a given CS that predicts the effects of amphetamine can elicit approaching behaviors or avoidance depending on the modality of the test.

Cognition, Behavior, and Memory
Poster Number 121 | Session 1

"Qualitative study of equivalence equivalence relations and cognitive functioning in multiple sclerosis"

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Multiple Sclerosis (MS) patients show a pattern of neuropsychological dysfunction, with deficits in attention, long term memory, working memory, analogical reasoning and executive functions. These cognitive abilities have shown correlations with the formation of "equivalence relations" (ER) in previous studies. The ER paradigm has been extended to the study of the analogical reasoning ability, by testing equivalence - equivalence relations (EER). The aim of the present study is to evaluate MS patients with a single case methodology, in order to explore the performance using an EER task and a conventional analogical reasoning (AR) assessment. Seven patients with clinically definite MS (2 males and 5 females) with ages between 18 and 46, and an educational range between 6 and 16 years, were evaluated with an EER task, the Brief Repeatable Battery in MS and the AR-WAIS sub-test. Subjects with lower performance on the AR-WAIS used physical similarity as response criterion in EER task. Only one subject, who obtained the highest performance on the AR-WAIS, used derived EER criterion. These results suggest a relationship between the performance on the EER task and conventional analogical reasoning ability.

Cognition, Behavior, and Memory

Poster Number 122 | Session 2

"To labilize or not to labilize? The importance of being proteasome"

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Once a memory is consolidated, the presentation of a reminder can induce reconsolidation, a process that has inherent stages: reactivation, labilización and restabilization of the memory trace. We are studying the labilization process in two models: context-signal memory in crab and fear conditioning in mouse. A previous study showed that the labilization process involved protein degradation by the proteasome. We found that the inhibition of the proteasome by MG132, block labilization. This drug can inhibit NF-kB during consolidation and this inhibition results in an amnesic effect but, when we administered MG132 previous the re-exposure we did not find an amnesic effect. When we co-injected MG132 with a NF-kB inhibitor we could not find the amnesic effect we expected. The same result was obtained when we co-injected MG132 with a NMDA receptor inhibitor. The GABA inhibitor bicuculline can enhance memory reconsolidation in the crab paradigm, but this effect was not found when it was co-injected with MG132. All together these results indicate that proteasoma is involved in memory labilization. We hypothesized that memory labilization implies partial synapses retraction involving proteasoma dependent protein degradation and then stabilization during the reconsolidation process.

Cognition, Behavior, and Memory
Poster Number 123 | Session 3

"Effects of enriched environment in middle-aged rats subjected to acute perinatal asphyxia"

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Information regarding the effects of perinatal asphyxia in middle-aged rats is scarce. On the other hand, exposure to enriched environment has been shown to be beneficial in different models of brain injury. The main aim of the present study was to assess if PA worsens some of the brain and behavioral alterations frequently observed in middle-aged rats. As well as, if life-long exposure to EE could prevent those alterations. Controls (CTL) and perinatal asphyxiated (PA) rats were assigned to enriched (EE) or standard environment (SE) at 21 days of age, for 18 months. Results showed that CTL and PA rats reared in SE did not differ in their locomotion and anxiety levels, while EE reduced locomotion and anxiety levels in spite of birth condition. Additionally, EE prevented the spatial reference and working memory impairments seen in PA rats reared in SE. Furthermore, EE diminished astrogliosis, increased synaptic density and expression of BDNF in CA1 region of the hippocampus in CTL and PA rats. It is concluded that life-long exposure to EE prevents the cognitive deficits observed in middle-aged rats subjected to perinatal asphyxia, possibly due to the morphological, ultrastructural and molecular changes associated with exposure to EE.

Cognition, Behavior, and Memory
Poster Number 124 | Session 1

"Pregnenolone in lateral septum nucleus affects memory acquisition of male rats via modulation of gabaergic system in a passive avoidance test"

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Pregnenolone (Preg) is a neuroactive steroid (NS) that plays a role in memory, among other functions. Previous results indicated that Preg injected into the lateral septum nucleus (LSN) impairs memory acquisition in an aversive memory paradigm. Our present objective was to study -in the same model- agonists and antagonists of glutamatergic and GABAergic systems, according to the following groups: 1) Control; 2) Ap7 1 µg/µl); 3) Bicuculine 9,8 µg/µl and 4) Muscimol 0,25 µg/µl. Injection of reagents (1µL) was done via a cannula implanted in the right LSN 30 min before training. After 24 hs the subjects were tested for retention. Our results showed that both muscimol and Ap7 have an amnesic effect, while

bicuculine showed no effect compared to the control group. It is important to note that any of the experimental groups did not affect any other significant behavior (motivation, motility and anxiety). It is possible to think of LSN requiring a delicate balance between glutamatergic and GABAergic actions, since the GABAergic agonist Preg disrupts this poise and impairs the acquisition of memory.

Cognition, Behavior, and Memory
Poster Number 125 | Session 2

"Object vs Subject Experiencer Psych Verbs: a step towards understanding the nature of the Syntax-Semantics Interface"

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In a study about sentence comprehension, Gattei, Vasishth and Dickey (2010) performed a self-paced reading task using Spanish Object Experiencer Psych verbs and Dative Agentive verbs. Both types of verbs differ in the way the semantic arguments are mapped over the syntactic structure, and can therefore show possible differences in processing coming from the syntax-semantics interface. The results showed that when the semantic arguments of a sentence are mapped indirectly over the syntactic structure (e.g. gustarle), readers take longer time to process the second argument than when the semantic arguments are mapped directly (e.g. gritarle). Besides, the study showed that readers also took longer time to read the second argument when the syntactic order was manipulated (SVO vs OVS) and the order of the sentence did not respect the semantic hierarchy required by the verb. The present study investigates whether both effects (type of mapping and semantic hierarchy order) occur due to differences corresponding to the semantic categories of the verbs (Psych vs Agentive verbs) or they also appear when two verbs of the same category (i.e. "gustarle" vs "quererlo/la") are compared.

Poster Number 126 | Session 3

"Mechanisms involved in the interaction between the reactivation of a consolidated fear memory and a stressful situation"

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Consolidated memories may result into a labile one after retrieval. It was proposed that this process can allow incorporate new environmental information to the original trace. One of the aims of the present research was to assess whether a stressful stimulus prior to reactivation would influence fear memory and what mechanisms are involved in such influence. Rats were subjected to a contextual fear conditioning using a single footshock. One day after, rats were subjected to a stressful situation. Half of the rats were re-exposed to the original conditioning context (test 1) for 3 min. one day after stress. A significant freezing increase was observed, which persisted 10 days after stress (test 2). Intra-BLA infusion with midazolam prior to stress prevented such increase. The intra-BLA infusion of an inhibitor of protein degradation (B-lac) did not prevent such increase. Although there is a clear interaction between stress and fear expression that leads to a higher fear memory, such increase is not dependent on protein degradation in BLA. What is more, knockdown of hippocampal BDNF impaired the interaction between fear memory and stress. These findings support the view that such interaction recruits mechanisms suggested to be involved in contextual fear memory consolidation.

Cognition, Behavior, and Memory
Poster Number 127 | Session 1

"To transfer or not to transfer. How to measure is the question..."

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Learning without transfer is useless. Whatever we learn, we learn it in a specific context. Whenever we need to retrieve that information, the context will have changed. But we will still need to use that information! The evolutionary strategy that the brain has achieved is generalization. During the last three years we have

been developing Mate Marote, an educational software to exercise different aspects of cognition for five-to-eight-year-olds. One of the goals of this project is to assess its specific impact and its transfer to other cognitive processes and academic performance. At the present more than 150 children have played many sessions at their own schools. As not all executive training produce transfer, this is a main issue for our project. Classically, cognitive transfer in humans is measured by the use and analysis of a batery of standardized tests. Surprisingly, nobody deeply analyses the testsí execution dynamics but just quantify some suggested specific measures. In order to assess the real impact that Mate Marote has in cognitive development, children were tested before and after training, and evaluations of the performance on the tests and games were extensively done. Here we show some interesting results that are very enriched by a profound data analysis.

Cognition, Behavior, and Memory
Poster Number 128 | **Session 2**

"Effect of IL-1ß on hippocampal signalling cascades involve in consolidation of fear memory"

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The effects of cytokines on cognitive processes have been extensively studied. Particularly, IL-1*ß* administered in dorsal hippocampus significantly impairs the consolidation of a contextual fear memory. The mechanisms by which this inhibition occurs in vivo remains to be elucidated. We previously reported that IL-1*ß* produced a decrease in glutamate release after contextual fear conditioning. Here we showed that this hypothesis is also sustained by the fact that treatment with D-cycloserine, a partial agonist of the NMDA receptor, reversed the effect of IL-1*ß* on contextual fear memory. Besides, IL-1*ß* reduces ERK2 phosphorylation, a MAPK critically involved in memory consolidation. We have demonstrated that *ß*-MSH reversed the IL-1*ß* detrimental effect on memory consolidation. Preliminary results demonstrated that *ß*-MSH reverted the pERK inhibition induced by IL-1*ß* and produced an increase in BDNF levels after the contextual fear conditioning.

Cognition, Behavior, and Memory

Poster Number 129 | **Session 3**

"Vesicular trafficking of NMDA like receptors in Neohelice granulata (Chasmagnathus granulatus)"

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The NMDA glutamate receptors (NMDARs) mediate the majority of excitatory neurotransmission in the nervous system. Molecular and electrophysiological properties suggest that they may be the Hebbian¥ coincidence detectors and has been implicated in synaptic plasticity, learning and memory. In the crab Neohelice granulata was demonstrated that the systemic administration of NMDAR antagonists (MK-801, APV) affects the memory consolidation, reconsolidation and extinction memory of the Context-Signal Memory. These results indicate that NMDA like receptors have been involved in long-term memory in this invertebrate model. Here we characterized the NMDA like receptors by Western blot assays and immunohistochemistry techniques. The next step was to evaluate the NR1 like receptor subunit expression as consequence of learning. Additionally, differential surface expression of NR1 like subunit was studied by semi-quantification of the intracellular and extracellular vesicular pool.

Cognition, Behavior, and Memory
Poster Number 130 | **Session 1**

"Effects of pretraining treatment with testosterone on successive and anticipatory negative contrast

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Previous research showed that the suppression of consummatory behavior that follows incentive downshift in male rats is attenuated by testosterone (T) administration during training. The present experiments assess the role of pretraining T administration on successive negative contrast (cSNC) and anticipatory negative contrast (cANC). In cSNC, groups received a single 5-min trial per day of access to sucrose solution. After 10 trials of access to 32% sucrose, Group 32-4 was exposed to a downshift to 4% sucrose for 5 additional trials and

its performance was compared to Group 4-4, which received access to 4% sucrose during 15 trials. The downshift leads to consummatory suppression in 32-4 relative to 4-4. Pretraining T administration enhanced consummatory behavior in rats from Group 4-4, but had otherwise no effects on cSNC per se. In cANC, groups received 2 trials per day separated by a short midtrial interval. For Group 4-32, the first trial each day was 4% sucrose and the second trial was 32% sucrose. For Group 4-4, both trials each day were 4% sucrose. Consummatory behavior in the first trial was suppressed in Group 4-32 relative to 4-4, and pretraining T administration increased responding for 4% sucrose independently of the incentive contrast manipulation.

Cognition, Behavior, and Memory
Poster Number 131 | Session 2

*"Reconsolidation in a word learning process"***Laura Kaczer**[2], Cecilia Forcato[2], María Eugenia Pedreira[2], Alejandro Wainselboim[1]

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The ability to learn new words is a fundamental process in language acquisition that is active throughout life, although little is known about the brain mechanisms supporting it. The current project aims to employ a combined linguistics-memory approach to understand how adults form a long-term association between a novel word and its meaning. In particular, we plan to evaluate a particular phase in the word-learning process: memory reconsolidation. When a long-term memory is recalled by a reminder, it enters a vulnerability phase (labilization), followed by a process of re-stabilization (reconsolidation). It has been suggested that this process makes the incorporation of new information possible. Here, we plan to analyze the presence of reconsolidation in new word learning, using an artificial language paradigm. Our hypothesis is that when we retrieve a novel word memory trace, there is a chance for reconsolidation to occur, allowing us to update or redefine the wordís meaning. In addition, we will address the neurobiological basis of reconsolidation using event- related potentials, measured with electroencephalography. This technique will provide useful information regarding the cortical changes observed during the reconsolidating phase.

Cognition, Behavior, and Memory

Poster Number 132 | **Session 3**

"Opposite actions of dopamine on aversive and appetitive memories in a crab"

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The understanding of how stimuli and reinforcements are represented in the central nervous system during memory formation is a current issue in neurobiology. In insects, it is widely accepted that dopamine (DA) mediates aversive reinforcements in aversive memory formation. Here we address the involvement of DA in aversive and appetitive memories in the crab Chasmagnathus. We found that DA-receptor antagonists impair aversive memory consolidation, in agreement with previous reports in insects. Moreover, exogenous administration of DA facilitates memory formation after a weak training protocol. By contrast with previous reports, DA treatment was found to impair appetitive memory formation, revealing a yet not described action of this amine. As a first step to elucidate the neuroanatomical correlates of DA action on memory, we mapped dopaminergic neurons in the central nervous system of the crab. Results of the current study together with those obtained in a previous work about the role of octopamine (OA) suggest that each amine (DA and OA) has a dual action on memory processes. On the one hand, they mediate the reinforcement signal throughout training and, on the other, they interfere with the formation of memory of the opposite sign.

Cognition, Behavior, and Memory
Poster Number 133 | **Session 1**

"The Tell-Tale Heart: Heart rate and decision-making in chess"

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Chess is used as an ecological but controlled model for higher order cognition and decision-making. In a chess game, each move is a decision. Players have a finite time budget and the goodness of each decision can be evaluated quantitatively. We are using this natural experimental setup to study heart rate (HR) as a physiological correlate of human decision-making. We found that HR depends of game states (available time -AT- and position value) and move properties in standard chess

games. Player AT decrease according to how much time is used in each move. We found a significant negative correlation between HR and AT, with HR increasing by almost 2-fold under time-pressure. HR is also modulated by the position value, increasing when the position is not balanced. The previous measures show that HR indexes the state-values of the game. HR also constitutes a signature of individual moves, increasing when players make blunders (bad choices). These results show that HR signals global and local variables during decision-making. Future studies should investigate the causality of this correlation, whether changes in HR impact decision-making or if the outcomes of the decision modify HR, or if heart and brain form a close-loop signaling parameters of decision-making.

Cognition, Behavior, and Memory
Poster Number 134 | **Session 2**

"Pradation risk may sculpt functional differences in identified brain neurons"

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As a biological organ, the brain is ultimately committed to increase the animalís fitness, by gathering and processing information and organizing the behaviour that allows them to succeed in complex dynamic environments. Thus, the designs and functioning of an animalís brain is assumed to be highly determined by selective ecological pressures. Previous studies on the crab Neohelice granulata identified brain neurons, termed LG, that proved to play a key role in the crabís escape response to visual danger stimuli (VDS) representing a predator attack. Here we show that three different populations of Neohelice, exposed to different risk of avian predation, present clear differences in their response to VDS. Populations exposed to higher risk showed stronger escape responses. Tests performed with other stimuli revealed no behavioural difference between them, thus ruling out an explanation of the response to the VDS in terms of unspecific effects. We have started to perform in vivo intracellular recordings of the LG neuronsí response to the VDS, our preliminary results suggesting that the behavioural differences are reflected by their performance. If confirmed, these results will represent one of the few examples of how ecological pressure shapes the functioning of individual neurons.
Cognition, Behavior, and Memory

Poster Number 135 | **Session 3**

"RK1/2 pathway underlie both the enhancement of anxiety-like behaviour and the facilitating influence on fear memory following stress"

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It is well known that an emotionally arousing experience usually results in a robust and persistent memory trace and the expression of enhanced anxiety. The present study explored the potential mechanisms involved in the influence of a prior stressful event on the consolidation of a contextual fear memory and on the onset of anxiety-like behavior. An activation of ERK2 in basolateral amygdala (BLA) was evident twenty four hours after the stressful experience. Moreover, such environmental challenge facilitated memory formation and increased anxiety-like behavior. In addition, pretreatment with the intra-basolateral amygdala infusion with UO126 (MAPKinase inhibitor) prevents the stress-induced facilitating influence on fear memory formation and on anxiety-like behavioural. Given that the activation of ERK1/2 pathway is essential for associative learning and the occurrence of anxiety, we propose that stress-induced facilitation of p-ERK2 in BLA is an important mechanism for the promoting influence of stress on both processes.

Cognition, Behavior, and Memory
Poster Number 136 | **Session 1**

"Optical imaging of different activation patterns in the lateral protocerebrum of the crab Chasmagnathus granulata during the construction of memories that can or not be expressed in long term"

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Memory is a sequence of processes that includes making an internal representation of the experience, storing and retrieving that information. In the Chasmagnathus¥s Context Signal Memory (CSM) paradigm, the iterative presentation of a Visual Danger Stimulus (VDS) induces a change in behaviour, from escape to freeze. We

focus on Weak (WTP) and Strong Training Protocols (STP). Long term memory (LTM) expression is disclosed after STP but not after WTP. Initially designed to induce only STM, WTP leads also to a latent LTM that is reactivated and could be expressed after improving reconsolidation. The crab¥s lateral protocerebrum (LP), formed by the medulla terminalis and hemiellipsoid body, is proposed as higher-order integration center involved in sensory processing and memory storage. Using Calcium imaging we are evaluating whether the amnesic effect of several drugs described to interfere with LTM, are due to interference on memory storage or to modulation of memory expression. Our preliminary results showed that specific changes in activity at the crab¥s LP induced by training and short-term tests (30¥) are abolished by an NMDA-receptor antagonist. This project¥s goal is to show that LTM expression is a distinct attribute of LTM storage.

Cognition, Behavior, and Memory
Poster Number 137 | Session 2

"Vulnerability of a memory related to drug experience: hippocampal participation"

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Hippocampal synaptic plasticity has been related to learning and adaptive processes developed during chronic administration of drugs of abuse. The existence of common neurobiological mechanisms mediating drug addiction and memory were suggested, but the specific adaptations that sustain addiction or memory along time are still missing. We previously found that changes in contextual cues linked to diazepam withdrawal prevented the expression of the memory associated to the drug experience. Recently it was shown that persistent phosphorylation by PKMZ is critical for the maintenace of hippocampal LTP and spatial memories. Considering all these, the aim of the work is to evaluate if the memory evoked by contextual cues linked to withdrawal experience, could be afected by intrahippocampal administration of the PKMZ inhibitor Zip. Results indicate Zip administration in dependent animals re-exposed to contextual cues prevented the expression of memory. Moreover, the enhancement in the hippocampal synaptic plasticity previously observed in dependent animals was also reversed, demonstrating the relevance of the hippocampus in the maintenance of the memory trace associated to drug experience and withdrawal.
Cognition, Behavior, and Memory

Poster Number 138 | Session 3

"Prenatal ethanol-exposure facilitates ethanol induced-second-order conditioning in infant rats"

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Epidemiological and pre-clinical studies indicate that prenatal exposure to ethanol is associated with heightened probability of problematic ethanol consumption later in life. In recent studies we have developed a second-order conditioning (SOC) place preference to analyze ethanol reinforcement. The present study assessed ethanol-induced SOC in infants rats exposed to ethanol during late gestation (maternal administration of 2.0 or 0.0 g/kg ethanol, gestational days 17 to 20). Two-week old rats derived from ethanol or vehicle-treated dams (total: 16 litters) were given ethanol (0.5, 1.0 or 2.0 g/kg) followed by the intraoral infusion of a conditioned taste stimulus (CS1). Unpaired controls were employed. The CS1 was then paired with a distinctive tactile cue (CS2, sandpaper). ANOVAs indicated ethanol-mediated reinforcement, indexed by measuring preference for the CS2 in a two-way test, in rats that had been exposed to ethanol in-utero but not in counterparts reared by vehicle-treated dams. These results suggest that prenatal experience with ethanol facilitates the acquisition of later ethanol-induced appetitive learning. These could be one the mechanisms underlying the permissive effect that prenatal ethanol exerts on later predisposition for alcohol intake.

Cognition, Behavior, and Memory

Poster Number 139 | Session 1

"Role of the system nucleus incertus/relaxin3 in pavlovian conditioning"

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Fear conditioning is one of the best-studied forms of emotional learning. The nucleus incertus(NI) designed the centers involved in the process of extinction and conditioning to context.The designed occurs through some projections that

GABA is co-released by the peptide relaxin3.Determine if the injury of the NI causes the modification of the patterns of Pavlovian conditioning. In male rats underwent an electrolytic lesion of the NI and after 20 days of recovery were subjected to fear conditioning protocol of 5 days.On 1 and 2 at 2 times a day, visited 2 types of context (ctx).Ctx2 was the conditioning and the extinction ctx1. The two contexts differed in shapes of the walls, touch and smell.Day 3 was the conditioning exposing the animals to 3 pairs of tone-shock.Day 4 was tested from the conditioning context in ctx2 and in ctx1.Day 5 is the day test of extinction. The controls spent the same protocols except the injury.Throughout all processes measured the amount of freezing using a load cell at the base of the box.The lesion of the NI no impact on significant changes in the level of freezing or during habituation and during conditioning.The injured animals had a greater resistance to extinction. Data suggest that the NI modulates some aspects of Pavlovian conditioning.

Cognition, Behavior, and Memory
Poster Number 140 | **Session 2**

"Re-examining the ontogeny of the context-preexposure facilitation effect in the rat"

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According to recent studies, the so-called context-preexposure facilitation effect (CPFE) is not observed until postnatal day 23. In these studies the foot-shock intensity employed was relatively high (1.5 mA) and context conditioning was inferred exclusively from a single behavioral measure (percentage of freezing). In adult rats studies employing a less intense foot-shock (1mA)showed evidences of contextual fear conditioning in the immediate-shock condition. The present study examined the CPFE on PD17 and PD23 by means of the analysis of multiple dependent variables. The experimental design was defined by three between-group factors: age (PD17 or PD23), preexposure (preexposed or non-preexposed) and foot-shock (0, 0.5 or 1.5 mA). In the present study we observed comparable levels of freezing than those reported by other authors using similar parameters. Additionally, the CPFE was limited to one dependent variable and detected at both ages. However, evidences of fear were found in a number of dependent variables regardless the pre-exposure treatment. Interestingly, fear responses

were generalized to an alternative environment. These results may have important implications for the analysis of the ontogeny of contextual fear conditioning.

Cognition, Behavior, and Memory
Poster Number 141 | **Session 3**

"Could pre-imaginal olfactory experiences modify the post-metamorphic behavior in a social insect?"

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In honeybee colonies the information of exploited resources is transferred among nest mates through mouth-to-mouth trophallaxis events. These social interactions also involve larvae as food recipients. We wonder if appetitive olfactory experiences that occur during pre-imaginal stages influence post-metamorphic odor-mediated responses. To address the issue we tested learning performance and memory retention in adults of 3/5 days old that underwent a pre-imaginal experience. Such experience was done by means of a scented-sucrose solution offered inside the hive and tested under the proboscis extension response (PER) paradigm in the lab. Results showed that precocious experience increased the PER-levels toward the pre-exposed odor suggesting retention of information gained prior to the emergence. Interestingly, high PER-levels to novel odors were also found accordingly to perceptual similarities between pre-exposed and different novel odors, resembling generalization in tested adults. Lastly, we found that even those bees that did not respond to the pre-exposed odor improved their learning performance in a PER-conditioning. Thus, pre-imaginal experiences may allow bees to assess food information very early in life with consequences in their learning abilities during adult stage.

Cognition, Behavior, and Memory
Poster Number 142 | **Session 1**

"Role of mPFC 5-HT2a receptors in the resolution of memory interference during retrieval"

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The medial Pre Frontal Cortex (mPFC) has been involved in complex integration of information necessary during response selection and interference. The interference theory refers to the idea that forgetting occurs because the recall of certain items interferes with that of other items. Thus, the retrieval of two memory traces that share certain neuronal circuits will activate the PFC to prevent interference resulting in the forgetting or suppression of one of the traces in favor of the other one. The mPFC is modulated by many neurotransmitter systems including the serotonergic system. In the mPFC one of the most important receptors is the 5-HT2aR. It is not clear which is the role of the serotonergic system in memory interference (MI) processes. In this work we evaluate the role of mPFC 5-HT2aR in memory interference using different versions of the spontaneous object recognition task in rats. We found that blockage of mPFC 5-HT2aR 15 minutes before retrieval alter the response in the temporal order and object in context tasks. Blockage of 5-HT2aR but not 5-HT2CR affects the ability of mPFC to avoid MI. Activation of 5-HT1aR also produces MI. Biochemically, increase MI correlates with an increase expression of immediate early genes in the perirhinal cortex.

Cognition, Behavior, and Memory
Poster Number 143 | Session 2

"Reward's memory impairment"

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The behavior of animals depends on their previous experience with rewards of different incentive value. Blockade of fl-adrenergic receptors leads to decline in memory of several tasks. The aim of this study was to analyze the administration's effect of a fl-adrenergic antagonist on a reward's downshift and omission memory. It was found that post-training administration of propranolol, results in a faster recovery of the consummatory response when the animals were downshifted from a 32% sucrose solution to a 4% sucrose solution, suggesting the existence of

memory impairment. On the other hand, the same drug administered immediately after the omission of the reward did not affect the extinction of the consummatory behavior. These data suggest that propranolol impairs memory of the downshift reward in an opposite way to the corticosterone's memory enhancing effect of this phenomenon. The results are discussed according to the role played by the noradrenergic system on incentive value's shifts memory and the asymmetry found in the results of consummatory extinction.

Cognition, Behavior, and Memory
Poster Number 144 | Session 3

"Exploring the limits of Bayesianity in the human brain"

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Consistently, we find that humans are near optimal (Bayesian) in processing and combining information from different sources in widely varying tasks. This poses the question of how this is achieved with our neural architecture. In particular, we are interested in how far this optimal processing can go, testing it in a task that is a good cadidate to expose its limitations. Fragmented empirical evidence points to the fact that optimality might be broken in the context of sequential decision making, and we thus set out to establish this by performing deliberately designed experiments. Here, we present the theory underlying the experiments, and describe in detail their design and implementation. Further, we show some preliminary results that confirm the hypothesis, along with a discussion of why this could be the case given our particular neural network.

Cognition, Behavior, and Memory
Poster Number 145 | Session 1

"Effect of object features and retinal position on the crab escape response induced by visual stimulation"

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The escape response to visual stimuli of the crab Neohelice granulata is being used as a model to investigate the neural control of visually guided behaviours. The lack of gaze movements and the compound eyes of these animals make it possible to predict the visual information that crabs use to initiate escape. In their natural habitat, crabs are exposed to a large variety of visual stimulus. Given the limited spatial resolution of their eyes, it has been proposed that crabs could use a simple rule based on the position in retinal elevation to recognize dangerous from harmless moving objects (Layne 1998). We found that the intensity of the escape response is largely independent of the stimulus position, whereas habituation differs across the visual field. The response proves to be highly dependant of the stimulus characteristics. For example, a virtual looming stimulus induces stronger responses than images moving in a trajectory of no collision. Paradoxically, the later stimulus (potentially less risky) provoked earlier responses that the former, in agreement with results obtained in field with fiddler crabs. Results are discussed regarding to the pressures of the ecological context, the constrains of the crab optical system and the organization of their visual nervous system.

Cognition, Behavior, and Memory
Poster Number 146 | Session 2

"Using all your fingers, Multi-touch as apparatus in experiments"

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Nowadays many of the tasks being done by subjects during behavioural experiments involve a computer, this is a hard restriction, because not every subject is capable of using it (ie: toddlers and very young children). Furthermore mouse/keyboard interfaces does not allow multiple input from one or even many subjects at the same time. That is why we started developing a multi-touch user interface (UI), it is software based on a ReacTiVision table which is much more approachable and usable (Zack, 2009). This UI is being implemented with a novel software framework that enables us to measure movement path and precision, response time, multi-agent input among others in real time and with high fidelity. The experiments are implemented as computer games, which allow us to display stimuli and interact with them in many ways. This system is in continuous growth, so we are considering the possibility to include video recording capability in order to recognize different players and their interaction around the table. Right now this system is being used in experiments with young children, aged between 3 and 8 years old, in an attempt to reproduce casual learning games (Gopnik, 2000).

Cognition, Behavior, and Memory
Poster Number 147 | Session 3

"Eye-hand coordination in serial tasks with preview"

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In action sequences, the eyes are generally fixated ahead of the stimulus being responded to, overlapping the processing of adjacent stimuli. What determines the timing of the saccade to the next item is initiated and which parts of the processing can be overlapped in time remain controversial. We designed a task based on a game often used to train typing skills, in which letters appear at the top of the screen and fall vertically, and are killed by typing. We found that participants use two different ways to coordinate hand and eye movements. One way is purely serial processing of each of the sequentially process targets. In this case, a target is fixated, and the gaze is maintained on the letter until its response is produced. In a second way, parts of the successive targets are processed in parallel. The gaze leaves the target and begins the processing of the following, before producing its response. A third mode of eye-hand coordination was what we called "double-fixations". These fixations were directed to the mid-point of two letters, and both were tapped. The proportion of fully-sequential-to-overlapping and the proportions of double fixations explain in part the variability in performance of participants.

Cognition, Behavior, and Memory
Poster Number 148 | Session 1

"Amnesia by inhibiton of dorsal hippocampus protein synthesis or NMDA receptors is overcome by previous OF exploration"

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To further investigate the participation of hippocampal protein synthesis and NMDA receptors (R) in memory formation, we trained rats in a one-step inhibitory avoidance (IA) task: when stepping-down from an isolated platform onto a grid-floor they got a 0.5 mA footshock; latencies to step-down in training and test

session, performed 24 h later, were recorded. Rats were injected intrahippocampus with the protein synthesis inhibitors Anysomicin or Cicloheximide (8 µg/side) 120 min before IA training, leading to anterograde amnesia. The NMDAR antagonist MK801 (2µg/side) was injected immediately after IA training, leading to retrograde amnesia. Other rats were let to explore twice an open field (OF) for 3 min, 24 h and 90 min before IA training, showing habituation to the arena. Although they received the drugs according to a similar protocol, these animals were able to express a conspicuous long term memory of IA. Our results corroborate the amnesic effects of the three drugs, but revealed that the previous exploration/ habituation to the arena allowed to overcome the amnesia, raising questions about dorsal hippocampus protein synthesis as unique source of synaptic plasticity proteins for this aversive memory formation, and the hippocampal NMDAR involvement there.

Cognition, Behavior, and Memory
Poster Number 149 | Session 2

"Behavioural and cellular changes triggered by sublethal doses of Stx2"

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Infection by Shiga toxin (Stx2)-producing enterohemorrhagic E. coli (STEC) causes hemorrhagic colitis, Hemolytic Uremic Syndrome (HUS) and neurological complications mainly in children. We developed a model to simulate some of the consequences of STEC infection. Our aim is to study the action of 2 sub lethal doses of Stx2 (Stx2sd) or saline at 4, 7 and 20 days in 20-25gr NIH mice. Mice were subjected to a set of behavioral tests: Open Field, Elevated plus maze, Object recognition and Rotarod. No changes in Open field, Elevated plus maze and Rotarod were recorded in none of the 3 concentrations tested. Significative differences were found at 4 and 7 days at the higher concentration in the Object recognition test. However, the change in memory is reverted at day 20. To study the effect of dsStx2 at a cellular level and to understand the reversion of the damage at 20d, we performed confocal double fluorescence studies in the hippocampus with: anti-Stx2, tomato lectin, Fluorojade-b, NeuN and GFAP. We observed a rise in the vascular area and GFAP marker which descends after 20 days. The present findings indicate that sublethal doses of Stx2 alter the memory of treated mice

and this is compatible with clinical reports of children who develop STEC derived encephalopaties.

Cognition, Behavior, and Memory
Poster Number 150 | **Session 3**

"Impact of different environmental experiences during development on adult rat behavior"

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Environmental influences play key role in refining neural development as well as on behavioral and cognitive functions. The aim of this study was to analyze the regulatory effects of different environmental qualities during development on neurobehavioral systems involved in memory, learning and defensive responses. A maternal separation (MS) protocol of 4,5h/day from birth until weaning and an enriched environment (EE) from weaning were applied as environmental manipulations on Wistar-derived male rats. At 60 d of age, exploratory behavior, emotional reactivity and cognitive responses were assessed using the open field, novel object recognition test (NORT) and step down inhibitory avoidance (IA) learning paradigm. We found that MS decreased grooming behavior while EE produced the opposite effect. Although groups didnít show significant differences when tested by NORT, animals with MS presented decreased latencies on the IA task and EE reversed this effect. Regarding defensive behaviors such as freezing and thigmotaxis, EE generated different responses according to the rearing experiences. These findings provide evidence for reversal of the behavioral effects of early life maternal separation. Such effects might involve compensatory changes associated with peripubertal enrichment.

• COMPUTATIONAL NEUROSCIENCE

Poster Number 151 | **Session 1**

"Predicting "in silica" the effect of new probable missense and intronic pathogenic mutations in Neuronal Ceroid Lipofuscinosis type CLN2"

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CLN2 Neuronal Ceroid Lipofuscinosis (NCL) prevails in Argentina (out of 13 types) showing mutations in the TPP1 gene of Tripeptidil Peptidase-I being recognized 72 pathogenic mutations. We aimed to study genomics incorporating bioinformatics. In 5 enzymatically deficient patients full screening with PCR and direct sequencing showed 5 substitutions -4 missense, and 1 intronic unpublished but previously mentioned (https://www.ucl.ac.uk/ncl/mutation.shtml). Later, the outcome was analyzed "in silica" with the selected programs MaxEntScan, ClustalW2, PolyPhen-2, SIFT, PoPMuSiC 2.1 and the database Protein Data Bank. The analysis revealed the potential impact of the substitutions on the protein showing mutually consistency, and supporting their possible pathogenic effect. Thus, the combination of bioinformatic softwares with the full screen of the TPP1 gene allowed predicting the probable pathogenic effect of substitutions in the DNA prior to performing experimental tests in the laboratory.

Computational Neuroscience
Poster Number 152 | Session 2

"Neuronal oscillations driven by noisy current inputs"

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The dynamics of a single neuron can be mathematically modeled by systems of differential equations whose variables describes the biophysical properties of the membrane. When driven with a constant strong current, the neuron spikes are regular. However, in realistic conditions the neurons operate with noise and receive fluctuating currents through their dendritic afferents. In this work, we present how the firing frecuency of the neuron varies with the application of a current with different noise levels. To achieve this, we simulate the neuronal dynamics with conductance models such as Hudgkin-Huxley and Morris-Lecar driven with a stochastic current. We observe that the firing frecuency of the neuron depends of the noise level of the applied current and, for certain levels of noise, the spike variability is maximum.

Computational Neuroscience
Poster Number 153 | Session 3

"The history of emotions:

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Morality plays a central role in the constitution of human nature. The social code of conduct emerges from a sophisticated integration of cognitive, emotional, and motivational mechanisms, internalized through an active process of cultural learning. Recent efforts explore clustering of social concepts by using adjectives that can be associated with behavioral or personality traits and may be assigned a valence. We compared this clusters with historical occurrences in literature by measuring similitude between this adjectives associated to social concepts using LSA trained with TASA database. For each pair of words we were able to measure their similitude in a quantitative semantic space, finding an intrinsic structure with moral emotions being quite close to all social concepts, as if they would be in the center of this semantic space. By analyzing the taxonomy of virtue by the blind classification process, we expect to reconstruct the theoretical categories of virtues and social concepts. Finally, we propose the definition of proximity based on the co-evolution (in time) of words using the Google Ngrams, from where the historical evolution of emotions may be inferred.

Computational Neuroscience
Poster Number 154 | Session 1

"Estimating mean rates with firing linear-nonlinear neuron models"

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The firing rate of a neuron depends on the mean and the variance of the input currents exciting the cell. Real neurons are exposed to variable amounts of noise, depending on the behavioral context in which they operate. Theoretical models allow us to predict the firing rate as a function of the stimulus attributes. One such theory has been developed for linear-nonlinear Poisson neurons that are characterized by a single receptive field. The model has been previously used to

predict the firing rate of conductance-based neurons, as the Hodgkin-Huxley model. In this work, we study the applicability of the linear-nonlinear Poisson approximation to describe the firing rate of several simplified and realistic neuron models. We find that the theory is approximately valid for small amounts of noise. As the noise increases, however, simulated neurons show an increase in the firing rate proportional to the standard deviation of the input current, not predicted by the theory. The discrepancies appear because in conductance-based models, rapidly fluctuating stimuli cannot be replaced by an average over constant stimuli.

Computational Neuroscience
Poster Number 155 | Session 2

"The GROT project: global repository of thoughts"

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One-to-one educational models awaken new educational scenarios, with recent initiatives delivering low-cost laptops to every student. At the present more than 400000 children and adolescents share the same digital platform. This provides a great amount of new possibilities, pushing a profound reformulation of teaching and learning, generating virtually infinite data from children. From the perspective of information technology, the challenge is how to interpret this vast corpus of cognitive development of kids and convert conceptual questions from the educational practice into quantifiable, analytic queries. Our goal is to develop a computational framework for information storage, access and analysis, which may enlighten ancestral cognitive questions. The project has three concrete objectives of progressive difficulty: 1) to develop a tool that includes educational games and applications and registers playing behavior; 2) datamining and analysis of registered data which may reveal the underlying mechanisms of thinking (as a first approach, we will attack memory, planning, numerosity, geometry and inhibitory control aspects); and 3) to generate interventions in the educational process of the childs towards a more equitative education, providing equal opportunities to every child.

Computational Neuroscience
Poster Number 156 | Session 3

"Acquisition of capacities visual motor to inclination of a dynamic simulation with delay"

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From learning model given by the exponential growth inhibited, represented by the initial value problem which simulates the learning of skills acquired is proportional to the maximum number of such capabilities to acquire, we propose a continuous model with delay. That information is analyzed student populations from country side schools in the province of Catamarca, obtained by applying a cognitive test to establish the relationship between the occurrence of possible alterations of toxic pollutants used in these areas with agricultural activity and school performance. The simulation is based on the delay of nursing students in the pace of its progress through the education system. The relationship grade - age is evident in those who repeat a grade or entering the education system in advanced age. The exponential growth model inhibited delay obtained was used for simulating the dynamics of Vensim software system, which discretize the differential equation into difference equation modeled.

Computational Neuroscience
Poster Number 157 | Session 1

"Understanding children's decision processes with planning algorithms and search heuristics"

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We studied the performance of children, 6 to 7 years old, playing a simple planning game (first appeared in Klahr 1985) that was part of a large scale experience in primary schools (Goldin et al, submitted). Our aim is to understand how children develop and implement planning strategies to solve the game. We adopt ideas and algorithms from artificial intelligence (AI) and reinforcement learning (RL) to model human decision processes. We first show that learning markovian models (as Q-learning) are insufficient to account for aspects of the data such as the

tendency to persist in specific movement directions (inertia). We then implement tree search forward planning algorithms and search heuristics to model the decision processes, to understand the construction of heuristics and the dynamics of plans. We used statistical techniques to fit the parameters for different computational models and later cross-validated them with the data.

Computational Neuroscience
Poster Number 158 | Session 2

"Genetic bases of mammalian inner ear evolution: Analysis of the Beta V-Spectrin gene"

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The mammalian inner ear has unique hearing capacities that distinguished it from the hearing organ of other vertebrates. Our main objective is to study the genetic basis underlying the evolution of the particular functional capacities of the mammalian inner ear. Using maximum likelihood methods we are analyzing the evolution of all inner ear proteins to reveal which of them were shaped by positive selection in the lineage leading to mammals. We hypothesize that proteins showing signatures of positive selection in mammals might have acquired new functional capacities that could underlie the particular capacities of the mammalian inner ear. We found in previous work that the motor protein prestin, a key player in the functional capacities of the mammalian inner ear underwent adaptive evolution in the lineage leading to mammals. Here we report that betaV-spectrin, a cytosqueleton component that interacts with prestin has accompanied such evolutionary trend, showing strong signals of positive selection. Our results suggest that a process of positive selection acting on these two proteins has shaped them to fit a mammalian specific function in the inner ear. The present work continues to delineate the genetic bases underlying the evolution of the inner ear in mammals.

• MOTOR SYSTEMS

Poster Number 159 | Session 3

"Low noise amplification of physiological recordings"

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Birdsong production involves the driving of a nonlinear biomechanical periphery (the vocal organ) by physiological instructions, originated in the bird's brain. These instructions are learned, and the result of that interaction is a diverse set of vocalizations. This makes birdsong a preferred animal model to study learned, complex behavior. Within a program aimed at unveiling the mechanisms underlying such learning process,we built a bio-prosthetic avian vocal organ that integrates a mathematical model of a bird's syrinx,when driven by a muted bird via physiological recordings. The on-line treatment of physiological recordings requires stability and low noise during the acquisition and amplification phase. We present a student\'s lab project which implements an ubiquitous solution, with perspectives to portability. We built a two-stage custom amplifier. Recordings are pre-amplified in-situ (inside a Faraday cage, where the subject is located) by means of a battery-operated stage. In a second stage, the signal is prepared for real-time integration to produce synthetic birdsong. The two-stage amplifier presented here is an affordable solution, suitable for the acquisition of physiological signals such as ECG, EMG and those resulting from mechanical transducers.

Motor Systems
Poster Number 160 | Session 1

"Subject-controlled bioprosthetic avian vocal organ"

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Songbirds are an ideal animal model to study general mechanisms underlying complex, learned motor behavior, as it is birdsong. While much effort focuses on neural control, we highlight the relevance of the interplay between the central mechanisms of motor control and the highly nonlinear peripheral biomechanical systems. The most widely studied songbird species is the zebra finch (Taeniopygia guttata). Its vocal organ is capable of generating a large variety of acoustic signals,

which range from simple whistles to highly complex sounds. In this work, we build an electronic syrinx with aims in the exploration mechanisms underlying birdsong production. We propose a mathematical model for the zebra finch syrinx, in which physiologically meaningful variables determine the acoustic features of the solutions; and we implement this model in a digital signal processor. This device is capable of recording the pressure gesture of a muted bird and integrating the model on-line to produce synthetic birdsong, which replaces the bird\\'s own auditory feedback. This subject-controlled prosthetic device allows for a set of altered auditory feedback experiments, where feedback is altered consistently with modifications of the motor instructions intended to produce them.

Motor Systems
Poster Number 161 | **Session 2**

"Acoustic observables of sound source-tract coupling"

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Birdsong has become a favorite animal model to study complex, learned motor behavior. The observed behavior, the complex set of vocalizations, results of the interaction between a pattern generator in the central nervous system and a nonlinear biomechanical device: the vocal organ. We focus on complexity of the behavior introduced by the dynamics of the vocal organ, which is capable of producing a variety of complex vocalizations even when driven by simple motor gestures. In the vocal organ, a sound source is attached to a vocal tract. If the tract does not act as a mere filter, but is acoustically coupled to the source, nonlinear phenomena are introduced, which can leave a signature in the acoustical features of vocalizations. Inspired by the sound production mechanisms of songbirds, we study a mathematical model of a vocal organ, in which a simple sound source interacts with a tract. We explore the system numerically, and by taking it to the weakly nonlinear limit, we examine its periodic solutions analytically. Nonlinear features of the solutions are proposed as the underlying mechanisms of observed phenomena in birdsong, such as "frequency jumps", enhancement of resonances, and the shift of the fundamental frequency observed in heliox experiments.

Motor Systems
Poster Number 162 | Session 3

"Exploring the vocal tract dynamics in a low dimensional parameter space"

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Many different models of the human vocal system exist that efficiently describe and synthesize single phonemes. However, as recent research shows, many properties of articulated speech rely on the dynamics of the vocal tract. The dimension of this problem is one open question. In other words, the temporal evolution of how many points in the vocal tract must we measure to know which phoneme its being articulated. In this work, we developed an experimental device using hall effect detectors, that allows us to directly monitor the dynamics of three points of the upper vocal tract. We measure the aperture of the center of the lips, the jaw and the position of the tongue. So far, we found that the different patterns obtained allow to recognize the majority of the Spanish consonants embedded in a fixed vocal context. These results suggest different bio-prosthetic applications for articulatory speech synthesis, exploiting the low dimensionality of the problem.

Motor Systems
Poster Number 163 | Session 1

"Dissecting the specificity of hindbrain premotor circuits"

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Motor behavior is the ultimate output of all brain processing and results from the complex sequence of muscle contractions and relaxations at precise times. A crucial step of control for this system is played by neurons monosynaptically connected to motor neurons, the so-called premotor circuit. We used monosynaptically restricted trans-synaptic rabies virus to visualize premotor neurons in the hindbrain, connecting to functionally distinct motor neuron pools (Stepien et al. 2010). Comparative analysis of 3D reconstructions revealed differences in premotor distribution from different motor neuron pools. We further demonstrated that the caudal most part of the reticular formation contains glutamatergic neurons

directly connecting to a specific subset of forelimb but not hindlimb motor neurons and these neurons receive direct cortical input. We postulate that these premotor neurons may be part of a modular system controlling co-activation of motor neurons activated during specific tasks such as reaching movements of the forelimbs.

Motor Systems
Poster Number 164 | Session 2

"Syllable breaking after cooling telencephalic nuclei unveils the presence of a second timescale in the birdsong motor pathway"

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Motor activities are affected by telencephalic areas in a way that is not fully understood. They may control motor behavior, not being affected by downstream motor circuits[Long & Fee 2008, Nature 456] In other perspective, there may be an interaction with the dynamics of these circuits that could have timescales that are different. Taking into account the latter hypothesis, deformation of motor gestures may be predicted in an experiment. Domestic canaries (Serinus canaria) were conjectured to have subharmonic responses in their intra air sac pressure patterns during singing. This may be the downstream circuits response to the activity of thelencephalic nuclei HVC[Trevisan et al. 2006,PRL 96;Alonso et al. 2009,PRE 79; Alliende et al. 2010, Dev. Neurob.70] In this work we modified experimentally the activity of HVC in canaries. We built a cooling device capable of lowering these nuclei temperature decreasing their instructions frequency. We measured simultaneously birdsong and intra air sac pressure patterns. In addition, we show the similarities between our results and a dynamical model with the possible signatures of brain circuits involved in this process.

Motor Systems
Poster Number 165 | Session 3

"Analysis of structural plastic changes underling L-DOPA induced dyskinesias in an animal model of Parkinson's disease"

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L-DOPA remains most efficacious drug for the treatment of Parkinsonís disease despite it induces severe motor complications (dyskinesia). L-DOPA-induced dyskinesias (LID) are correlated with post-synaptic plastic changes in D1 pathway striatal neurons deprived of DAergic innervation. It is well known that L-DOPA induces persistent molecular and that LID are rarely reversible. Our current hypothesis is that striatal structural changes imply an increase of the D1 pathway synaptic connectivity. To test it, we developed an animal model of LID in transgenic mice expressing red or green fluorescent markers under control of the D1 (striatonigral neurons) or D2 (striatopalidal neurons) receptor promoter, respectively. Mice were injected with 6- hydroxydopamine in the medial forebrain bundle and treated with increasing doses of L-DOPA. As expected we found a dose-dependent induction of LID. In these animals we are analyzing the structural modifications in each neuronal type by means of morphological analysis of their dendritic trees. We expect that this work will allow a broad comprehension about the structural changes that take place in the striatum as a consequence of L-DOPA treatment and its relation with the development of dyskinesias.

Motor Systems
Poster Number 166 | **Session 1**

"Reconstruction of motor gestures in birdsong"

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Behavior emerges as the interaction between a nervous system, a peripheral biomechanical device and the environment. In birdsong production, this observation is particularly important: songbirds are an ideal animal model to unveil how brain structures reconfigure themselves during learning of a complex behavior as song. Therefore it is important to understand which features of behavior are controlled by independent tuning of neurophysiological parameters, and which are constrained by the biomechanics of the peripheral vocal organ. In this work we show that

many of the acoustic features in the Zebra finch song are in fact conditioned by the biomechanics involved. We show how to reconstruct the physiological parameters used to drive the avian vocal organ during birdsong production from recorded song. The procedure involves fitting the time dependent parameters of a model. It is implemented as dynamical system ruling the behavior of the oscillating labia that modulate the air flow during birdsong production, together with the equations describing the dynamics of pressure fluctuations in the vocal tract. Finally, we compared the reconstructed instructions with direct measurements of the physiological parameters during song and synthesized a realistic song.

Motor Systems
Poster Number 167 | Session 2

"Effects and adaptations induced by long term treatment with dopamine agonists in an animal model of Parkinson's disease"

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L-DOPA and pramipexole are frequently used drugs in the treatment of Parkinsonís disease. L-DOPA is the most effective to alleviate motor disability but induces abnormal involuntary movements (AIM). Pramipexole have been proposed to have neuroprotective effects and less induction of AIM. To evaluate possible differences at the gene expression level, rats were injected with 6-hydroxydopamine in the striatum and a partial lesion model was characterized. After surgery, animals were treated with L-DOPA or pramipexole at doses that produced similar therapeutic benefit. We analyzed 3 independent groups of normal and lesioned rats by DNA microarray technology in order to compare the genetic profile of the groups: normal/vehicle, lesioned/vehicle, lesioned/L-DOPA, and lesioned/pramipexole. We found that chronic treatment with L-DOPA and pramipexole induced changes in gene expression. We demonstrated that besides the differences seen in the pharmacological profile and the clinic effects of L-DOPA and pramipexole, these drugs also have important differences at the gene expression level. The analysis of differentially expressed genes induced by these drugs allowed us to elaborate new hypothesis, being the most valuable result of massive gene analysis.

• NEURAL CIRCUIT PHYSIOLOGY

Poster Number 168 | **Session 3**

"Research project: assessing the physiological impact of NMDAR ablation in interneurons on prefrontal cortex activity during postnatal development"

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Using genetically modified mice we have recently shown that ablation of NMDA receptors (NMDAR), exclusively in corticolimbic parvalbumin positive interneurons and during early postnatal development, results in a schizophrenia-like syndrome. Notably, no such abnormalities were detected when the conditional knockout of NMDAR occurred after adolescence. This project is aimed to elucidate the physiological changes that take place trough postnatal development in this animal model of schizophrenia. We will focus in the medial prefrontal cortex (mPFC) since it has been postulated as the primary site of cortical dysfunction in schizophrenia. To analyze the electrophysiological activity at different ages we will conduct multichannel-tetrode and local field potential recordings in anesthetized mice to study unitary activity and it engagement in cortical rhythms like gamma oscillation. Also, we will evaluate circuit level connectivity by stimulation of monosynaptic excitatory afferents to mPFC. We speculate that NMDAR hypofunction in interneurons during early postnatal development will result in abnormal GABAergic control of pyramidal firing and coordination, leading to increased, uncoordinated activity in adult animals. This project is supported by grants from ANPCyT, CONICET and NARSAD.

Neural Circuit Physiology
Poster Number 169 | **Session 1**

"Gradual changes in body sodium status induce a specific pattern of behavioural, renal and brain activity"

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We analyzed the effect of gradual changes in body Na status induced by iv infusions of hypotonic (0.075M) and mild(0.3M)/high(1.5M) hypertonic Na solutions on drinking, renal and brain activity responses. Male rats were iv infused with either of the 3 solutions or saline(0.15M) as a control (0.15ml/min/20min). For behavioural and urinary measurements, rats had access to water during the infusion period and 2h after. For immunohystochemical studies other group of rats was infused without access to water and brains were collected 60min after the 20min inf. Our results indicate that both 0.3M and 1.5M groups had a gradual increase in renal Na-excretion but only in the 1.5M group, thirst was elicit. Consistently, the telencephalic circumventricular organs (CVOs) were activated in 1.5M group while the supraoptic vasopressinergic/oxitocinergic (AVP/OT) cells were gradually activated in both, 0.3M and 1.5M groups. The 0.075M group had a different K-renal-excretion suggesting the existence of aldosterone-mediated cell-volume regulation. Our data reveals that a gradual increase in body Na status first triggers the renal response and the hypothalamic AVP/OT cells activation without any drinking response. A higher level of Na overload elicits thirst and the activation of telencephalic CVOs.

Neural Circuit Physiology
Poster Number 170 | Session 2

"Role of long term depression in corticostriatal postnatal maturation in vivo"

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It is assumed that neuropsychiatric disorders like obsessive-compulsive and attention deficit hyperactivity disorder stem from corticostriatal malfunction. However, the current knowledge of the functional postnatal development of these circuits and whether dopamine (DA) pathways are involved in that process is scarce. We have previously shown that early DA depletion delays the functional maturation of the corticostriatal system: striatal spontaneous activity is higher in young mice than in adult mice, but spontaneous activity remains high in adult mice that received early DA-depleting lesions (at PD2). Here we induced corticostriatal long term depression (LTD) in vivo by a saturation protocol using high frequency stimulation (HFS) of the prefrontal cortex, in young and adult mice

with or without neonatal DA depleting lesion. Repeated HFS induced cumulative plastic changes. While the susceptibility to HFS is higher in young mice and adult lessioned mice than normal adults, maximal depression after repeated HFS is the same in all groups. We propose that striatal spontaneous activity reached during development determines the capacity to suffer LTD. In control adults, HFS could recruit less spontaneous active neurons so that one attempt of induction causes a lesser degree of LTD.

Neural Circuit Physiology
Poster Number 171 | Session 3

"Strategies toward identifying the source of endogenous 5-HT involved in the SSRI effect"

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Selective serotonin reuptake inhibitors (SSRI) evoke synaptic activity in identified neurons from leech ganglia, through the activation of a cord spanning interneuron. We asked about the source of endogenous 5-HT involved in this effect. Local and remote Retzius neurons and interneurons 21/61 are possible candidates (IIRNC2010). To answer the question we used two strategies: 1) to inhibit the activity of the candidates and block the effect, and/or 2) to stimulate the activity of the candidates and reproduce the effect. Based on the connectivity of the serotonergic neurons in the leech ganglia, hyperpolarizing (-10 nA) one of them inhibits the rest. However, we did not know if a hyperpolarizing current applied to a serotonergic soma in a ganglion arrives to the terminal that innervates the adjacent ganglion without decay, and thus blocks the release of 5-HT. Thus, we studied if applying a hyperpolarizing current (-10 nA) in an S cell affects the excitability of the S cell from an adjacent ganglion, taking into account that they are both connected by an electrical synapse. Results shown that the signal decay with distance (n = 3) and therefore it is not a valid strategy. The stimulation of serotonergic neurons by different protocols has not been successful (rate of success = 3/16).

Neural Circuit Physiology
Poster Number 172 | Session 1

"Participation of sex chromosome complement in brain pattern activation during acute sodium depletion"

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In order to evaluate whether sex chromosome complement (SCC) differentially modulates brain activity during acute sodium depletion (DEP) we used the 4 core genotype mouse model in which the effect of gonadal sex and SCC is dissociated, allowing comparisons among XX and XY females as well as in XX and XY males. Gonadectomized male and female mice were submitted to DEP by a combined treatment of the diuretic furosemide and a low sodium diet. Twenty one hours later, the mice were perfused and their brains were subjected to Fos inmunoreactivity (Fos-Ir) procedure. The statistical analysis shows that DEP elicits a significant increase in the Fos-Ir at the vascular organ of the lamina terminalis and median preoptic nucleus irrespectively of sex and CCS factors. Activation of subfornical nucleus however is affected by the interaction of treatment and CCS. At brainstem level, the nucleus of the solitary tract and parabrachial lateral nucleus shows a similar pattern of activation since mice bearing SCC-XX show an increased neuronal activity at both nuclei. Furthermore, DEP-XX males show a higher Fos-Ir at the area postrema level. Our results indicate that SCC may modulate brain activity of nuclei closely involved in hydrosaline homeostasis.

Neural Circuit Physiology
Poster Number 173 | Session 2

"Changes in pallidal activity during cortical activation depend on striatal output in an animal model of Parkinson's disease"

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Excessive synchronization between basal ganglia and cortex is a hallmark of

Parkinsonís disease. The underlying mechanisms remain unclear. The aim of the present study was to determine whether striatal output drives oscillations in the parkinsonian GP during the condition known as "activated state". In order to do that, we recorded GP activity before and during the administration of an antangonist of NMDA receptors inside the striatum through reverse microdialysis. Synchronization of pallidal firing to the activated ECoG is increased in 6-OHDA rats across a wide range of frequencies, specially at low and beta frequencies. Importantly, pallidal coupling with cortical rhythms in control rats was negligible, indicating a truly anomalous nature of cortico-pallidal synchronization in the parkinsonian condition. Modulation of pallidal firing by the low frequency components of the activated ECoG was markedly reduced during AP-5 infusion. Striatal NMDA receptor blockade had less marked effects on beta synchronization. It reduced the number of neurons modulated by low but not high cortical beta rhythm. Thus, intrastriatal AP-5 infusion reduced cortico-pallidal synchronization in 6-OHDA rats in a frequency dependent manner.

Neural Circuit Physiology
Poster Number 174 | Session 3

"Stable neural ensemble recordings in awake head-fixed rat"

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We present an easy to implement Head-fixed setup for multielectrode recordings in awake rats. Animals were implanted with an "X" shaped aluminum piece. During the recording sesions, the implant was screwed in four points to two acrylic pieces, which are in turn fastened to the ear bars holders of a standard stereotaxic apparatus. The setup was implemented successfully in a classical conditioning task: a water deprived rat was presented with 1 KHz tones, which predicts reward (0.06 ml of sweetened water), and 8 KHz tones not paired with reward. Simultaneous recordings with tetrodes were made in Prefrontal Cortex and Ventral Tegmental Area. Stable (up to 40 minutes) single unit activity of many neurons in both areas was obtained. The shape of the implant can be easily modified for recordings in different brain areas. Also, implementation of other recording techniques like fast-cyclic voltammetry,microinjection and calcium imaging could be achieved.

Neural Circuit Physiology
Poster Number 175 | Session 1

"A switch from integrative to orthogonal input processing by hippocampal adult-born neurons"

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Immature granule cells (GCs) born in the adult hippocampus present higher excitability than mature GCs, and require weaker inputs to be recruited. These properties suggest that while mature GCs are presumably more selective in their responses, immature GCs could be recruited by different inputs, allowing integrations to occur. To test this hypothesis, we worked on hippocampal slices from adult mice injected with a retrovirus to express RFP in newborn GCs. We monitored GCs activity by loading them with the calcium-sensitive dye OGB-1 AM. Two electrodes were placed to stimulate independent medial perforant path inputs at different intensities. Under these conditions, the majority of mature GCs responded to only one of the two stimuli, but most immature GCs were recruited by both. Blocking GABAergic inhibition increased mature GCs input integration but did not modify immature GCs. Our results suggest that immature GCs integrate different sources of information whereas mature neurons process incoming information in an orthogonal manner. These functions could imply a specific role of both GCs populations in learning and memory, such as temporal associations and dissociations of space and object information.

Neural Circuit Physiology
Poster Number 176 | Session 2

"Serotonergic system involvement during states of body hypertonicity"

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Our previous studies demonstrated the serotonergic (5HT) system participation in the inhibition of sodium appetite induced by peritoneal dialysis. In this model, Fos expression, used as a marker of neuronal activity, decreases when animals are sodium depleted and increases when animals are in the process of restoring sodium balance through ingestion, in serotonergic dorsal raphe nucleus (DRN) cells.

Pharmacological and lesion studies have also demonstrated the contribution of 5HT system in renal sodium excretion. Taken together, the evidence suggests that body sodium levels modulate the activity of the 5HT-DRN system. Our aim was to study the 5HT system involvement during a hypertonic sodium overload (HSO). The HSO was performed by a sc injection of 0.6 ml/100g bw of 2M NaCl solution. Ninety minutes later the rats were subjected to immunohistochemical detection of Fos and 5HT. Our results indicate that the HSO significantly increased 5HT-DRN cells activity and also induced a significant increase in Fos expression along brain stem, lamina terminalis and central extended amygdala nuclei. In summary, a HSO itself can increase the activity of 5HT-DRN cells together with specific brain cell groups, possibly to modulate sodium balance and avoid an extracellular volume expansion.

Neural Circuit Physiology
Poster Number 177 | **Session 3**

"Neural basis of goal directed behaviour"

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The main objective of our project is to identify mechanisms that sustain episodes of animal locomotion following a brief sensory stimulus. The ability of the nervous system to sustain information online beyond the temporal pattern of sensory stimuli is an essential feature to achieve goal directed behaviour, a behaviour that is fundamental from invertebrates to humans. In the medicinal leech, an identified cascade of excitation that initiates swimming has been traced from mechanosensory neurons that receive the tactile stimulus to the swim oscillator circuit that innervates motoneurons. However, the mechanisms that maintain swimming are less clear. In this network a neuron identified as 204 constitutes a characterized scenario to study sustained activation. When swimming is elicited by sensory stimuli, cell 204 is excited and its activity is sustained throughout the entire swim episode that can last several seconds. Cells 204 are considered the excitatory drive for the swim central pattern generator. What is the source of this sustained activation? Because it is well characterized, the nervous system of the leech offers peerless experimental setting to answer this question. In the poster we will discuss the experiments and strategies we are planning for this project.

Neural Circuit Physiology
Poster Number 178 | Session 1

"Spatio-temporal pattern of efferent connectivity of adult-born dentate granule cells"

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Adult neurogenesis provides a continuous pool of new dentate granule cells (DGCs) to the hippocampus. The maturation process of adult-born DGCs takes from six to eight weeks. Whereas the development of afferents to new DGCs has been extensively characterized, the pattern of efferent connectivity remains poorly understood. In the adult hippocampus, mossy fibers grow through the hilus to reach pyramidal cells in the CA3 region. This led us to hypothesize that mossy fibers of immature DGCs primarily innervate proximal GABAergic interneurons and lately they synapse on pyramidal cells. To test this hypothesis we retrovirally transduced adult-born DGCs to express channelrhodopsin-2 (ChR2). Newborn DGCs were stimulated by flashes of blue light in order to characterize their connectivity by recording on putative postsynaptic targets in acute slices. Preliminary recordings on pyramidal neurons suggest that 6 to 8-week-old DGCs project functional monosynaptic contacts as evidenced by evoked glutamatergic EPSCs. Moreover, light stimulation also evoked disynaptic IPSCs on pyramidal neurons. These results suggest that adult-born DGCs perform functional synaptic connections on CA3 neurons by 6 weeks. We are currently investigating the functional output connectivity of younger DGCs.

Neural Circuit Physiology
Poster Number 179 | Session 2

"ABAergic regulation of the cardioinhibitory response in the crab Neohelice granulata"

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Upon the presentation of a visual danger stimulus (VDS) as well as a relatively harmless visual stimulus (OFF) to the crab Neohelice granulata, heart arrest or bradycardia were elicited. We further investigated the regulation of this cardioinhibitory response

upon visual stimulation in the presence of GABA antagonists. The administration of the non-competitive antagonist of GABA-A receptors, Picrotoxin, partially reduced the heart arrest, while no effect was observed using OFF. However, the administration of the competitive antagonist Bicuculline showed no effect on the cardioinhibitory response. In order to investigate the neuroanatomical substrate of this response, histological and immunohistochemical methods were used to examine the localization of the cardiac ganglion and GABA-like immunoreactivity within the cardiac system. Haematoxylin-eosin and Masson trichrome stains facilitated the identification of a distinct region compatible with the cardiac ganglion. GABA-like immunoreactivity detected in this region is originated from a single pair of processes close to the ganglion, giving rise to varicose branches that surround the somas of large neurons. Taken together, these findings support the idea that GABA mediates the extrinsic inhibition in this cardiac response.

• NEUROCHEMISTRY AND NEUROPHARMACOLOGY

Poster Number 180 | **Session 3**

"Potentiation of the homomeric rho1 GABAc receptor function by H2O2 is mediated by the intracellular cysteine residue Cys-364"

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Reactive oxygen species (ROS) are generated as by products of the cellular oxidative metabolism and secondary to the activation of NMDA and AMPA receptors. They are implicated in signalling pathways and oxidative stress, particularly during normal aging and neurodegenerative disorders. Numerous neurotransmitter receptors and ion channels are modulated by ROS. We reported previously that H2O2 significantly potentiates GABAc receptor function. In the present study we characterized the mechanism underlying this modulation. Homomeric GABA rho1 receptors were expressed in Xenopus laevis oocytes and GABA-evoked chloride currents recorded by two-electrode voltage-clamp in the presence or absence of H2O2. Potentiation of GABA rho1 receptors by H2O2 was dose-dependent, reversible, voltage independent and strongly depended on GABA concentration. GABA rho1 receptors subunits contain three cysteine residues: two extracellular at the cys-loop (C177 and C191) and one intracellular (C364). Chemical protection of cysteine residues by the membrane-permeable sulfhydryl alkylating reagent (NEM) prevented ROS potentiation. Furthermore, site directed mutagenesis of the

intracellular cysteine by alanine (C364A) rendered receptors insensitive to H2O2, suggesting a single modulatory site.

Neurochemistry and Neuropharmacology
Poster Number 181 | Session 1

"Pharmacology of nicotinic receptors from C. elegans muscle"

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Caenorhabditis elegans has emerged as a model organism useful for drug screening. It expresses two types of muscle nicotinic receptors (AChRs): levamisole-sensitive (L-AChR) and nicotine-sensitive (N-AChR). L-AChRs are targets of anthelmintic drugs, which act as specific and potent agonists. We here explore activation of both AChR types by the nematocide drug oxantel. Single-channel recordings from C. elegans muscle cultured cells show that oxantel elicits openings with amplitudes similar to those of L-AChRs. However, the frequency of opening is significantly reduced with respect to that observed in the presence of ACh, indicating reduced activation. In whole-cell recordings from cells expressing N-AChRs, peak currents elicited by oxantel are 45% of those elicited by ACh. Molecular docking studies show binding of oxantel to both types of AChRs, with similar binding energies but different orientations at the binding pocket. We conclude that oxantel behaves as a partial and non selective agonist of both L- and N-muscle AChRs, thus showing a different pharmacological profile compared to that of typical anthelmintic agents. These results contribute to the understanding of the pharmacology related to neuromuscular transmission in the model organism.

Neurochemistry and Neuropharmacology
Poster Number 182 | Session 2

"Stereospecific modulation of α9α10 nicotinic cholinergic receptors by L-ascorbic acid"

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The activation of a9a10 nicotinic receptors in cochlear hair cells can ameliorate

acoustic trauma. Therefore, enhancing a9a10-mediated currents through the use of a positive modulator may be helpful in the prevention or treatment of noise-induced hearing loss. We have previously characterized L-ascorbic acid (L-ASC) as a positive modulator in a9a10 injected X. laevis oocytes by two-electrode voltage-clamp recordings. In the present work we aimed to analyze the underlying mechanism of this potentiation. Oxidized L-ASC blocked acetylcholine(ACh)-evoked responses at a 3mM concentration (72±5% block at 1μM ACh, n=3), suggesting that reducing L-ASC is the active compound. Alternatively, the equally reducing stereoisomer D-ASC also had a blocking effect (55±10% block at 1μM ACh, n=3), indicating that a stereospecific interaction is also necessary. To test if a redox mechanism is involved in L-ASC potentiation we mutated the extracellular residues CC192/193SS. This substitutions did not abolish L-ASC potentiation (92±10% potentiation at 1mM Ach, n=6), ruling out the participation of these residues in the mechanism. Altogether, our results indicate that ASC potentiates a9a10-mediated responses through an allosteric and/or redox mechanism which does not involve C192-C193 disulfide bond.

Neurochemistry and Neuropharmacology
Poster Number 183 | Session 3

"Neonatal X radiation: hippocampal morphology and anxiety state"

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Ionizing radiation (Rx) is a physical agent broadly used in therapeutic medicine, being the developing Central Nervous System particularly vulnerable. Since a decrease in anxiety levels has been observed in neonatally irradiated rats, which could underlie several memory changes, and considering an implication of GABA in both mechanisms, the aim of the present work was to address the involvement of the GABAergic system in these Rx-induced alterations. Moreover, since a relationship between dendritic spines, GABAergic neurotransmission and memory processes have been postulated, the density of dendritic spines in the hippocampus (Hip) was also assessed. Male Wistar rats were irradiated with 5 Gy of X rays in their cephalic ends between 24 and 48 hours after birth. GABAA receptor binding assay and morphological assessment of mushroom dendritic spines were performed in Hip of 30-days-old rats. Results show that the decrease in anxiety levels observed in

irradiated rats is correlated with an increase of GABAA receptor density observed in synaptosomes from Hip of irradiated rats. Moreover, the increase in the number of mushroom dendritic spines Faloidine (+) in CA1 region of the Hip of irradiated group could explain, at least in part, the increase of GABAA receptor density.

Neurochemistry and Neuropharmacology
Poster Number 184 | Session 1

"Neuroprotective effect of a new rhEPO analogue with low erythropoietic activity"

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Although recombinant human erythropoietin (rhEPO) is a promising therapeutic agent for neuroprotection, its use might lead to hematological side-effects such as red cell mass increase and platelet aggregation. Using a rat model of traumatic brain injury (TBI), the present study explored the neuroprotective potential of a novel physicochemically-related rhEPO analogue with low erythropoietic activity (rhNEPO). Male Sprague-Dawley rats were subjected to a moderate focal cortical contusion. Following TBI, animals were treated with daily ip. applications of rhNEPO or rhEPO. After 3 days, rats were killed in order to measure contusion volume, hippocampal neuronal loss and neural apoptosis. Hemoglobin was also quantified before TBI and after therapy. Treatments with rhNEPO or rhEPO were able to slightly reduce the damaged area without differences in the contusion volumes. Interestingly, both molecules significantly reduced the loss of CA2-3 hippocampal neurons in about 50% when compared with placebo and they prevented apoptosis in the pericontusional area. Besides, only rhEPO increased hemoglobin, suggesting that rhNEPO might be a good candidate for TBI therapy because it shows the same neuroprotective action than rhEPO but does not elicit erythropoiesis significantly.

Poster Number 185 | Session 2

"Nerve terminal alterations in frontal cortex from rats with experimental autoinmune encephalomyelitis (EAE)"

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Multiple sclerosis (MS) is a human inflammatory demyelinating disease of the central nervous system that leads to motor, sensory, and cognitive deficits. Although it has been classically considered a white matter pathology, cortical lesions has increasingly received attention as they may contribute to disease progression and emergence of cognitive deficits. EAE is the classical model that mimics many of the clinical and pathological features of MS. While EAE research has been mostly focused on spinal cord inflammation/demyelination, the extent of cortical alterations are still not completely known. Besides corroborating the previously found diminution in Ca2+-dependent glutamate release from frontal cortex synaptosomes of EAE rats, herein we show that this alteration is accompanied by increased levels of the GABA synthesizing enzyme GAD65/67, without changes in the content of glutamatergic and GABAergic terminals. Concomitantly, differential activation upon stimulation of kinases was observed. Moreover, these changes are reversed when the animals are completely recovered from the clinical signs of the disease. These data indicate that the machinery of neurotransmitter release is affected in EAE, and shed light into the mechanism of neuronal dysfunction in the frontal cortex.

Neurochemistry and Neuropharmacology
Poster Number 186 | Session 3

"Effect of vanadium exposure through lactation: Biochemical and histological studies in neonate wistar rats brain"

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We have shown that administration of sodium metavanadate (NaVO3) to adult rats resulted in high lipid peroxidation levels, astrogliosis, heat shock protein (Hsp 70) expression, oxide nitric syntase activation, reactive oxygen species formation and

alteration of the oxidative defence system in hippocampus (Hc) and cerebellum (Cer). Since V was reported to be present in milk of V-treated nursing rats, the aim of this work was to study the effects of NaVO3 on the CNS of neonate rats exposed through lactation. 8 Wistar rat litters were randomly assigned to the following groups: V-treated: offspring of dam i.p. injected with 3 mg/kg bw of NaVO3 from the 10th to the 21st post natal day. Control: offspring of dam injected with saline solution. Brain areas were removed for lipid peroxidation assay by the thiobarbituric acid (TBA) reaction, and brains were used for: NADPHd histochemistry and anti-Hsp 70 and anti-GFAP immunohistochemistry. The relative optical density of the NADPHd stained layers and the GFAP (+) astrocyte surface area in Cer and Hc were measured. Although TBA levels and NADPHd activity didnít show differences between experimental and control groups, astrogliosis and an Hsp 70 activation was detected in Hc and Cer of V-exposed pups.

Neurochemistry and Neuropharmacology
Poster Number 187 | **Session 1**

"Latrunculin A in the Nac inhibited the expression of motor sensitization and increased AMPA receptors after cocaine in chronically stressed animals"

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Drug addiction is associated with long-term changes in the synaptic function, including the actin cytoskeleton. There is evidence about the proactive influence of stress on drug addiction. This study investigated whether the adaptations induced by repeated cocaine in the actin cytoskeleton, the surface expression of AMPA receptors (AMPAR), and the size of postsynaptic density (PSD) in the nucleus accumbens (NAc), occur and are relevant in a repeated immobilization stress-induced model of cocaine sensitization in rats. The levels of F-actin, actin-binding protein (ABP) and AMPAR were determined by Western, and the size of PSD measured by electron microscopy. The effects of latrunculin A and CNQX in the NAc were studied on motor activity. In the NAc, a decrease in p-cofilin and p-cortactin, concomitant to an increase in AMPAR and the size of PSD, was found in the stress plus cocaine group. Latrunculin A in the NAc inhibited motor sensitization and increased AMPAR. This study shows that stress-induced changes in the actin cytoskeleton, the size of PSD and AMPAR partly parallel the alterations elicited by sensitization to repeated cocaine and that actin dynamics regulate

AMPAR expression in the NAc and underlie the expression of cross-sensitization between stress and cocaine.

Neurochemistry and Neuropharmacology
Poster Number 188 | **Session 2**

"Involvement of the gabaergic system in the induction of the neuropathological alterations in experimental autoinmune encephalomyelitis (EAE)"

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EAE is an inflammatory and demyelinating disease used as model of multiple sclerosis, which is characterized by a strong cellular response against myelin antigens and neuronal compromise. In order to better understand the events that lead to the progressive neuronal dysfunction, we previously explored the contribution of glutamate release in frontal cerebral cortex from rats with EAE. We found a significant decrease of glutamate release regulated by GABA from synaptosomes of EAE rats during the acute stage of the disease. In order to determinate the GABA system participation in EAE progression, now we evaluated the effect of GABAergic agents with different action mechanisms on disease induction, proliferation of T cells, and glutamate release. The results indicate that meanwhile treatment of rats post-EAE induction with GABAA agonists (diazepam, muscimol) prevented the development of the disease, a GABAB agonist (baclofen) worsened clinical signs. Additionally, the GABAA agonists added in vitro inhibited T-cell proliferation and increased glutamate release but the GABAB agonist acted stimulating T-cell proliferation and decreasing glutamate release. In conclusion, these results indicate that the GABAergic system modulate the EAE development depending on the activated pathway.

Neurochemistry and Neuropharmacology
Poster Number 189 | **Session 3**

"Homomeric (NO)1 GABAc receptor function is potentiated by S-nitrosylation"

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Nitric Oxide (NO) is a gas messenger produced in neurons that can modulate the activity of neurotransmitter receptors. We have previously shown that NO potentiates GABAc receptor function. Earlier studies reported the S-nitrosylation of cysteine residues in NMDA and GABAa receptors by NO. Thus, we examined if GABAc receptors can undergo analogous modifications. Homomeric rho1 GABAc receptors (GABAr1R) were expressed in oocytes and GABA-evoked responses electrophysiologically recorded in the presence or absence of the NO donor DEA/NONOate (DEA). GABAr1 responses were significantly enhanced in a dose-dependent, fast and reversible manner by DEA. The specific NO scavenger CPTIO prevented these effects. Each GABAr1R subunit contains three cysteine residues, namely: two extracellular at the cys-loop (C177 and C191) and one intracellular (C364). Site directed mutagenesis of the C177 and C191 renders non-functional receptors, but C364 can be safely exchanged by alanine. The chemical protection of these cysteine residues by sulfhydryl reagents (NEM, MTSEA) prevented DEA effects on GABAr1R. Meanwhile, wild type receptors and GABAr1C364AR were similarly potentiated by DEA. These results suggest that NO enhances GABAc receptor function through S-nitrosylation occurring at the cys-loop.

Neurochemistry and Neuropharmacology
Poster Number 190 | Session 1

"Role of Nitric Oxide in the alterations in HPA Axis induced in prenatally stressed offspring"

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A great amount of evidence has shown that prenatal restraint stress (PRS) induces alterations in hypothalamic-pituitary-adrenal (HPA) axis responsiveness. Evidence has pointed out to a role of the hippocampus in modulating the response to stress. The aim of the present work was to find a possible link between the alterations induced in nitric oxide (NO) and the reported alterations in HPA axis. Methods: pregnant

Wistar rats were restrained three times a day during the last week of pregnancy. Offspring was sacrificed at early ages and at adulthood for corticosterone (cort) determination and NADPH diaphorase histochemistry. Anxiety-like behaviour was evaluated in an open field. Results: We found an increase in immovility time in the open field in PRS rats, together with a decrease in cort secretion and NADPH diaphorase staining. To evaluate a link between NO and cort, animals were treated with an nNOS inhibitor prior to sacrifice. We found that nNOS inhibition led to a decrease in cort secretion in controls, but not in PRS rats. Discussion: Alterations in HPA axis due to PRS has previously been established. The results found here point to a role of nNOS in the modulation of corticosterone secretion, and to a participation of NO in PRS induced alterations.

Neurochemistry and Neuropharmacology
Poster Number 191 | **Session 2**

"In search of a specific agonist for the α9α10 nicotinic cholinergic receptor"

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The a9a10 nicotinic acetylcholine (ACh) receptor (nAChR) mediates synaptic transmission between efferent olivocochlear neurons and outer hair cells of the cochlea. This receptor differentiates itself from other nAChRs with its distinct pharmacological profile. In light of the proposed role of this system in protection against acoustic damage (Taranda, Plos Biology, 2009), it is desirable that specific, efficacious agonists be identified. In this study, we used molecular modeling to design a series of ACh analogs which we subsequently tested in Xenopus oocytes expressing the a9a10 receptor by electrophysiological recordings with the two-electrode-voltage-clamp technique. Preliminary results revealed that four of the original compounds tested proved to be agonists. Of these, one compound, 2-((3-methoxy-3-oxopropanoyl)oxy)-N,N,N-trimethylethanaminium iodide, produced a maximum response 150% of that elicited by ACh, but with an EC50≥508.2μM (EC50ACh=16.24±1.07 μM). We have subsequently designed a second generation of drugs based on this lead compound in an effort to maintain this high efficacy while improving the affinity. Interestingly, our results suggest that the a9a10 receptor can be more efficaciously activated by compounds other than the native agonist.

Neurochemistry and Neuropharmacology
Poster Number 192 | Session 3

"*Preproenkephalin knockout mice did not show sensitization to the behavioral effects induced by cocaine and failed to show the cocaine-induced increases in ERK activation and AMPA cell surface expression in striatum and nucleus accumbens*"

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Opioid receptors and endogenous opioid peptides, mainly enkephalin, are largely distributed in the mesolimbic system. However, their contribution to cocaine - induced sensitization on behavioral and associated molecular parameters has been poorly studied. Male C57B/6J wild type (WT) and preproenkephalin knockout (KO pENK) mice were daily treated with cocaine (15mg/Kg i.p.) and vehicle for 9 days followed by a cocaine challenge (7,5mg/Kg) on days 15 and 21 where the locomotor activity was measured. In addition, male C57B/6J WT mice received the same treatment but 30 min. before each cocaine injection, the animals were administered with naloxone (1mg/Kg s.c.). The day 21 the nucleus accumbens, striatum, hippocampus and prefrontal cortex were dissected and GluR1, dopamine transporter, ERK and CREB levels were measured by western blot. Penk KO mice did not show sensitization to the behavioral effects induced by cocaine and failed to show the cocaine-induced increases in ERK activation and AMPA expression evidenced in the WT mice. Wild type mice pretreated with naloxone did not show the cocaine- induced molecular changes. These results indicate that preproenkephalin system is strongly involved in the long-term plasticity underlying sensitization to cocaine.

Neurochemistry and Neuropharmacology
Poster Number 193 | Session 1

"BDNF and NR2B/PSD-95 complex modulation by atorvastatin leads to an improvement in the spatial learning and memory after transient focal cerebral ischemia"

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Cerebral ischemia is a cerebrovascular disease mainly generated by the occlusion of middle cerebral artery. The most sensitive areas to this injury are cerebral cortex and hippocampal regions, generated neuronal death and deficits in learning and memory. Statins, HMG-CoA reductase selective inhibitors have shown a protective role in cerebral ischemia but its mechanisms are not well understood. Main aim was to evaluate the effect of atorvastatin on proteins involved in synaptic plasticity in ischemic rats. Initially, male rats were subjected to focal ischemia t-MCAO, subsequently received treatment with atorvastatin 10mg/kg at 6 hours post-ischemia for 3 days in a daily doses. The behavioral test was made by Morris water maze (MWM). Animals were perfused or decapitated to realize cellular and biochemical analysis respectively. Our results show cerebral recovery (tested by Nissl, MAP-2, NeuN and fluorojade), reassociation of NR2B/PSD-95 complex and recovering of the BDNF protein levels in cerebral cortex and hippocampus. Finally, ATV treatment improves learning and memory function to 15 days post-ischemia. All data together suggests that early administration of ATV during three days protects the synapses and recovers the cognitive function in a late period after ischemia.

Neurochemistry and Neuropharmacology
Poster Number 194 | **Session 2**

"Melanin concentrating hormone (MCH) decreases presumed serotonergic neuronal activity in the dorsal and median raphe nuclei"

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Hypothalamic neurons that utilize melanin-concentrating hormone (MCH) as a neuromodulator project to the serotonergic dorsal (DRN) and median raphe nuclei (MnR). These nuclei are involved in the control of sleep. MCH-labeled tanycytes are present in these nuclei and are specialized in transporting substances from the CSF to the neuronal parenchyma, suggesting that MCH could be absorbed

from the CSF and produce a tonic effect on raphe neurons. Thus, the aim of the present study was to analyze the effects of microinjections of MCH into the lateral ventricle on the neuronal activity of raphe neurons. Adult rats (n=13) anesthetized with urethane were prepared for standard extracellular recording of DRN and MnR neurons. Thereafter, we analyzed the electrophysiological properties of the neurons after MCH (5 mcg) or saline microinjections into the lateral ventricle. MCH decreased the firing rate in 70% of the DRN recorded (from 3.3±2.9 to 1.7±1.6 Hz) and in 90% of the MnR neurons recorded (from 5.5±3.1 to 2.8±2.2 Hz). Most of the units were presumably serotonergic according to their electrophysiological properties. We conclude that MCH regulates the DRN and MnR neuronal activity and part of this effect could be mediated by volume conduction through the CSF.

Neurochemistry and Neuropharmacology
Poster Number 195 | Session 3

"The brain RAS is involved in the neuroadaptative responses induced by amphetamine in a two-injection protocol"

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A single or repeated exposure to psychostimulants induces long-lasting neuroadaptative changes. Different neurotransmitter systems are involved in these responses including the neuropeptide angiotensin II. Our study tested the hypothesis that the neuroadaptative changes induced by AMPH produce alterations in brain RAS components. Wistar male rats (250-300 g), pretreated with AMPH (5mg/kg, ip) were used 7 or 21 days later to quantified AT1 receptors by western blot and mRNA-Angiotensinogen (AOGEN) in caudate putamen (CPu) and accumbens nucleu (NAcc). In another group of animals treated in the same way, bearing intra-cerebral cannula, the locomotor activity was tested after AMPH challenge (0.5 mg/kg) injection. The animals received an AT1 blocker, losartan (8 ug/ul/side) or saline 5 min before the AMPH challenge. We found an increase of AT1 receptors density in both studied areas. AMPH modified the basal and challenge induced expression of mRNA-AOGEN in NAcc and CPu at 7 and 21 days after treatment. Finally The AT1 receptors blockade in NAcc increased the locomotor activity induced by AMPH challenge. Our results support the hypothesis for a key role of brain RAS in the neuroadadptaive changes induced by AMPH.

Neurochemistry and Neuropharmacology
Poster Number 196 | Session 1

"Neuroprotective properties of modafinil on methamphetamine-induced glial activation in mouse striatum"

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Modafinil (MOD) is a stimulant used as a cognitive enhancer and is also prescribed to treat psychostimulant addiction. Methamphetamine (METH) intake produces deleterious effects in brain areas. In order to determine if MOD was able to counteract METH-induced glial activation, mice were treated with a METH "binge" protocol (4x5mg/kg, i.p., 2h apart) co-administrated with MOD (2x90mg/kg, i.p., 1h before 1st and 4th METH injections). After two days, METH treated group exhibited striatal reactive microgliosis and enhanced GFAP immunostaining but normal levels of tyrosine hydroxylase (TH) and dopamine transporter (DAT). When METH was co-administered with MOD, glial activation values and TH and DAT levels were similar to those found in saline/MOD groups ($p < 0.001$). Six days after treatment, astrogliosis was still present in METH treated animals without signs of microgliosis ($p<0.001$) along with diminished TH and DAT levels ($p<0.05$). In MOD+METH group activated astroglia and no signs of microgliosis or altered levels of TH and DAT were found ($p<0,05$). Our results suggest that MOD managed to block glial activation by an early intervention in the temporal onset of METH-induced dopamine terminal toxicity. Grants: PICT 2007-1009/2008-2019, PIDRI-PRH 2007, PIP 11420100100072.

Neurochemistry and Neuropharmacology
Poster Number 197 | Session 2

"Differential expression of α1 subunit and ß2-3 subunit of GABA-A receptors in neonatal chick forebrain submitted to an acute stess. Modulation by noradrenaline hicks forebrain"

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Gamma-aminobutyric acid, the mayor inhibitory neurotrasmitter in the brain, mediates inhibition via GABA-A receptors(GABA-A R).The GABA-A R consist on five subunit and the assembly of different combinations allows the constructions of different types of GABA-A R, each having specific pharmacological and funtional properties.The mayority of GABA-A R are composed of two α subunit, two β subunit and one γ. The α subunit are associated with pre- and post-synaptic sites, while β subunit is associated only with post-synaptic sites.Ours previuos findings in 4-6-days-old chicks, showed difference on behavioral responses and on GABA-A R recruitment after Open Field and/or central administration of noradrenaline. In the present report, different doses of noradrenaline (0.0025;0.010;0.050;1.000 ug/ul)injected before an Open Field induced a differential expression of α1 subunit and β2-3 subunit of GABA-A R on cell surface, detected by immunoperoxidase, in brain tissue. We observed that α1 subunit is higher Optical Density due to increased number of labeled cells than β2-3 subunit. These results indicated that the differences observed between expression of both subunit corresponded to an increased of post-synaptic subunit expression.

Neurochemistry and Neuropharmacology
Poster Number 198 | Session 3

"Effects of α-and ß-thujone on anxiety behavior in neonatal chicks: involvement of GABAA receptors"

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Essential oils are mixtures of several volatile and aromatic substances predominantly composed of terpenes. They have various pharmacological properties, anesthetics, analgesics, gastric sedative agent that may have profound consequences on animal behavior. α-thujone and β-thujone are bicyclic monoterpenes with neuroactive properties due to an effect on neuronal intracellular signaling or by modulating ionic currents of ionotropic receptors. GABAAR can be modulated by various natural compounds such as flavonoids, glycosides, terpenes, among others. Alterations in the activity and density of GABAAR are involved in anxiety, fear, depression and chronic substance abuse. The aim of this paper is to study the effects of in vivo administration of α-thujone and β-thujone on anxiety behaviour

and the recruitment of GABAAR, in neonatal chicks. Terpenes icv administered (78 µmol per chick, ~0.24 mg/kg) induced an anxiogenic effect, observed as latency increased to ambulate and locomotor activity decreased in an Open Field of both experimental groups compare to control (saline).In addition, α-thujone only modulated synaptic inhibition through the decrease of flunitrazepam-sensitive-GABAAR recruitment during Open Field, avoiding the increment induced by acute stress.

• NEUROENDOCRINOLOGY AND NEUROIMMUNOLOGY

Poster Number 199 | **Session 1**

"The effect of sleep loss on the reproductive function of male rats"

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This study aims to evaluate the influence of selective sleep loss on sexual behavior and hormone levels as well as sperm quantity and viability in male rats. Sexually experienced rats were subjected to paradoxical sleep deprivation (PSD) for 96 hours or sleep restriction (SR) for 21 consecutive days. The control group (CTRL) was kept in their home cages throughout the experimental protocol. Sexual behavior was evaluated following the exposure of the PSD or SR paradigm and then the hormone and sperm variables were measured. The PSD was able to significantly decrease sexual behavior, but the SR group showed no effect compared to the CTRL group. With respect to their hormones, the PSD rats had a significantly lower testosterone levels compare to CTRL group. Sleep deprivation protocols did not change progesterone, follicle-stimulating hormone (FSH) or luteinizing hormone (LH) in relation to CTRL group. Regarding the semen analysis, both the PSD and SR groups presented a lower sperm viability compared to the CTRL group. However, the decrease in the number of live sperm was larger in the PSD group than in the SR group when compared to the CTRL rats. These findings demonstrate that sleep loss can promote marked changes in the reproductive system and particularly affect sperm viability.

Neuroendocrinology and Neuroimmunology
Poster Number 200 | Session 2

"Involvement of kinin B2 receptors in the development of the pilocarpine-induced epilepsy in female rats"

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Aims: Several studies indicate that in the central nervous system and peripheral tissues, estrogen regulates the expression of B2 receptor and reduces cytokine production and inflammatory responses. Accordingly, the present work aimed to investigate the expression of kinin B2 receptor in non-castrated and castrated female rats, submitted to the pilocarpine model epilepsy. Methods and Results: The animals were divided in four groups: OVX + SE ovariectomized female rats, which presented status epilepticus (SE); SE: intact female rats that presented SE; OVX ovariectomized female rats that received saline instead pilocarpine; SAL intact female rats that received saline instead pilocarpine. The results showed a decrease of immunoreactivity of kinin B2 receptor in the hippocampal formation during the acute and silent but not in chronic periods of this model in SE group when compared to SAL group. In contrast, the immunoreactivity in OVX + SE group was increased during the acute and silent periods when compared with OVX group. Conclusion: This study showed that the expression of kinin B2 receptor is modified in female rats during epileptogenesis.

Neuroendocrinology and Neuroimmunology
Poster Number 201 | Session 3

"Immunologic studies in a model of Autism Spectrum Disorders (ASDs)"

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Autism spectrum disorders (ASD) are a heterogeneous group of neurodevelopmental disorders defined behaviorally by abnormalities in social, verbal, and nonverbal communication. The etiologies of ASD are likely to be the result of a variety of

numerous genetic, neurological, environmental, and immunological interactions that lead to a general behavioral phenotype defined as ASD. It is widely agreed that a subset of patients with ASD show abnormal immunity and it has been proposed that autoimmunity may play a role in the pathogenesis of ASD. However, immune findings in ASD patients are often inconsistent mainly due to the heterogeneous, ill-defined subject groups. Rett Syndrome is a neurodevelopmental disorder caused by mutations in Methyl Cytosine Binding Protein 2 (MeCP2) and mouse models of Rett have been widely used for studying ASDs. Using this monogenic model, we propose to evaluate the role of altered immunity in the pathogenesis and/or maintenance of ASDs. For that purpose we will evaluate the presence of autoantibodies against brain proteins, as well as markers of neuroinflamation in mouse models of Rett, at early postnatal ages and after the symptoms appearance. These studies will help to further understand the pathogenic mechanism of ASDs.

Neuroendocrinology and Neuroimmunology
Poster Number 202 | **Session 1**

"Inhibitory role of Diazepam on autoinmune inflammation in rats with experimental autoinmune encephalomyelitis"

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Glutamate and GABA are the respective main excitatory and inhibitory neurotransmitters in the central nervous system and both may be involved in the neuronal dysfunction in neurodegenerative conditions. We have recently found that glutamate release was decreased in isolated synaptosomes from the rat cerebral cortex during the development of experimental autoimmune encephalomyelitis (EAE). We have evaluated the relevance of the GABAergic system in EAE by treating rats challenged for the disease with the GABA agonist diazepam. Administration of diazepam during six days starting at day 6 or 11 after EAE induction leads to a marked decrease of the disease incidence and histological signs in spinal cord. Cellular reactivity and antibody responses against the encephalitogenic myelin basic protein were also diminished. Beyond the effects of diazepam on the autoimmune, inflammatory response, we report also a positive effect on neurotransmission. Treatment with diazepam inhibited the previously described reduction in glutamate release in the frontal cortex synaptosomes from EAE animals. These data suggest that an endogenous inhibitory GABAergic system

within the immune system is involved in the diazepam effect on EAE and indicate that increasing GABAergic activity potently ameliorate EAE.

Neuroendocrinology and Neuroimmunology
Poster Number 203 | Session 2

"Ghrelin indirectly activates hypophysiotropic CRF neurons"

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Ghrelin is a stomach-derived hormone that acts on specific hypothalamic neurons and potently increases appetite. Also, ghrelin activates the CRF (Corticotropin-Releasing Factor)-producing neurons of the hypothalamic paraventricular nucleus (PVN) and, as a consequence, the neuroendocrine hypothalamic-pituitary-adrenal axis (HHA). However, the neural circuits mediating this activation are mostly uncharacterized. Here, we studied the circuits by which ghrelin activates the hypophysiotropic CRF neurons in vivo. We found that peripheral and central administration of ghrelin to wild type mice strongly activates the marker of cellular activation cfos in CRF-producing neurons. Also, ghrelin increase CRF mRNA expression in the PVN and the HHA axis at peripheral level. When directly administrated on the PVN, we found that ghrelin also activates the CRF-producing neurons and the HHA axis, without any significant effect on food intake. Unexpectedly, we found that CRF neurons do not express ghrelin receptor as indicated by in situ hybridization histochemistry and ghrelin binding studies. Thus, we conclude that ghrelin activates hypophysiotropic CRF neurons indirectly, likely via a local pre-synaptic mechanism.

Neuroendocrinology and Neuroimmunology
Poster Number 204 | **Session 3**

"Early maternal separation and stress during adulthood: Implications for glucocorticoid receptors and anxiety-like behavior under treatment with tricyclic antidepressant amitriptyline"

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During the development of the Central Nervous System, early environment promotes different pathways of expression of molecules and different neurocircuits in the brain which will determine diverging responses to stress in the adult. This involves two types of nuclear receptors for corticosteroid: mineralocorticoid receptor (MR) and glucocorticoid receptor (GR). They mediate in a coordinate manner the steroid control of HPA activity and behavior. The aim of this work was to measure the immunoreactivity of GR and MR in Hippocampus and the anxiety-like behavior after chronic variable stress (CVS) in rats that were previously early maternally separated (MS). During the CVS protocol in adulthood, animals were treated with antidepressant Amitriptyline (AMI) (10 mg/Kg i.p.). There was a rise in the number of GR positive cells in CA3 in response to CVS combined with MS or with AMI. Regarding MR, CVS evoked a rise in the number of positive cells in CA1 and CA3 ($p < 0,05$) which was prevented by AMI treatment in CA3. On anxiety-like behavior, CVS evoked a marked tendency to increase anxiety which turned significant when CVS was applied to MS rats ($p<0,05$). AMI exerted an anxiolitic effect on CVS animals ($p<0,05$) but could not reverse the effect of MS combined with stress.

Neuroendocrinology and Neuroimmunology
Poster Number 205 | **Session 1**

"Effect of maternal viral infection on postnatal development in a mouse model of autism"

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Autism is a developmental disorder characterized by impediments in communication and social interaction and by stereotyped behaviors. We hypothesized that there

is a developmental critical window in which maturation and consolidation of the neural systems responsible for these symptoms typically occur and that they are altered in the patients. Different environmental factors have been linked to autism, in particular, epidemiological studies showed an association between outbreaks of viral diseases and a higher incidence of autism. In this work we used a mouse model of autism: prenatal exposure to valproic acid (VPA) validated in the lab, and combined it with a model of maternal viral infection. Pregnant females were injected with VPA at GD13 and with PolyI:C at GD17. We evaluated the postnatal behavior in the 4 experimental groups. To control for unspecific effects on maternal care, we also evaluated maternal behavior during the first postnatal week. Female pups were sacrificed at different postnatal ages and their brains evaluated for glial activation by immunohistochemistry. This study will help us to identify early alterations in brain development and glial activation that lead to autism-related behaviors and to evaluate the effect of maternal infection on these alterations.

Neuroendocrinology and Neuroimmunology
Poster Number 206 | Session 2

"Cell death mediated apoptosis by a Candida albicans infection in central nervous system (CNS)"

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Neurocandidiasis is a serious form of bloodstream infection with 50% of mortality, associated with congenital or acquired immunodeficiencies. With the aim to explore the ethiopathogenic mechanisms involved in this mycosis, we developed a murine in vivo model. C57/Bl6 mice were injected iv with 5.105, 1.106 or 2,5.106 viable yeasts of C. albicans and the colonization assessed 4, 12, 24, 48 and 72h post-infection. Candida was able to impinge, get through the BBB and settle down in the brain parenchyma as was confirmed by the recovery of the CFU 4h after the microorganism administration. The immunostaining with anti-GFAP and anti-CD11b (Astrocytes and Microglia markers respectively), revealed the presence of both reactive astro- and microgliosis (IF). Interestingly, we detected neuronal degeneration associated to the infection (FJB/A-Cu-Ag stain) and a significant number of TUNEL+ cells. The local balance between pro- (IL-1ß and

TNF-α) and anti-inflammatory (TGF-ß and IL-10) cytokines indicated a break-up of the niche homeostasis promoting a Th1 profile (ELISA). This model reproduces human pathology and provides evidence not reported yet in support of neuronal degeneration and apoptosis+ and/or pyroptosis+ cells after a systemic induced infection with C. albicans.

Neuroendocrinology and Neuroimmunology
Poster Number 207 | **Session 3**

"Early retinal changes in an experimental model of Type 2 Diabetes characterized by postprandial hyperglycemia"

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The aim of the present study was to develop a model of type 2 diabetes in rats (combining diet-induced insulin resistance and a slight ß-cell secretory impairment) in order to study early retinopathic changes. Rats received drinking water and citrate buffer i.p. (Group 1), tap water with 30% w/v sucrose and citrate buffer i.p. (Group 2), tap water and streptozotocin (STZ, 30 mg/kg i.p., Group 3), and 30% sucrose and STZ (Group 4). At 6 and 12 weeks of treatment fasting and postprandial glycemia, fructosamine and serum insulin levels were assessed. In addition, i.p. glucose and insulin tolerance tests were performed. Retinal function, retinal morphology, retinal lipid peroxidation and NOS activity were also evaluated. At 6 and 12 weeks of treatment, animals of Group 4 showed significant differences in most metabolic tests as compared with the other groups. At 12 weeks of treatment, a significant decrease in the ERG a- and b- wave and oscillatory potential amplitudes, a significant increase in retinal TBARS levels, NOS activity, and glial fibrillary acidic protein in M¸ller cells were observed in Group 4. Only hyperglycemia resulting from the combination of a sucrose-enriched diet and STZ injection induced significant biochemical and functional retinal alterations.

Neuroendocrinology and Neuroimmunology
Poster Number 208 | **Session 1**

"Model of depression in rats: Effect of tianeptine on hipocampal GR

and MR expression, MR/GR balance, anxiety and hedonic behavior"

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The aim of this work was to determine the effect of chronic treatment with 10 mg/Kg of tianeptine on GR and MR expression, GR/MR balance in the dorsal hippocampus, anxiety and hedonic behavior in male adult Wistar rats separated from their mother as neonates and submitted to variable chronic stress for 24 days, which is considerate an animal model of depression. GR and MR levels were determined by immunohistochemistry in the layers CA1, CA2, CA3 and dentate gyrus of dorsal hippocampus. Plus Maze test was performed in order to calculate anxiety indexes. Sucrose intake and preference were used as anhedonic indexes. Our results show that maternal separation significantly decreased MR- positive cells in all layers of rats treated with vehicle, while the treatment with tianeptine reverted this effect; and MR/GR balance was decreased in the layer CA3. On the other hand, stress significantly increased the immunoreactive cells to GR in the layer CA2 and tianeptine increased GR levels in layer CA3. Anxiety indexes were not altered by any of the treatments. In the Sucrose Preference Test, stressed animals showed a tendency to reduce this index at the end of treatments. On the contrary, stressed and treated with tianeptine groups showed a decrease of anhedonic behavior.

Neuroendocrinology and Neuroimmunology
Poster Number 209 | **Session 2**

"Cold exposure activates Thyrotropin Releasing Hormone (TRH)-producing neurons in specific brain nuclei"

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TRH mediates cold-induced thermogenesis via both neuroendocrine and autonomic mechanisms. It is known that the TRH neurons located in the hypothalamic paraventricular nucleus (PVN) regulate the neuroendocrine thyroid axis. In contrast, what populations of TRH neurons mediate the activation of the sympathetic mechanisms induced by cold exposure is unclear. Here, we

systematically examined the distribution of TRH neurons activated in response to cold exposure throughout the adult rat brain. To map the activation TRH neurons, we used double immunohistochemistry for the marker of cellular activation, cfos, and the TRH prohormone. To further assess the functional consequences of the cfos expression in TRH neurons, we evaluated the preproTRH mRNA levels in brain micro-dissections where TRH is produced. Our analysis was focused on hypothalamic regions including the PVN, the pre-optic area, dorsomedial nucleus and the lateral hypothalamus, and areas of the medulla including raphe obscurus, raphe pallidus (RPa) and parapyramidal regions. Our data indicated that only the TRH neurons located in the PVN and the RPa are activated in animals exposed to cold. Thus, these groups of TRH neurons are the main candidates participating in cold-induced thermogenic mechanisms.

Neuroendocrinology and Neuroimmunology
Poster Number 210 | **Session 3**

"Study of the interaction between ghrelin and high fat diet-induced acute food intake in mice"

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Ghrelin is a stomach-derived hormone that acts on homeostatic and hedonic brain centers to increase food intake. We have shown that ghrelin administration affects the rewarding value of high fat diet. However, it is currently unclear how the neural circuits activated by either ghrelin or high fat diet (HFD) interact to modulate eating. Here, we quantified acute food intake in wild type mice injected with ghrelin and exposed to either HFD alone, regular chow alone or a combination of both diets. Also, we performed immunohistochemistry for cfos in the mouse brains to map the neuronal circuits activated in each experimental condition. We found that i-ghrelin increases food intake of regular chow; ii-HFD alone strongly activates food intake; and ii-ghrelin fails to further affect HFD intake or short-term HFD preference. Also, we found i-ghrelin increases cfos expression mainly in hypothalamic nuclei; ii-HFD intake increases cfos mainly in centers of the mesolimbic pathway; and iii-the combination of HFD ingestion and ghrelin administration increase cfos expression in both hypothalamic and mesolimbic pathways. Thus, we conclude that HFD potently induces food intake due to the activation of hedonic centers, which override the acute effects of ghrelin on eating.

• SENSORY SYSTEMS

Poster Number 211 | Session 1

"Neuroprotective effects of brief ischemia pulses in an experimental model glaucoma"

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Glaucoma is a leading cause of acquired blindness which may involve an ischemic-like insult to retinal ganglion cells and optic nerve head (ONH). We investigated the effect of a weekly application of brief ischemia pulses (ischemic conditioning) on the retinal damage induced by experimental glaucoma. Glaucoma was induced by weekly injections of chondroitin sulfate (CS) in the rat eye anterior chamber. Retinal ischemia was induced by increasing intraocular pressure to 120 mmHg for 5 min; this maneuver started after 6 weekly injections of vehicle or CS and was weekly repeated in one eye, while the contralateral eye was submitted to a sham procedure. Glaucoma was evaluated in terms of: intraocular pressure (IOP), retinal and visual pathway function (electroretinogram (ERG), and visual evoked potentials (VEPs)) and histology of the retina and ONH. Retinal thiobarbituric acid substances levels were assessed as an index of lipid peroxidation. Ischemic conditioning significantly preserved ERG, VEPs, retinal and ONH structure from glaucomatous damage, without changes in IOP. Moreover, ischemia pulses abrogated the increase in lipid peroxidation induced by CS. These results suggest that induction of ischemic tolerance could constitute a new therapeutic strategy in glaucoma treatment.

Sensory Systems

Poster Number 212 | Session 2

"Morphological and physiological characterization of columnar neurons from the second optic ganglion of the crab Chasmagnathus granulatus"

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Crabs are highly visual animals, they have well-developed compound eyes located on mobile stalks containing an important part of their nervous systems. Just bellow the retina, there are three serially arranged retinotopic optic ganglia. There are giant neurons in the third optic ganglion (LGs) that command the crabís escape response to a visual danger stimulus. This response declines after a few stimulus presentations giving rise to a short or long term memory depending on the training protocol. This behavioural modification is a consequence of plastic changes in LGs neurons. However, the changes induced by massed training occur in columnar elements that are presynaptic to the LGs neurons. The aim of this work is to study the presynaptic site of plasticity. To stain these presynaptic columnar elements, we applied tracers onto the first optic ganglion in order to stain columnar neurons that project from this ganglion to the second one. We found that these columnar neurons arborize in three different horizontal layers. Studies with calcium imaging showed that the response of these columnar neurons does not decline with successive stimulus presentations. This result indicates that the site of plasticity is between these columnar neurons and the LGs.

Sensory Systems
Poster Number 213 | **Session 3**

"Study of visual pathways in an experimental model of glaucoma in rats"

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Glaucoma is a leading cause of blindness, characterized by progressive loss of visual functions, retinal ganglion cell (RGCs) death and optic nerve head atrophy. Glaucoma has been conceived as a disease limited to the eye, but axons of RGCs have extraorbital and intracranial components. The aim was to study retinal projections in an experimental model of glaucoma in rats. Glaucoma was induced by weekly injections of chondroitin sulfate in the eye anterior chamber for 15 weeks. Visual pathway activity was analyzed through visual evoked potential recordings (VEPs), and major retinal projection areas (superior colliculus (SC), lateral geniculate nucleus and suprachiasmatic nuclei) through anterograde transport of cholera toxin β-subunit (CTB). In the glaucomatous visual pathway, the VEP N2-P2 component amplitude and transport of CTB from retina to all areas examined were significantly reduced, compared to vehicle. Furthermore, CTB was significantly

accumulated in the optic nerves of glaucomatous eyes and glial immunoreactivity was significantly increased in the contralateral SC to glaucomatous eye. In sum, these results indicate that glaucoma induces significant alterations in extraocular visual pathway, and might cause trans-synaptic degeneration in retinal projection areas.

Sensory Systems
Poster Number 214 | Session 1

"Neurogenesis in the zebrafish retina: glutamatergic control of cell proliferation"

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The retina of zebrafish grows throughout animalís life from an intrinsic germinal region at the periphery of the retina called ciliary marginal zone (CMZ). This animal model allows studying cell proliferation and differentiation processes to generate new cell types of the adult retina. On the other hand, glutamatergic transmission from photoreceptors to bipolar cells (BC) and from BC to ganglion cells (GC) mediates light processing and information transmission to the visual brain centers. We blocked the depolarization of ON BC, which are activated in response to light, through an agonist (L-AP4) of mGluR6 receptors, whose activation by glutamate hyperpolarizes ON BC. We also used an antagonist of AMPA receptors DNQX, to block excitatory synapses between photoreceptors and OFF BC and between BC and all GC (although part of the response is NMDA receptor-mediated). DNQX treatment significantly increased proliferating cell number (measured by BrdU incorporation in the CMZ). Moreover, a still non significant increase of L-AP4 in cell proliferative activity has been observed. Therefore, these and previous results might indicate a relevant role of glutamate (and hence light and dark signals) in regulating mitotic activity and cell addition for retinal growth in the adult zebrafish.

Poster Number 215 | **Session 2**

"The Role of Visual Cues in Auditory Distance Perception"

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In humans, multisensory interaction is important for improving the detection of stimuli of different nature and reduce the variability of response. In some cases, the information collected by a sense can affect other perceptual modalities. It is known that the presence of visual information affects the auditory perception in the horizontal plane (azimuth) but there are few researches that study the influence of this information in the auditory distance perception (ADP). In general, the data obtained from these studies are contradictory and do not completely define the way in which visual cues affect the perception of the apparent distance of a sound source. In the present work, psychoacoustic experiments were performed considering ADP in humans by including and excluding visual cues. The data shows that the apparent distance from the source is affected by the presence of visual information and that subjects can store in their memory a representation of the environment that later improves the perception of distance.

Sensory Systems
Poster Number 216 | **Session 3**

"The exposure to an enriched environment prevents acute visual ischemic injury in adult rats"

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Retinal ischemia may provoke blindness. There is no effective treatment for retinal ischemic damage. Recent evidence supports that exposure of adult amblyopic rats to an enriched environment (EE) promotes full visual acuity recovery. We analysed the effect of exposure to an EE in an acute retinal ischemia model. Adult male Wistar rats were exposed to a standard environment (SE) or EE. EE consisted of big cages with food hoppers, wheels and different objects repositioned once/day and fully substituted once/week. After 3 weeks of exposure to SE or EE, unilateral

ischemia was induced, and afterwards, animals were returned to their respective environment for 2 weeks. Retinal function (electroretinography, ERG) and histology, anterograde transport to the superior colliculus (SC) and glial reactivity in the SC (GFAP staining) were analysed. In control animals, ischemia induced a significant decrease in ERG a- and b- wave amplitude and retinal ganglion cell (RCG) loss, GFAP staining increase in the SC and a decline in anterograde transport. The exposure to an EE partially prevented retinal dysfunction, RCG loss and GFAP stain in the SC. Anterograde transport was fully preserved. These results suggest that the exposure to an EE could become a new strategy for retinal ischemia treatment.

Sensory Systems
Poster Number 216 | **Session 3**

"Perception of principal pitch in asymmetrical vibratos: a new experiment and revision of models"

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Vibrato is a performing technique widely used by singers in all cultures, consisting in a quasi-periodic frequency modulation. Despite multiple frequencies are present in a vibrato note, when listening to a singer it is possible to extract a single pitch, called principal pitch. This principal pitch is equal to the geometric mean of the instantaneous frequency (f0) if the frequency modulation profile is symmetric, but it is still a matter of debate what is the principal pitch when frequency profile is nonsymmetric. Although exists evidence pointing to a "stability-sensitive weighting" mechanism which gives less weight to segments of the sound where f0 is changing rapidly, it is not clear a) the criteria used to assign weights and b) the way in which this is implemented by the auditory system. We present i) results from experiments of pitch perception for asymmetric vibratos as a function of the rate of change of frequency and ii) a comparison between predictions of several pitch perception models for our data and data of previous works.

Sensory Systems
Poster Number 217 | **Session 1**

"Distal axonopathy of the visual pathway in experimental diabetes"

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Diabetic retinopathy (DR) is a leading cause of acquired blindness. Visual function disorders have been demonstrated in diabetics with very early retinopathy or even before the onset of retinopathy. Although DR has long been recognized as a vascular disease, it is becoming increasingly clear that retinal ganglion cells (RGC) are also affected by diabetes. The aim of the present work was to analyze the visual pathway in an early step of experimental diabetes. Diabetes was induced in Wistar rats by an injection of streptozotocin (STZ). At different times after diabetes-induction, the visual pathway was morphometrically evaluated. A deficit in the anterograde transport from the retina to the superior colliculus at 6 weeks post STZ-injection was observed. At this time point, no RGC loss or substantial alterations in the superior colliculus were found. A large increase in astrocytes reactivity occurred in the distal portion of the optic nerve, which coincided with a significant axon loss. Moreover, profound myelin alterations and altered morphology of oligodendrocyte lineage were observed. The present results suggest that axoglial alterations at the distal portion of the optic nerve could be the first structural change in the diabetic visual pathway.

Sensory Systems
Poster Number 218 | Session 2

"More than just supporting: new insights into the roles of supporting cells in the postnatal and adult inner ear"

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Despite that all inner ear hair cells and their associated sensory nerve terminals are surrounded by supporting cells (SCs), the roles of these non-neuronal cells in the postnatal period remain poorly understood. Recent evidence indicates that SCs have many glia-like characteristics, including expression of erbB receptors. We are using genetically modified mice to investigate the roles of inner ear SCs. Our

studies show that SCs are critical for the formation and maintenance of vestibular and cochlear inner hair cell synapses and for the long-term survival of spiral ganglion neurons. We show that BDNF produced by SCs is critical for synapse formation/maintenance in the vestibular system, while supporting cell-derived NT3 plays similar roles in the cochlea. Expression of BDNF and NT3 is regulated by the NRG1/ErbB signaling pathway, pointing to reciprocal trophic interactions between sensory neurons and SCs in the formation and maintenance of a functional sensory epithelium. Together, these studies indicate that SCs are actively engaged in the promotion of maturation, function and maintenance of the inner ear and that this is, at least in part, mediated by reciprocal signals between sensory neurons and SCs involving NRG1-erbB and BDNF-TrkB/ NT3-TrkC signaling.

Sensory Systems
Poster Number 219 | **Session 3**

"Behavioral response to conspecific alarm cues in tadpoles of Rhinella arenarum: the involvement of the olfactory and vomeronasal organs"

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Early warning about the presence of a predator and triggering of the appropriate avoidance behavior is crucial for prey survivorship. Chemical signals released by disturbed or injured conspecifics may provide decisive information to prey animals. In the present work we found that tadpoles of the common toad, Rhinella arenarum, show an alarm reaction when an extract from injured conspecifics is presented. Activity (measured as time spend moving) decreases in a dose-dependent manner in tadpoles exposed to the extract. In contrast, tadpoles of R. arenarum exposed to extract from injured heterospecific tadpoles (Hypsiboas pulchellus) do not change their behavior. Moreover, in order to assess the involvement of the olfactory and/ or the vomeronasal organ in alarm cues detection, we evaluate activation of the chemosensory neurons of these sensory systems. Taken together, the results show that a species-specific alarm cue exists in R. arenarum. The dose-dependent response observed demonstrates that the stimulus (or stimuli) detected by animals is specific, and codifies imperative information for animalsí survivorship. Finally, measurements of neuronal activity suggest that the olfactory, and not the vomeronasal organ, would participate in this alarm cue detection.

Sensory Systems
Poster Number 220 | Session 1

"Learning modifies odor mixtures representation in the honey bee antennal lobe"

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In the present work we ask, how animals extract information about key components present in complex odor mixtures. The antennal lobe is the first processing center for olfactory information in the insect brain. The local network conformed by excitatory and inhibitory local neurons transforms the olfactory information before it leaves the antennal lobe to other brain areas. The information about rewards converges with odor information in the antennal lobe network. We propose that the association between odors and reward modifies the weight of lateral inhibitions and shifts the representation of a mixture towards the representation of the more relevant components. In the present work we train animals to pure odors and perform calcium imaging in output neurons of the antennal lobes to measure neural activity patterns elicited by pure odors and the respective binary mixtures. We report results, in which we observe that the experience modifies mixture presentation. A comparison of the activity patterns elicited by the mixtures and the patterns expected based on the representation of the pure odors suggests that learning induces modification in the competitive interactions among components.

Sensory Systems
Poster Number 221 | Session 2

"Polarity in sensory neuron axonal transport: relevance to regenerative capacity in the central and peripheral nervous system"

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The differential regenerative capacity of the central and peripheral branches of the spinal sensory neurons is a classic model for evaluating the components molecular, cellular, pharmacological and physiological that contribute to successful or failed nerve regrowth. This is due to an unusual anatomic characteristic of these neurons,

they send one bifurcated axon into both the peripheral and central nervous systems. While the former is capable of regenerating after injury, regeneration of the same axon in the central root fails when it encounters the central nervous system environment. This model provides a versatile system for identifying elements that contribute to, or limit, the regenerative capacity of the nervous system. One contributor to the different central and peripheral capacity for regeneration is the transport of proteins down the two branches after injury to the corresponding branches. Here, we describe the characteristics of two forms of nerve injury that can be used to investigate the inherent and limited capacity of the nervous system to recover after damage. Proteins associated with regeneration are transported bidirectionally after injury to a single branch, and contribute differentially to the regenerative capacity in two very distinct environments.

Sensory Systems
Poster Number 222 | **Session 3**

"Protein nitration: Is it part of the oxidative stress associated with light induced retinal damage?"

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Continuous illumination (CI) induces the degeneration of retinal photoreceptors. Electron Paramagnetic Resonance demonstrated an increase of NO in CI retinas peaking at 24 hs while Western blot (WB) showed a peak of iNOS expression. Our hypothesis is that NO may be involved in the oxidative stress induced by CI, that is why protein nitration was studied. Sprague Dawley rats were continuously illuminated with white light (12000 lux) for 24 hs, 48 hs, 5 and 7 days while control rats (CTL) were kept at light/dark cycles of 12/12 h. The eyes of CTL and CI rats were fixed and processed with the PAP immunocytochemical (ICC) technique using a nitrotyrosine (Ntyr) antibody. Other eyes were processed by WB and quantified by Image J Software. Ntyr immunoreactivity (IR) was observed in Inner Nuclear Layer (INL) and Ganglion Cell Layer (GCL) in CTL rats. After CI, Ntyr IR was observed in Outer Nuclear Layer (ONL) as well as in INL and GCL. WB analysis showed a band pattern that differed among control, 5 and 7 days of CI. The observed results showed a change of nitration patterns in CI rats both by ICC and WB. The increase of NO nitrates tyrosine residues in proteins which alter their

structures and functions leading, with other free radicals, to oxidative damage and retinal degeneration.

Sensory Systems
Poster Number 223 | **Session 1**

"Moderate noise exposure differentially affects hippocampal and auditory structures in developing rats"

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Noise is one of the causative factors of hearing loss. However, the vulnerability of the auditory system to different noise levels seems to be variable. On the other hand, noise can also affect extra-auditory areas such as the hippocampus (Hip). Therefore, the aim of this work was to elucidate if exposure to moderate noise induces histological and functional changes in the auditory pathway. Since hippocampal-related behavioral changes were found in noise-exposed rats, hippocampal histological assessment was made to investigate if potential auditory changes could underlie Hip alterations. Male Wistar rats of 15 days were exposed to white noise (95-97 dB, 2h/day) and separated into acute (AE, 2h/day) and chronic exposure (CE, 2h/day for 15 d) groups. The integrity of the auditory pathway was evaluated by recording auditory brainstem response (ABR). Histological assessment of cochlea and Hip was also performed. Results showed no significant differences in ABR in noise-exposed rats, without changes in cochlear histology. In contrast, histological changes were found in Hip of exposed rats. These data suggest that AE and CE to moderate noise are capable of inducing hippocampal histological impairments in developing rats, without affecting auditory function and morphology.

• SYNAPTIC TRANSMISSION AND EXCITABILITY

Poster Number 224 | **Session 2**

"Prenatal stress effect on glutamate transporters, GLT1 and GLAST"

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Episodes of stress suffered by the mother during pregnancy generate changes in the fetal environment affecting nervous system development of the offspring. Our previous studies have shown that offspring of stressed rats exhibited higher levels of glutamate receptors, astroglial reaction and reduced dendritic arborization with synaptic loss. Since metabolism of glutamate is linked to astroglia, the results suggest that glutamate pathways might be impaired in the prenatally stressed rats. To study the effect of prenatal stress (PS) on the glutamate system, pregnant rats were subjected to restrain stress. Frontal cortex and hippocampus of PS and control (C) rats were evaluated by mass spectroscopy to measure the content of glutamate and other metabolites. In this animal model we also evaluated the glutamate transporters, GLT1 and GLAST. Using gliosomes and synaptosomes we measured 3H-glutamate uptake and the GLT1 AND GLAST proteins and mRNA levels. Our results show that glutamate uptake of adult PS rats is significantly higher and we observed an overexpression of GLT1 protein and its mRNA in the hippocampus. This would indicate that PS produces long-term changes on the expression of glutamate receptors and alters the normal glutamatergic synaptic transmission of the adult brain.

Synaptic Transmission and Excitability

Poster Number 225 | **Session 3**

"The immediately releasable pool of mouse chromaffin cell vesicles is coupled to P/Q calcium channels by the synaptic protein interaction site"

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The immediately releasable pool (IRP) is a group of ready releasable vesicles closely associated with voltage dependent Ca2+ channels. We have previously shown that exocytosis of this pool is specifically coupled to P/Q Ca2+ current. Accordingly, in the present work we found that the Ca2+ current flowing through P/Q-type Ca2+ channels is 8 times more effective to induce exocytosis in response to short stimuli that the current flowing through L-type channels. To investigate the mechanism that may generate the coupling between IRP and P/Q-type channels we transiently expressed in chromaffin cells peptides corresponding to the synaptic protein interaction site (synprint) of Cav2.2, an amino acidic sequence of the Ca2+ channel that serves to maintain a close physical coupling with vesicles in synaptic terminals. This treatment reduced the efficiency of Ca2+ current to induce exocytosis to similar values as ω-agatoxin-IVA. In addition, the same treatment markedly reduced IRP exocytosis, but did not affect the exocytosis provoked by sustained electric or high K+ stimulation. In conclusion, our results indicate that synprint is a crucial factor for the establishment of the functional coupling between IRP vesicles and P/Q-type Ca2+ channels.

Synaptic Transmission and Excitability
Poster Number 226 | Session 1

"Ultra-short agonist application. A novel approach for the kinetic study of ligand gated receptors"

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The experimental determination of the molecular mechanisms of ligand-gated ion channels activation often requires the ability to change solutions quickly. Ultra-short applications, i.e., shorter than the opening time of the receptor (20-100 μs), reveal the current signature of the partially activated states that precede channel opening. The solution exchange system that deliver the shortest pulses on outside out channel preparations is controlled by a piezo actuator that moves the interface between the solutions streams on the patch pipette. The time response of this switcher is limited by mechanical vibrations that appear at increasing velocities of the piezo. In this study, we apply an optimization method based on Fourier

transforms that allowed us to damp the oscillations. In open tip experiments, we obtained pulses of 26.3 ± 1.08 µs (n =150, measured at 50% of exchange) that achieved an exchange index of 92.54 ± 1.31 % (n =150, measured as the height of the solution exchange). The ability of applying ultra short pulses would open an experimental window to the molecular events that occur between the binding of that agonist and the opening of the channel even for the fast gating receptors.

Synaptic Transmission and Excitability
Poster Number 227 | Session 2

"Short-term synaptic plasticity at the medial olivocochlear hair cell synapse in α9L9íT knock-in mice"

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Cochlear amplification is regulated by medial olivocochlear (MOC) neurons that synapse onto outer hair cells (OHCs). This synapse is mediated by the a9a10 nicotinic receptor (nAChR). We have generated a mouse with a point mutation (L9íT) in the a9a10 nAChR that produces longer-lasting inhibitory postsynaptic currents (IPSCs) and changes the magnitude and the dynamics of the efferent-mediated inhibition of cochlear responses (Taranda et al. Plos Biology, 2009). Our goal is now to determine if there is a consequent change in the short-term plasticity (STP) properties of the MOC-hair cell synapse. Synaptic activity was recorded in voltage-clamped inner hair cells (IHCs) from excised apical turns of wild-type (wt) or a9L9íT knock-in (kin) mouse cochleas (P9-11), during electrical stimulation of the MOC fibers. Evoked IPSCs in kin mice had longer decay times and smaller amplitudes. In wt IHCs, prolonged high frequency stimulation produced an increase in the postsynaptic response during the 1st second, followed by depression that produced ~80% decay in the response. In kin mice, peak responses were reached after ~3 sec of stimulation and then decayed by 50%. These results show that changes in the dynamics of the nAChR induce dramatic changes in the MOC-hair cell synapse STP properties.

Synaptic Transmission and Excitability
Poster Number 228 | **Session 3**

"Design strategies for improving dynamic clamp performance"

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Dynamic clamp is an electrophysiological technique capable of interfacing an electronic device and a living neuron in real time. In particular, this method can simulate the currents that pass through ionic channels. The technique relies on the creation of a control loop between the injected current and the recorded membrane potential. Since the delivered current is determined by the intracellular voltage, it is necessary to develop a solving algorithm that calculates that current on the basis of several parameters, e.g. the membrane potential and the instantaneous values of activation and inactivation variables. In this work, we analyze several strategies to optimize this procedure, including optimal data rounding based on experimental uncertainties, among others. Therefore, it was possible to speed up the design and to use fewer logic resources in the implementation of a dynamic clamp device, thus achieving a higher instrument performance. Additionally, we design and build a dynamic clamp system capable of reproducing the electrical effects of the fast inactivating K+ channel, which is present in neocortical pyramidal neurons. The developed device, implemented on a Field Programmable Gate Array, effectively sets up the control loop, with an operation frequency of 500 kHZ.

Synaptic Transmission and Excitability
Poster Number 229 | **Session 1**

"A semiconductor-based photoconductive stimulation device"

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The ability to generate complex patterns of cell excitation is of key importance in the research of the dynamic behaviour of excitable tissues. In that sense, it is desirable to have a device capable of stimulating several individual neurons simultaneously with a specific temporal pattern of stimulation. To achieve this objective, we take advantage of the fact that the incidence of light is an effective way of modulating the electrical conductivity of a Silicon monocristaline surface. It

is possible to create a "virtual" extracellular stimulation electrode by aiming a laser at any specific location of a Silicon substrate, thereby producing a local current in the generated photoconductive pathway. By targeting the laser to the region of interest, local photocurrent can stimulate neurons in the light path. Within some limits, it is possible to obtain specific temporal patterns of stimulation by varying the bias voltage applied to the substrate, and by taking into account the impedance characteristics of the substrate. In this work, we present preliminary results of the device functioning. The technique presented here could be particularly efficient for the extracellular stimulation of dissociated neurons cultured on silicon wafers, with minimal physiological manipulation.

Synaptic Transmission and Excitability
Poster Number 230 | **Session 2**

"Local network activity controls neuronal maturation in the adult dentate gyrus"

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The adult hippocampus continuously generates new cohorts of immature neurons with increased excitability and plasticity. In this work we show that local network activity regulates the rate of maturation of adult-born neurons along the septo-temporal axis of the hippocampus. Confocal microscopy and patch clamp recordings were combined to assess marker expression, morphological development and functional properties in retrovirally labeled neurons over time. The septal dentate gyrus displayed higher levels of basal network activity and faster rates of newborn neuron maturation than the temporal region. Voluntary exercise enhanced network activity only in the temporal region and, in turn, accelerated neuronal development. The role of electrical activity was further supported by the observation that neurons exhibited a delayed maturation when their intrinsic electrical activity was reduced by the cell-autonomous overexpression of Kir2.1. Finally, intrinsic neuronal activity promoted long-term survival of new granule cells. Our findings reveal a novel type of activity-dependent plasticity acting on neuronal maturation, functional integration and survival of adult-born hippocampal neurons.

Synaptic Transmission and Excitability
Poster Number 231 | Session 3

"Reduction of presynaptic calcium influx with higher amplitude excitatory post-synaptic currents in S218L Cav 2.1 knock-in migraine mouse model"

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We used KI S218L mice to study Ca2+ currents and excitatory postsynaptic currents (EPSCs) at the calyx of Held. During whole-cell patch-clamp recordings, a shift of the peak of the I-V curve to more negative potentials was observed. Steady-state activation curves were also significantly shifted between WT and KI mice, activating at more negative potentials in KI. On the other hand, presynaptic calcium currents (IpCa) evoked by action potential (AP) waveforms had less amplitude in KI than WT. Additionally, Ca2+ current facilitation after 100 Hz train of APs was significant reduced in KI compared to WT mice. Both EPSC amplitudes and miniature EPSC frequencies were significant higher in KI than WT. The synaptic activity in the KI showed less rate of short term depression with a faster recovery after either 10 or 100 Hz frequency trains. Our results suggest that the calcium channel activation shift might be increasing calcium influx at resting membrane potential. Such calcium concentration increment would lead to a decrement in action potential-induced calcium influx due to channel inactivation. Moreover, an acceleration of calcium dependent process on the exocitosis machinery might explain the observe increment in EPSC amplitude regardless of the observed reduction in IpCa.

Synaptic Transmission and Excitability
Poster Number 232 | Session 1

"Ghrelin increases glutamate release from rats hippocampal synaptosomes"

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Ghrelin (Ghr) is a peptide that participates in the modulation of different biological processes. In our laboratory we have shown that Ghr modulated memory acquisition and consolidation; intra-hippocampal Ghr administration increased the nitric oxide synthase (NOS) activity in a dose dependent manner, and reduced the threshold for LTP generation in dentate gyrus. The biochemical memory cascade is initiated by the glutamate (Glu) release followed by the activation of AMPA and NMDA receptors, increasing [Ca2+]i, NOS activity and GMPc levels. The possible mechanisms underlying the Ghr-facilitatory effect on memory have not been clarified. NO enhances the Glu release and this evoked release is important to LTP. In the present work we studied, the effect of Ghr on Glu release using an in vitro preparation of rat hippocampal synaptosomes incubated in the presence of Ghr. Glu was monitored fluorometrically. Data points were obtained at 1-s intervals during 20 min. Results are expressed as % of change in Glu release in relation to the control group. The addition of Ghr to the medium increases Glu release reaching 30% with the dose of 0.03 nM. In conclusion, results provide additional evidence about the neurobiological bases of Ghr action in hippocampus.

Synaptic Transmission and Excitability
Poster Number 233 | Session 2

"Effect of repetitive cocaine "binge" administration on GAD65/67 and T-type subunits levels from GABAergic somatosensory thalamic neurons in mice"

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A cocaine "binge" induces an enhancement in both GABAergic transmission and T-type calcium currents on Ventrobasal (VB) thalamic neurons, which were prevented by the systemic administration of T-type calcium channel antagonists mibefradil and 2-octanol. However, it is unknown if protein levels of GAD65/67 or T-type calcium channel subunits are affected by cocaine. The objective of this study

was to assess, using Western Blotting, the effects of acute and chronic cocaine "binge" on GAD65/67, Cav3.1 and Cav3.3 T-type subunits protein levels compared to saline administration from mice thalamic VB and Reticular (RetN) nuclei. One day after a chronic "binge" protocol (one "binge" a day, for 3 days) an increase in protein levels of Cav 3.1 was observed in the VB nucleus (ANOVA p=0.01), while no changes in GAD65/67 levels were found. After a fourth "binge" administration, no changes were observed in VB Cav 3.1 protein levels. GAD67 levels were increased in RetN nucleus (ANOVA p=0.024). No differences were found in GAD65/67 protein levels in the VB nucleus. Changes in the expression levels of these two key proteins may help understand alterations in thalamocortical networks of long-term cocaine abusers. Grants: PICT 2007-1009/2008-2019, PIDRI-PRH 2007, PIP 11420100100072.

Synaptic Transmission and Excitability
Poster Number 234 | **Session 3**

"Muscarinic and adrenergic neurotransmission at CA1 area of rat hippocampus"

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Adrenergic and muscarinic neurotransmission in rat hippocampus are involved in synaptic plasticity. We have shown that muscarinic toxin 1 (MT1) from Mamba snake venom (an M1 muscarinic receptor agonist and M4 antagonist) also binds to hippocampal α-adrenergic (α-a) receptors. In radioligand binding assays in rat hippocampal membranes, MT1 inhibited the binding of the muscarinic ligand 3H-N-methylscopolamine (3H-NMS, Ki=180±7 nM) and that of the α-a antagonist 3H-prazosin (3H-PRZ, Ki=83±3 nM), with maximal inhibition of 49±5% and 34±4%, respectively. Furthermore, PRZ inhibited 3H-NMS binding (Ki=5.1±1.2 μM). Further assays are carried out to estimate inhibition parameters for the α-a agonist phenylephrine (PE) on 3H-PRZ and 3H-NMS binding. We registered field excitatory postsynaptic potentials (fEPSPs) in CA1 area of rat hippocampal fresh slices. Perfusion with MT1, PRZ and PE modified fEPSPs. MT1 (1μM) and PRZ (10μM) significantly increased fEPSP (41.4±5.2%, n=5; 15.8±3.2%, n=6, respectively); while PE (10μM) decreased fEPSP (23.0±9.0%; n=4). These indicate that activation of α-a receptors inhibits basal transmission at CA1 synapse. Co-perfusion of MT1+PRZ and MT1+PE

would help to discriminate MT1 action on α-a receptors.
Synaptic Transmission and Excitability
Poster Number 235 | **Session 1**

"Facilitation and depression determine timing of synaptic responses at the inner hair cell ribbon synapse"

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The auditory system must represent different features of the acoustic environment, including the temporal structure of incoming sounds. It is well established that auditory nerve fibers are able to fire at a particular phase of low-frequency periodic stimuli. This capacity is driven by the transmitter released from inner hair cells (IHC) in the cochlea and is known to occur regardless of the intensity of the stimulus. In the current study, we decided to investigate the mechanisms by which IHC are able to release neurotransmitter with constant timing even with stimuli of different intensities. We used simultaneous recordings from IHC and postsynaptic boutons of auditory nerve neurons. We firstly observed that with trains of square pulse at 250 Hz IHC released neurotransmitter with constant delay. Synaptic responsiveness was evaluated at the end of these trains of different intensities, observing that stronger stimuli produced deeper synaptic depression and longer delays. Therefore, during repetitive stimulations a given pulse promoted release by allowing Ca2+ in the cell, but simultaneously, provoked depression due to partial depletion of vesicles. We propose that an equilibrium would be established between pulse intensity and depression, determining a constant synaptic timing.

Synaptic Transmission and Excitability
Poster Number 236 | **Session 2**

"GABA receptors in Caenorhabditis elegans embryonic muscle cells"

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GABA receptors mediate fast inhibitory neurotransmission in both vertebrates and invertebrates. They are targets of anxiolytic and antiepileptic drugs as well as of commonly used insecticides and antihelminthics. Here we investigated GABA receptors in the free-living nematode Caenorhabditis elegans, which is a model

for the study of the nervous system as well as a model of parasitic nematodes. Nematode muscle contains, in addition to two types of nicotinic receptors, a GABA receptor. We used a primary culture to explore the functional properties of GABA receptors from L1 muscle cells at the macroscopic and single-channel levels. Rapid application of GABA to L1 muscle cells elicits inward currents that decay fast due to desensitization. In cell-attached patches, 1-10mM GABA activates channels of ~ 2.6 pA (membrane potential: +100mV) and a mean duration of about 0.22 ms. The frequency of channel openings increases as a function of GABA concentration. The analysis of single-channel properties shows a homogenous channel population, indicating the presence of a single GABA receptor subtype. Our study contributes to the understanding of muscle GABA receptor properties and, in turn, provides new avenues for exploration of anthelmintic therapies.

Synaptic Transmission and Excitability
Poster Number 237 | **Session 3**

"Angiotensin II AT2 receptor blocker reverses the neuroprotective action of preconditioning in the brain of hypoxic-ischemic neonatal rats"

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AT2 receptors (AT2-R) are involved in the control of cell proliferation and tissue regeneration. In a previous report we observed that AT2-R were up-regulated in the preconditioned neonatal brain after ischemia, and their activation could be neuroprotective contributing to the beneficial effects of AT1 receptor antagonists on neurological outcome of cerebral ischemia. Seven- day old WKY rat pups were submitted to preconditioning (PC) and then to common carotid unilateral artery ligation (I) / 2 min asphyxia on day 8 (H). The lesioned group (L) was submitted only to I/H. A third group was PC+I/H and injected 5 µl of PD123319 (1.69mM) in the lateral ventricle after PC. Control animals received mock treatment. The animals were sacrificed at 7 days post lesion (PND15). Ki67 (a marker of cell proliferation) and GFAP (reactive astrocytes) were analyzed by IHQ (DAB). The treatment with the AT2 blocker reversed the effects of preconditioning, increasing the GFAP+ label ($p<0.001$ vs PC) in both hemispheres and dampening the increase of Ki67+ unidentified cells in the area of lesion ($p <0.01$ vs. PC).We hypothesize that AT2 receptors may participate in the neuroprotective action of preconditioning by direct or indirect regulatory actions on the AT1 receptors. Subsidio ANPCyT, PICT

2006-451.
Synaptic Transmission and Excitability
Poster Number 238 | Session 1

"M1 muscarinic receptor positively modulates neurotransmission in CA1 area of rat hippocampus"

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Although muscarinic cholinergic modulation of neurotransmission in rat hippocampus has been widely investigated, the involvement of the different receptor subtypes is not clear yet. Muscarinic toxin 2 (MT2) is a selective M1 receptor agonist and M4 antagonist, with higher affinity for M1. MT2 enhanced field potentials (fEPSPs) at CA1 synapses in rat hippocampal slices. This effect was blocked by pirenzepine, a selective antagonist for M1 and M4 receptors. fEPSPs and spike population recordings were performed in CA1 area of rat hippocampal acute slices to study the effect of MT2 on input-output curves (I/O). MT7, a selective M1 antagonist, was co-perfused with MT2. MT2 shifted I/O (log IC50: control 0,65±0,05; MT2 0,51±0,04). MT7 abolished MT2ís effect on fEPSP and I/O. Considering that MT2 has significant affinity for α-adrenergic receptors, binding assays were performed in hippocampal membranes to evaluate MT7 inhibition of 3H-N-methylscopolamine (3H-NMS, muscarinic ligand) and 3H-prazosin (3H-PRZ, α-adrenergic ligand) binding. MT7 inhibited 3H-NMS but not 3H-PRZ binding. Maximal inhibition of 3H-NMS was close to M1 ratio in rat hippocampus. These results show that M1 receptors positively modulate neurotransmission in CA1 synapses.

Synaptic Transmission and Excitability
Poster Number 239 | **Session 2**

"The effect of calcium dyes on the observed dynamics of calcium signals"

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Calcium signaling is ubiquitous across cell types. Intracellular calcium signals are observed in intact cells with a minimum disruption using calcium dyes. The most commonly used indicators fluoresce upon calcium binding, thus providing information on the calcium-bound dye concentration. Their presence affects the calcium dynamics since dyes act as calcium buffers. Cells have buffers, usually proteins, that bind calcium altering its spatio-temporal dynamics. In this work we address the issue of to what extent the observed signals vary depending on the dye that is used. To this end we perform experiments in Xenopus Laevis oocytes using confocal microscopy and two dyes, Fluo 4 and Rhod 2, to observe signals that arise due to calcium release from the endoplasmic reticulum through IP3 receptors (IP3Rís). The dyes probed differ in their binding kinetics and dissociation constant. However, we have found that it is possible to choose their concentrations in such a way that, when using separately, the properties of the observed signals remain undistinguishable. We also observe signals in the presence of both dyes using a multispectral microscope. In this way we are able to study the competition between the two dyes probing the effect of their different kinetics on the observed signals.

Synaptic Transmission and Excitability
Poster Number 240 | **Session 3**

"5-Bromo-2`deoxyuridine is toxic to olfactory epithelium cells at concentrations widely used for studies of cells lineage"

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The 5-bromo-2\\'-deoxyuridine (BrdU), is a thymidine analogue that incorporates in to the DNA of dividing cells during the S-phase of the cell cycle. This analogue is widely used for the study of mitosis and cell lineage in vitro and in vivo. However, there are reports that questioned the validity of its use in some biological systems where BrdU showed cellular toxicity. The current work examines the effect of BrdU incorporation on the phenotype of cells in the olfactory epithelium (OE). We found a decrease in mature olfactory receptor neurons (ORNs) as a function of BrdU exposition time when we used 10 mM BrdU. We also observed an increase in the number of apoptotic cells after the BrdU administration. Moreover, while the epithelial structure in BrdU treated animals was maintained, the histochemical analysis showed alterations in secretory vesicles from the sustentacular cells. BrdU 0,1-1 mM did not show the same toxic effects. We conclude that the use of BrdU 10 mM (doses of current use in the literature) has toxic effects on ORNs, activating classical cell death pathways and altering the histomorphology of the sustentacular cells. These results demonstrate the importance of using the correct dose of BrdU in neurogenesis studies both in vivo and in vitro.

Synaptic Transmission and Excitability
Poster Number 241 | **Session 1**

"Decisions on magnitude and parity in two consecutive tasks"

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The stimuli are digits. There are two consecutive decision making tasks. During the first task, the participant decides whether the stimulus "prime" (1, 2, 3, 4, 6, 7, 8 and 9) is greater or less than 5. During the second task, the participant decides whether the target (1, 4, 6 and 9) is even or odd. In Task 1, the participant is instructed not to answer in 50% of the trials (depending on the format of the stimulus). We measure the RT to solve task 2, as a function of the time between the two stimuli and analyse how the task on the prime conditions the task on the target. The results shows an effect of Congruence between Tasks: answers to the target are facilitated when the hand to respond about its parity is the same hand that would have been used if asked about its magnitude (as in task 1). It is notisable that the answer (to the target) is also facilitated when the hand to respond about magnitud in task 1 is the same hand that would have been used if asked about its parity (as in task 2). Both effects are stronger when the prime belongs to the set of digits chosen as targets and is not present when the prime was ignored.

Synaptic Transmission and Excitability
Poster Number 242 | **Session 2**

"Functional changes of Cys-loop receptors generated by electromagnetic fields"

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Due to the rapid advances in communication technologies, the population is increasingly exposed to electromagnetic fields (EMFs). This has raised concern about potential health effects resulting from exposure to them. We here studied the influence of EMFs on AChR and 5HT3A receptors. We recorded macroscopic and single-channel currents from cells expressing these receptors and exposed to EMFs before or during agonist application. Exposure to EMFs (15 Hz-120 kHz) produces a significant decrease of the peak current, an increase on the rise time and no changes in the decay time constants of macroscopic currents activated by ACh. The peak current decreases as a function of EMF frequency (IC50 54 kHz). The effect on 5HT3A receptor currents is 3-fold more profound than on AChR currents, thus indicating different sensitivity of Cys-loop receptors to EMFs. The single-channel amplitude of AChR channels is not affected by the EMF, revealing that the reduction of macroscopic currents is originated from a change in the activation kinetics and not in the ion permeability of the receptor. In accordance with this, open time histograms change in EMF-exposed AChRs. Our results reveal that EMFs affect ligand-gated ion channel function and open doors to understand the mechanistic of such effect.

Synaptic Transmission and Excitability
Poster Number 243 | **Session 3**

"Functional maturation of striatal cholinergic interneurons during adolescence"

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Acetylcholine released by a small population of interneurons, referred to as "tonically active neurons" (TANs), is a main modulator of striatal function. The tonic activity of cholinergic interneurons depends on intrinsic mechanisms. Action potentials open Cav channels and activate Ca2+ dependent K+ currents. This leads to an afterhyperpolarization (AHP) with three phases: fAHP, mAHP, and sAHP, which produces the activation of Ih. Previous results show that intrinsic properties of TANs mature during adolescence. We propose that KCNQ channels with atypical properties, activated by Cav through the hipocalcin, mediate a protracted maturation of the AHPs. Increases in one or more components could explain the more marked firing adaptation in adult compared to juvenile TANs. Firing frequency adaptation defines neurons computational properties. Neurons with more marked IsAHP and Ih may act as resonators and codify the signals in the temporal precision of each spike. We propose that differences in the frequency adaptation between juvenile and adult TANs determine how they codify environmental cues during instrumental learning.

Synaptic Transmission and Excitability
Poster Number 244 | **Session 1**

"A methodological approach to monitor fluctuations on vesicular membrane potential"

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Introduction.Synaptic vesicles are intracellular structures that contain neurotransmitters wich are liberated to the synaptic cavity allowing the communication between two cells.In the same form that plasmatic membrane separates the intra and extracellular media, vesicles are defined by a lipid membrane.Therefore, can be also considered an electrical capacitor with specific proteins mediating the ion exchange.The contribution of different conductances to the vesicular resting potential has been suggested but not described.Methods. We design a methodological approach to monitor changes in ionic permeability across the vesicular membrane wich combines a GFP fused to a vesicular membrane protein, and the hydrophobic ion dipicrylamine (DPA). DPA molecules moves inside the dielectric in response to changes in the membrane potential producing the quenching of GFP by a FRET phenomena.We setup the preparation of membrane sheets using a sonifier, this allow us to have full access to the intact docked vesicle. Imaging was performed using a laser TIRF microscope coupled to a Hamamatsu

Orca 12ER CCD camera. Results. After loading DPA we perfused solutions with different ionic composition recording fluorescent fluctuations at the level of single vesicle wich manteins its integrity.

Synaptic Transmission and Excitability
Poster Number 245 | **Session 2**

"GABA regulates the release of acetylcholine (ACh) at the medial olivocochlear (MOC) efferent-inner hair cell synapse through presynaptic GABAB(1a,2) receptors"

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Before the onset of hearing, inner hair cells (IHCs) of the mammalian cochlea are innervated by MOC efferent fibers. Although ACh is the main neurotransmitter, GABA is present at MOC synaptic terminals. We have previously shown that GABA modulates the cholinergic input at MOC-IHC synapses by acting on GABAB receptors. To further evaluate the role of GABAB in the modulation of ACh release, synaptic currents evoked by electrically stimulating the efferent fibers were recorded in voltage-clamped IHCs from isolated mouse organs of Corti of GABAB knock-out (KO) mice isoforms. GABAB receptors are formed by the GABAB2 subunit with either the GABAB(1a) or the GABAB(1b) subunit. We compared the effects of the agonist baclofen in GABAB(1a-1b), GABAB(1a) and GABAB(1b) KO mice. Baclofen (1μM), caused a significant reduction in the quantum content (m) of evoked release in both, wild-type (32.3±6.9%, p 0.05) and GABAB(1b) KO mice (26.7±7.7%, p 0.05). Baclofen did not affect m in GABAB(1a-1b) or GABAB(1a) KO mice (p>0.05, n=5). Our results show that ACh release at the MOC-IHC synapse is negatively regulated by GABA acting on presynaptic GABAB(1a,2) receptors.

Synaptic Transmission and Excitability
Poster Number 246 | **Session 3**

"Voltage-dependent conductances modulate the amplification and propagation of signals in nospiking neurons"

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A pair of nonspiking neurons in the leech nervous system, the NS cells,is present in each midbody ganglion and displays a very extensive arborization.Given the wide influence of these neurons on effector neurons(e.g. they are capable of modulating motor behaviors),it has been of interest to analyze how signals are integrated by the NS neurons. In spite of the fact that they do not display Na+-dependent spikes they exhibit a widespread distribution of voltage-dependent-Ca++-conductances(VCC). To study how VCCs are involved in the integration and propagation of inputs in the neuritic tree of the NS neurons we have analyzed the calcium signals evoked by brief depolarizing pulses of different amplitudes.To modulate the expression of the voltage-dependent conductances, the study was performed at different membrane potentials(Vm). The experiments were performed in isolated leech midbody ganglia, in which NS neurons were loaded with the Ca++ probe Oregon Green 488 BAPTA-1 and recorded in a confocal microscope. The results obtained at negative potential (-60mV, where increasing depolarizing signals produce no-linearly increasing Ca++ transients) suggest that VCC in NS neuron aid the propagation of signals throughout its extensive neuritic arbor.

Synaptic Transmission and Excitability
Poster Number 247 | **Session 1**

"Functional development of the medial olivocochlear efferent innervation before the onset of hearing"

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During development, inner hair cells (IHCs) are innervated by medial olivocochlear fibres. At P9-11 transmitter release is supported by P/Q and N-type voltage-gated Ca2+ channels (VGCC) and negatively regulated by BK channels. The quantal content (m) of release increases between P5-7 and P9-11. This is accompanied by dramatic changes in the short term plasticity (STP) properties. To determine the basis for these developmental changes in synaptic transmission, postsynaptic responses were monitored in voltage-clamped IHCs while electrically stimulating

the efferent fibres in isolated mouse organs of Corti. At P5-7, w-AgatoxinIVA reduced m to 37±6% whereas 1 microM w-Conotoxin GVIA failed to block release, revealing that P/Q- but not N-type VGCC partially support release at this stage. To test whether BK channels modulate transmitter release, P5-7 cochleas were incubated with Iberiotoxin, a BK channel antagonist. As reported for P9-11, m increased to 192±11% of control. The readily releasable pool (RRP) size increased from 4.7±1.0 vesicles at P5-7 to 10.7±2.3 vesicles at P9-11. Our results suggest that both differences in subtypes of VGCC that support transmitter release as well as differences in the RRP size, underlie the observed developmental changes in synaptic transmission.

• COGNITION, BEHAVIOR AND MEMORY

Poster Number 248 | **Session 2**

"Memory impairment induced by scopolamine: storage or expression deficit?"

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CF-1 male mice were trained in an inhibitory avoidance task. Pre-training administration of scopolamine (SCP, 0.50 mg/kg, ip), a non selective muscarinic acetylcholine receptor antagonist, impaired retention performance at different training-test intervals. Post-retrieval intra dorsal hippocampal infusions of choline (Ch, 0.80 µg/hippocampus), a α7 nicotinic acetylcholine receptor agonist, either enhanced or impaired retention performance depending on the training conditions (mild or high footshock respectively). Ch (0.80 µg/hippocampus) administered immediately after retrieval reversed the impairment induced by pre-training SCP (0.50 mg/kg, ip) administration. Ch effects were time-dependent and also depend on the interval that mediates between training and the first retrieval session (2, 7, 15 and 21). Our results suggest that SCP memory impairment could be attributed to a deficit on memory expression rather than memory formation and also that, modulation of memory reconsolidation by α7 nicotinic hippocampal receptor depends on the time elapsing from training.