

**Abstracts from the
First Meeting of Argentine Biology Societies**



**I Reunión Conjunta de
Sociedades de Biología de
la República Argentina**

**AGOSTO 2007
HUERTA GRANDE // CORDOBA**

**XVI Annual Scientific Meeting of Córdoba Biology Society
(Sociedad de Biología de Córdoba)**

**IX Meeting of Argentine Biology Society
(Sociedad Argentina de Biología)**

**XXV Annual Meeting of Cuyo Biology Society
(Sociedad de Biología de Cuyo)**

With the participation of

**Rosario Biology Society
(Sociedad de Biología de Rosario)**

**Tucuman Biology Association
(Asociación de Biología de Tucumán)**



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IX Annual Meeting
Argentine Biology Society



XVI Annual Meeting
Córdoba Biology Society



XXV Annual Meeting
Cuyo Biology Society



Rosario Biology Society



Tucumán Biology Association.

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L.1.**HARVESTMEN (*ARACHNIDA*) AND BIOGEOGRAPHIC PATTERNS***Acosta LE**CONICET – Cátedra de Diversidad Animal I, Facultad de Ciencias Exactas, Físicas y Naturales, Universidad Nacional de Córdoba.*

Harvestmen (Arachnida: Opiliones) are generally recognized as good model-organisms in biogeographic studies. Their diversity patterns suggest a close dependence to humidity conditions, while their limited vagility would explain the strong degree of endemism observed in some regions. In this lecture I present several distributional patterns of Argentinean and Neotropical opilionids, referring to both endemic and widespread species, either supporting or contrasting the above mentioned features. Ecological niche modeling (bioclimatic profile, method Bioclim, software Diva-GIS; climatic layers of 2.5 min resolution: Worldclim 1.4) was employed to analyze the potential ranges of mesopotamian, chacoan and yungas harvestmen, with special attention to species with a disjoint Mesopotamia-yungas range. The most limiting factors in each sector are discussed (in most of the range, temperature variables seem more relevant than humidity variables). General patterns of diversity in the area were analyzed using the current harvestmen data base (100 spp., >1000 georeferenced records). Aside, patterns typical to mountain-dwelling species are shown, evidencing a remarkable degree of endemism, even at genus level and in sectors of presumable environmental homogeneity. Andean harvestmen reflect the geographical complexity of the area, which embraces a variety of environments depending on elevation, orientation and isolation of ranges; examples are provided from the Argentinean yungas, as well as Andean taxa from Bolivia and Peru, most of them Gonyleptidae Pachylinae. Finally, some cases of relictual species found in the xeric western Argentina, otherwise an area completely lacking harvestmen, are given.

*Financial support: CONICET and SECyT-UNC.***L.2.****NEUROGENESIS IN THE ADULT HIPPOCAMPUS: REWIRING THE BRAIN***Schinder AF**Instituto Leloir, Buenos Aires.*

The hippocampus is a cortical structure involved in learning and memory, and contains self-renewing neural stem cells (NSCs) that can generate neurons throughout life. The transformation of a NSC into a mature neuron is a complex process involving discrete phases such as proliferation, phenotype determination, migration, maturation, functional integration, survival. Combining *in vivo* retroviral labeling with electrophysiology and confocal microscopy we have recently revealed a precise developmental sequence for the maturation of adult-born neurons that recapitulates the one occurring during development (*Espósito, J Neurosci 2006*): neuronal determination is followed by synaptogenesis of dendritic GABAergic terminals, continuing with formation of glutamatergic contacts, and ending with perisomatic GABAergic synapses. Compared mature granule cells born in the developing and adult hippocampus we have found that they all converge into a homogeneous population with indistinguishable afferent connectivity with regard to both glutamate and GABA (*Laplagne, PLoS Biol 2006; Laplagne, Eur J Neurosci 2007*). Adult-born neurons can fire action potentials in response to an excitatory drive, exhibiting a firing behavior comparable to that of neurons generated during development. These observations suggest that neurons born in such distinct environments may be under control of homeostatic mechanisms underlying this convergence of function. Revealing these basic mechanisms of synapse formation and neuronal function seems critical to understanding network plasticity in the adult brain.

L.3.**INTRACELLULAR VESICULAR TRANSPORT: ROLE OF RAS INTERFERENCE I (RIN1) AND RABEX5 ON ENDOSOME FUSION DRIVEN BY EPIDERMAL GROWTH FACTOR RECEPTOR***Galvis A, Balmaceda V, Barbieri MA.**Department of Biological Sciences, Florida International University, Miami, FL, USA. E-mail: barbieri@fiu.edu*

Fluid phase and receptor-mediated endocytosis involve a series of intracellular membrane fission and fusion reactions that play an essential role in the regulation of the efficiency of both processes. An endosome-endosome *in vitro* fusion assay is described using two different ligands. The first one-biotin conjugated epidermal growth factor-is rapidly and efficiently internalized to the endosomal compartment via the epidermal growth factor receptor (i.e., receptor-mediated endocytosis), and the second one –avidin-beta-galactosidase- is internalized via pinocytosis (i.e., non receptor-mediated endocytosis). Both ligands were localized in endosomes as determined by fractionation on Sucrose gradients. Incubation of vesicles prepared from the two set of cells resulted in vesicle fusion as indicated by the formation of EGF-biotin-avidin-beta-galactosidase complexes. Under our experimental conditions, fusion was time-, ATP-, KCl, and temperature-dependent. Furthermore, Rab5 was required in both set of vesicles as well as from the cytosolic fraction. Interestingly, Rin1 and Rabex-5, two Rab5-exchahng factors, also were required for optimal endosome fusion driven by epidermal growth factor receptor. However, these Rab5-exchahng factors were required in one set of vesicles. Finally, we found that the endosome-associated Rab5-exchange factors are required for the heterotypic fusion between early endosomes.

L.4.**ETHIC, CLINICAL TRIALS AND THE PHARMACEUTICAL INDUSTRY. A DEVELOPMENT, THREE ACTORS AND THEIR TROUBLES***Bottasso O.**Instituto de Inmunología, Facultad de Ciencias Médicas, UNR.*

A new drug development is an undertaking that requires a joint activity from participants coming from the academic and industrial fields within a strict adherence to ethical principles guiding biomedical research. The first controlled clinical trial was performed in United Kingdom in 1946 to assess the efficacy of streptomycin in patients with tuberculosis. Such study provided the basis for carrying out a countless number of clinical trials that enabled the development of rationally successful therapies for a broad spectrum of medical problems. Since the 80 decade, a series of gradual and disturbing deregulations have led, however, to a frank preponderance of pharmaceutical industry over the academic institutions. To provide a few but significant examples: nowadays the implementation of clinical trials is mostly under the control of industry because of the increased laxity of control agencies together with a significant number of supposedly innovations which in essence are new versions of known compounds. Within this new scenario ethical principles are also affected.

S.1.**BIOLOGICAL APPLICATIONS OF NANOMATERIALS***Barbero CA.**Departamento de Química, Universidad Nacional de Río Cuarto
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The relationship between biology and nanomaterials works both ways. On one hand, nanomaterials could be applied in biological research and on the other biological phenomena could inspire or even build nanomaterials.

In the present communication we show the synthesis of carbon nanotubes functionalized with semiconductor (CdSe) quantum dots and their use to dye cells. To avoid cadmium toxicity, novel small nanoparticles or metallic clusters are synthesized by reduction/stabilization with conducting polymers or chemical reduction inside inverse microemulsions. The cluster shape and size are measured using scanning tunneling microscopy.

The synthesis, dispersion and use of conducting polymer nanofibers in photothermal cancer therapy are tested. Successful internalization is shown using fluorescence spectroscopy. The polyaniline nanofibers tagged with dansyl and dispersed using poly(vinylpyrrolidone) are not citotoxic.

Additionally biocompatible polymer (e.g. polystyrene) surfaces are nanostructured using Direct Laser Interference photolithography into lines or hemispherical cavities. The surfaces can be applied as templates for cell growth and differentiation. Finally, interpenetrated networks of conducting polymers in thermosensitive hydrogels are switched using microwaves opening a new method of drug delivery.

Self assembly in biological systems inspires the electrostatic self-assembly of polyelectrolytes, functionalized carbon nanotubes and carbon microparticles. The hierarchical properties of some assemblies produce novel properties unattainable with only one scale.

On the other hand, polymer nanostructures could be used as templates in biominerilization. Incorporation of functional groups ($-SO_3^-$, $-COO^-$) into polyaniline backbones changes the effect of the polymer on the morphology and crystallography of $CaCO_3$ deposits grown chemically.

The work was financed by SECYT-UNRC, FONCYT, CONICET and SECYT-ECOS Sud. C.A. Barbero is a permanent research fellow of CONICET.

S.2.**NANO-NEUROSCIENCES AND SEL-ORGANIZED BIOSTRUCTURES IN NEUROBIOLOGICAL PROCESSES***Maggio B.**Dpto. Química Biológica-CIQUVIC. Fac. Cs. Químicas. UNC. 5000 Córdoba. E-mail: bmaggio@dqb.fcq.unc.edu.ar*

The hallmark of biomolecular structure is the acquisition of spontaneous molecular, supra-molecular and topological self-organization over a size range of nanometers to micrometers. Synergic factors determine their thermodynamic stability and the topographic distribution of components. This results in lateral and transverse tensions leading to segregation of structural domains that can determine biorecognition and reactivity. There can be no Nanotechnology without advances in Nanoscience. Nano-neurosciences attempt to employ the self-organization of biomolecules to modulate the differentiation, protection and regeneration of nerve cells through controlled adhesion/growth/differentiation for stimulation/response/regulation of nerve cell activity, neural molecule tracking and deposition of neural molecules, membranes and cells over supports conditioned at the nanoscale level. The “top-down” technologies, reasonably adequate on the micro-scale, are increasingly experiencing lack of precision for the control of molecular nano-biostructures at the nanoscopic level without an exceedingly high instrumental cost required to control their organization and interactions. By contrast, the spontaneous “bottom-up” self-organization of biomolecules contains intrinsic capacity to generate specific structures with self-regulated properties and reactivity. This represents an enormous advantage: it is not necessary to pre-design nanoframes or nanotemplates for their assembly but only understand the determining molecular codes and factors to direct them to specific defined responses (Maggio *et al.*, 1981; Maggio, 1994; Maggio *et al.*, 2004-2006).

S.3.**SELF-ASSEMBLED NANOPARTICLES AND MONOMOLECULAR LAYERS AS TOOLS IN THE MOLECULAR PHARMACOLOGY**

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In the classical pharmacological paradigm, interesting drugs are those capable to interact in a reversible manner with a binding site at a receptor protein. In the case of membrane bound proteins, which are mechanically connected with its molecular environment through the lateral pressure (π), the coupling between protein structure and function may be exerted through lipids. In agreement with this statement, the binding of [^3H]flunitrazepam (FNZ) to the gamma-amino butyric acid receptor (R-GABA_A) in conditions of constant planar curvature (Langmuir-Blodgett films), exhibited a different type of kinetics that changed from hyperbolic to sigmoid upon increasing the π from 15 to 35 mN/m. This could be due to a cooperative effect similar to that expressed by allosteric enzymes, which may reflect the mechanosensitivity of the R-GABA_A. Moreover, the qualitatively changing kinetics could also indicate that the binding kinetic order is coupled to the fractal topological dimensionality of the molecular organization of the receptor surroundings, revealed by atomic force microscopy data. On the other hand, the partition of FNZ in bilayers, affected the size of lipidic nanoparticles as well as the morphology of cells, which were used as membrane models. This reflected curvature changes working as relaxation mechanisms of the curvature stress generated by the partition of the drug in the outer regions of the bilayer. These evidences suggested that starting from perturbations at the single molecular level, self-organizing phenomena emerge at higher hierarchical levels where finally the pharmacological effect is expressed. The introduction of the concepts of complex systems science and dynamical organization of living systems would contribute significantly to the understanding of the problem of side effects of pharmacologically active drugs. It is amazing how relative becomes in this context the concept of specificity of ligand recognition. New approaches of mesoscale modeling and dissipative particle dynamics may help understanding where the limit is when defining the concept of specificity. In this way self-assembled nanoparticles and films behave as useful tools to explore essential features of the drug-membrane interaction problem.

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S.4.**IDENTIFICATION AND CHARACTERIZATION OF VIRULENCE GENES OF *MYCOBACTERIUM TUBERCULOSIS* AND EVALUATION OF MUTANT STRAINS IN THESE GENES AS CANDIDATE VACCINE AGAINST TUBERCULOSIS**

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The genome of *Mycobacterium tuberculosis* and *Mycobacterium bovis* harbors four paralogous *mce* genes, all encoded in an operon structure consisting of eight genes. To investigate whether *Mce* proteins are essential for virulence, we generated knock-out mutants in *mce1*, *mce2* and *mce3* operons of *M. tuberculosis* and evaluated their ability to multiply in a mammalian host. The intratracheal route was used to infect BALB/c mice with the Δ *mce3*, Δ *mce2* and Δ *mce1* mutants. After infection, all mice infected with the Δ *mce* mutants survived, while those infected with the wild-type strain died.

Given the variable protective efficacy generated by *M. bovis* BCG (Bacillus Calmette-Guerin), there is a concerted effort worldwide to develop better vaccines that could be used to reduce the burden of tuberculosis. Rational attenuated mutants of *Mycobacterium tuberculosis* are vaccine candidates that offer some potential in this area. In a collaborative research with Dr. Rogelio Hernandez-Pando's group from Instituto Nacional de la Nutrición, México DF, we assessed Δ *mce3* and Δ *mce2* as candidate vaccine against *M. tuberculosis* in mice. Used as subcutaneous vaccines, 60 days before intra-tracheal challenge with H37Rv or a hypervirulent strain of *M. tuberculosis* both mutants induced a higher level of protection than BCG, 72% and 63% of the mice vaccinated with the *mce-2* and *mce-3* mutants, respectively, survived for 16 weeks after the challenge, compared to 30% of those vaccinated with BCG.

The characterization of transcriptional regulation of genes that play critical roles in virulence could not only provide better understanding of the process of pathogenesis but also lead to new ways to prevent or control mycobacterial infection. In order to study the regulatory process involved in *mce* operon expression we obtained a knockout mutant strain of *M. tuberculosis* H37Rv in *Rv1963c* gene that encoding a putative TetR transcriptional factor. The mutation results in a significant increase in the expression of *mce3* genes either *in vitro* or in a murine cell macrophages line, indicating that it is a transcriptional repressor of *mce3* operon. In addition, by using a promoter-*LacZ* fusions approach and a bioinformatics tool we also demonstrated that Rv0586 repress the expression of *Mce2* proteins and a putative endonuclease IV, encoded in the *end* (Rv0670) gene.

S.5.**INTRACELLULAR LIVING OF *MYCOBACTERIUM TUBERCULOSIS*: A MATER OF DEATH OR LIFE***Lerena C, Calligaris S, Colombo MI.**Laboratorio de Biología Celular y Molecular, IHEM-CONICET, Facultad de Ciencias Médicas, Universidad Nacional de Cuyo, Mendoza, Argentina.*

Many bacteria have evolved mechanism to invade host cells and survive in the intracellular environment. *Mycobacterium tuberculosis* lives within the macrophage, a cell whose function is the elimination of microbes. This bacterium interferes with the normal maturation of the phagosome and resides in compartments modified to avoid degradation by hydrolytic enzymes. *Mycobacterium*-containing phagosomes lack the proton pump, thus preventing acidification. We have evidence that in cells incubated under conditions that stimulate the autophagic pathway mycobacterium-containing phagosomes acidify and acquire markers of lysosomal compartments resulting in a loss of bacterial viability. Similar results were obtained in cells incubated under full nutrient media but in the presence of rapamycin, a drug that induces autophagy. We have also analyzed the intracellular fate of *Mycobacterium marinum*, a natural pathogen of fish that occasionally affects humans causing the “fish tank granuloma”. Similar to *M. tuberculosis* the survival of this bacterium was hampered in host cells subjected to conditions that increase autophagy. With the purpose of finding some new molecules that might modulate the intracellular fate of pathogenic mycobacteria we analyzed the behavior of these pathogens in cells overexpressing proteins involved in the autophagy pathway. Our results indicate that autophagy induction counteracts the intracellular trafficking block imposed by mycobacteria and results in a harsh intracellular environment for *Mycobacterium* replication. These observations suggest that avoiding the autophagic pathway is a matter of death or life for *Mycobacterium* and highlight the significance of this pathway as a target for controlling the persistence, latency, and transmission of tuberculosis.

S.6.**LIGAND GATED ION CHANNELS: FROM STRUCTURE TO PATHOLOGY***Bouzat C.**Instituto de Investigaciones Bioquímicas de Bahía Blanca (INIBIBB). Bahía Blanca, Argentina. E-mail: inbouzat@criba.edu.ar*

Ligand-gated ion channels, such as nicotinic (AChR) and serotonin 5HT₃ receptors, mediate rapid responses throughout the nervous system. The molecular mechanism of activation is not known. However, its modification leads to pathological processes. To determine how the binding of agonist to the extracellular region is coupled to the opening of the ion pore, we constructed chimeric subunits combining the extracellular domains of the Acetylcholine Binding Protein or neuronal α7 AChR with the transmembrane region of 5HT₃. The analysis of single-channel and macroscopic currents of the chimeric receptors revealed that a network of loops located at the interface between the binding and transmembrane domains allows functional connection between both domains and it governs the shape and strength of the synaptic response. *C. elegans* is a non mammalian model for the study of synaptic transmission, for testing drugs, and it is also a model of parasitic nematodes. Nematode muscle AChRs are targets for anthelmintic drugs, which act as potent agonists. We showed that, in contrast, they are partial agonists of mammalian AChRs, and we identified residues located at different faces of the binding site that govern the selectivity of anthelmintic drugs for AChR subtypes. We described for the first time at the single channel level the activation of *C. elegans* muscle AChRs by ACh and anthelmintic agents and showed functional changes in mutant worms.

S.7.**REGULATORY MECHANISMS OF cAMP LEVELS IN LEUKEMIC CELLS OR HOW TO PREVENT CELL DIFFERENTIATION***Davio C.**Laboratorio de Radioisótopos, Facultad de Farmacia y Bioquímica, Universidad Nacional de Buenos Aires.*

Cancer constitutes the second cause of death in the world. So it is highly relevant to elucidate the molecular mechanisms that induce cells to loose their normal maturation program with the aim to discover new therapeutic targets and therefore more clinically efficient drugs. The induction of cell differentiation proved to be an important therapeutic tool for hematopoietic malignancies, particularly leukemia. In this respect, histamine has modulatory properties on cell proliferation and differentiation acting through specific GPCRs. In leukemic cells, histamine causes great increases in cAMP through the activation of H2 receptors. Our studies in leukemic cell lines clearly reflect the importance of time and magnitude of the cAMP response on cellular differentiation. Processes such as desensitization, degradation and cyclic nucleotide efflux tightly regulate this response. We showed that by modulating diverse components of the rh2 signaling pathway as GRK2-mediated receptor desensitization, cAMP efflux through multidrug resistance proteins (MRP4 and MRP5) as well as the magnitude of cAMP response it is possible to induce leukemic cell differentiation. These findings provide new insights that may eventually contribute to the development of new therapeutic strategies for cancer treatment.

S.8.**MAPKS REGULATE GENE EXPRESSION AT MULTIPLE LEVELS**

Coso OA, Degese MS, Hochbaum D, Coluccio Leskow F, Abadia B, Naipauer J, Tanos TB.

IFIBYNE-CONICET. Depto. de Fisiol. Biol. Mol. y Celular, Facultad de Ciencias Exactas y Naturales, UBA.

Living cells react to extracellular signals eliciting a variety of responses that include several biochemical changes that may lead to morphological modifications and even change cell fate. These are performed by changes in the activity of pre-existing proteins or in the quality and quantity of proteins present arising from alterations in the pattern of gene expression. While signals are sensed mostly at the cell surface, the genes, responsible for protein expression, lie in the cell nucleus. The transduction of signals between the cell membrane and the nucleus is performed by a series of molecular events that involve the interaction of receptors, GTP binding proteins, adaptors, protein kinases and transcription factors as major players conforming what is known as a signaling pathway or cascade.

We have focused on the characterization of members of the cascades that regulate AP-1 activity, a transcription factor composed mainly by the products of the *c-jun* and *c-fos* genes.

We will show how different MAP kinase (MAPK) signaling pathways influence AP-1 activity acting at the gene promoter level, phosphorylation of the newly produced proteins and also by altering the quantity and quality of specific mRNAs present. We understand that all these processes, mRNA production, transcription factor activation, mRNA processing and mRNA stability; are regulated by analogous mechanisms that involve proteins that bind to nucleic acids, and have their activities regulated by signaling cascades.

S.9.**IMPORTANCE OF VEGETAL ANATOMY IN QUALITY CONTROL OF MEDICINAL PLANTS**

Cortadi AA.

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Medicinal plants are an important option in health problems, either as a local validated resource in primary care or as industrialized phytotherapy. WHO recommends the inclusion of traditional components, and utilization of local resources, such as medicinal plants in health programs. Basic condition is quality, not only intrinsically, but supported by reproducibility of security and efficiency parameters, of main importance in this type of therapeutic agents, since they are complex systems. We present anatomical studies (AS) to establish the micrographic parameters, and quality control and standardization of crude drugs only of Argentina and Latin America native flora, well-known as medicinal ones. As an example we present the anatomy of stems & leaves of *Passiflora caerulea* L. (*Passifloraceae*) substituted by a native species, *Cucurbitella asperata* (Gill.) Walp. Anatomical difference was established through hairs presence in *Cucurbitella asperata*, absent in *Passiflora caerulea*; *Cecropia adenopus* Mart. ex Miq. (*Cecropiaceae*) & *Tetrapanax papyrifer* (Hook.) C. Koch (*Araliaceae*). Distinctive anatomical character was the presence of trichome & stomata. AS of *Uncaria tomentosa* (Willd. ex R. & S.) DC. & *Uncaria guianensis* (Aublet) J. F. Gmelin cortex, showed differences among fibers longitude, calcium oxalate crystals & starch grains; *Pffafia glomerata* (Spreng.) Pedersen, *Pffafia paniculata* (Mart.) O. Kuntze, widely used in Brazil, have roots with unusual structure. Both species are differentiated by peridermis, ring number of vascular bundles, & cell size. *Tabebuia impetiginosa* (Mart. ex DC.) Standl. & *Tabebuia heptaphylla* (Vell.) Toledo, differences between internal cortex are evidenced by phloem fibers longitude, sclereids & presence or not of oxalate crystals, foliar architecture is decisive. *Achyrocline satureioides* (Lam.) DC., *Achyrocline flaccida* (Weinm.) DC. and *Gnaphalium gaudichaudianum* DC. Characteristics are bracts exomorphology and number of trichome glands. The species' AS are rapid, simple and efficient.

S.10.**THERAPEUTICAL VALIDATION FOR MEDICINAL USE OF *BACCHARIS GRISEBACHII* HIERON (ASTERACEAE) FROM SAN JUAN PROVINCE, ARGENTINE***Feresin GE.**Instituto de Biotecnología–Instituto de Ciencias Básicas Universidad Nacional de San Juan, Avda. Libertador Gral. San Martín 1109 oeste. San Juan. CP: 5400. E-mail: gferesin@unsj.edu.ar*

The aerial parts of the shrub *Baccharis grisebachii* Hieron (Asteraceae), known as “quilchamalí” in the andean provinces in Argentina, are recommended in infusions to treat gastric ulcers, as a digestive, local antiseptic and cicatrizant. We were carrying out the study of the chemical and biological activities of this species, in order to the therapeutical validation in the traditional medicinal use from San Juan Province in Argentina during several years. Studies in mice showed no toxic effects, mortality or side-effects of *B. grisebachii* up to 8 g kg⁻¹. It showed, as well as, anti-inflammatory, antipyretic and analgesic significant effects in animal models. The 20% infusion presented citoprotective gastric effect in mice. The exudate and seriated extracts showed activity as free radical scavengers and inhibited lipoperoxidation in erythrocytes. The most active free radical scavengers measured by the DPPH decoloration assay were the *p*-coumaric acid derivatives drupanin and trans-ferulic acid O-hexan-3-onyl-ether and the flavonoid quercetina. The activity towards the superoxide anion was mainly due to the flavonoid constituents. 5,7,4'-Trihydroxy-6-methoxyflavone and quercetin. The xanthine oxidase inhibitory effect of the extracts can be related with the *p*-coumaric acid derivatives, drupanin, 4-acetyl-3,5-diprenylcinnamic acid and trans-ferulic acid O-hexan-3-onyl-ether. Both *p*-coumaric acid derivatives and flavonoids inhibited lipoperoxidation in erythrocytes. The highest activity was found for the *p*-coumaric acid derivatives 4-acetyl-3-prenyl-ethoxycinnamate, 3-prenyl-4-(4'-hydroxydihydrocinnamoyloxy)-cinnamate and trans-ferulic acid O-hexan-3-onyl-ether and the flavonoids 5,7,4'-trihydroxy-6-methoxyflavone, quercetin, 5,7,4'-trihydroxy-6,3'-dimethoxyflavone and 5,7,4'-trihydroxy-6,8-dimethoxyflavone. The resinous exudate showed activity towards dermatophytes and bacteria. The compounds isolated, 3-Prenyl-*p*-coumaric acid and 3,5-diprenyl-*p*-coumaric acid were the best active towards *Epidermophyton floccosum* and *Trichophyton rubrum*. Antimicrobial and chemical composition the essential oil of *Baccharis grisebachii* showed activity against all the bacteria of the panel which includes methicillin resistant *Staphylococcus aureus* and was active against all fungi assayed, being *Trichopyton mentagrophytes*, *T rubrum*, *Cryptococcus neoformans* and *Aspergillus* spp. the most susceptible species. The major constituents (concentrations higher than 3.5 %) were thymol (18.3%), thymol methyl ether (16.7%), thymyl acetate (10.9%), alpha pinene (7.2%), alpha humulene (7.2%) and globulol (3.7%). The results of the study on antimicrobial activity, gastric cytoprotective, antioxidant, antiinflammatory and antipyretic effects support the traditional medicinal use of *Baccharis grisebachii* in San Juan Province (Argentina).

S.11.**BIOTECHNOLOGICAL STRATEGIES IN THE PRODUCTION AND TRANSFORMATION OF PLANT SECONDARY METABOLITES***Kurina Sanz M.**INTEQUI-CONICET. Área de Química Orgánica Facultad de Química, Bioquímica y Farmacia. Universidad Nacional de San Luis. Chacabuco y Pedernera. 5700-San Luis. Argentina. E-mail: mkurina@unsl.edu.ar*

Plants are a valuable source of a wide range of secondary metabolites, which are used as pharmaceuticals, agrochemicals, flavours, fragrances, colours, biopesticides and food additives. This is one of the causes of the unsustainable utilization of natural resources. The agronomic culture of many wild species is not always a viable alternative. Biotechnology offers an opportunity to exploit the cell, tissue, organ or entire organism by growing them *in-vitro* to get desired compounds. One of the advantages of this technology over the conventional agricultural production is the independency of geographical and seasonal variations, as well as the possibility to achieve a defined production system, which ensures the continuous supply of products, uniform quality and yield.

Chemist and biotechnologist have developed research projects that bring into play the knowledge of the different allelopathic strategies that plants use to interact with other components of their environment. Other interesting point is to emulate these abilities by using cell cultures of different organisms as source of enzymes to transform natural or xenobiotic substrates into more valuable compounds in environmental friendly procedures.

Our group has been established the *in-vitro* cell cultures of some plant endemic genus from the Cuyo Region in Argentina. In that sense we have studied the accumulation of their main secondary metabolites under different culture conditions and stress situations.

We have also studied the production of high-value food metabolites by biotransformation of plant secondary metabolites and derivatives. The high reaction specificity, together with the low substrate selectivity that show the enzymes of interest result in a great advantage to catalyze regio, chemo and stereoselective reactions on interesting natural products.

Financial assistance: UNSL (Project 22Q-505) and CONICET (PIP 6228).

1.**ACUTE TOXICITY OF *Plantago major* IN MICE**

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Plantago major is popularly known as “llantén”. The antiulcerogenic activity of *Plantago major* was demonstrated on different experimentally induced gastric ulcer models in rats. The aim of the present study was to determine the acute toxicity of the infusion of *Plantago major* in mice. Infusion (10%) of the aerial parts was prepared.

Acute toxicity was produced according to the method of acute toxicity, Guidenace No. 423, OECD. Twenty four hours before the experiments, Rockland mice were fasted. *Plantago major* infusion was administered to five lots of six mice (3 males and 3 females), *p. o.*, 0.2 ml/animal. The control group received water and the test groups received infusion (5, 50, 300 and 2000 mg/kg). The animals were observed carefully every 2 days to record toxic manifestations, and also to measure body mass. After the 14 days experimental period mice were sacrificed. The organs were observed macroscopically and the relative weights (organ/body) were determined.

No toxic symptoms or death occurred in each infusion group. The general conditions of all mice were also normal. There were no significant alterations in body weight during the experiment. The relative weights of kidney, liver, lung, spleen and heart were not statistically different from control (ANOVA and posterior comparison by Tukey-Kramer). The infusion of *Plantago major* did not present acute toxic effects.

2.**ACTION CITOPROTECTOR GASTRIC OF PLANTAGO MAJOR**

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The ethanol affects the mucous gastric by producing severe damage in its structure, which leads to the production of oxygen's free radicals that they would initiate the process of lipoperoxidation. In the present work is studied this mechanism by means of the etanolic extract of the *Plantago major*'s action on the mucous gastric insulted. The extract was evaluated in rats Wistar under protocol and standardized conditions. Ethanol 96° was administered across orogastric probe, as offensive substance. There were in use 18 rats distributed in 3 lots, lot 1(control), lot 2 (injury), lot 3 (experimentally). The techniques used as warning of the production of ROL are Malonildialdehido (MDA) and total Antioxidants (TAS) as mechanism of defense. The removed stomachs were used for the study in optical microscopy. The obtained results were: MDA (nmol/ml/min) L1: 0.39 ± 0.11, L2: 1.30 ± 0.30, L3: 0.56 ± 0.22. TAS (mmol/l), L1: 1.85 ± 1.05, L2 1.03 ± 0.55, L3 1.7 ± 0.39. The optical microscopy of the samples of stomach showed in the L2, the mucous gastric presented loss of its histoarchitecture. The L1 and L3 in return did not show histological alterations. The biochemical obtained parameters indicated a correlation with the studies of optical microscopy, of what it concludes that etanolic extract etanolic of the Pm exercises an effect citoprotector on the gastric mucous insulted.

3.**DENGUE-2 VIRUS ENTRY INTO VERO CELLS**

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Dengue virus (DENV) produces a spectrum of clinical illness ranging from silent infection to either dengue fever or more severe and fatal diseases. Different mechanisms of virus entry into cell, such as receptor mediated endocytosis and direct fusion at the plasma membrane, have been described. In the present study we analysed the effect of inhibitors of different endocytic pathways on DENV-2 entry into Vero cells by virus yield inhibition assays. We assayed inhibitors of receptor mediated endocytosis (monodansilcadaverine), clathrin mediated endocytosis (chlorpromazine and sucrose), caveola mediated endocytosis (nystatin) and macropinocytosis (amiloride). We further evaluated the effect of lysosomotropic agents (ammonium chloride and concanamycin A), and cytoskeleton disrupting agents (cytochalasin D, colchicine and nocodazole). A significant reduction of virus production was observed in cultures treated with amiloride, lysosomotropic substances, nocodazole or colchicine. Electron microscopy studies showed that virus particles are engulfed by cell structures that resemble the macropinocytosis process. Taken together, these results suggest that DENV-2 entry into Vero cells occurs by macropinocytosis and that the internalization pathway is dependent on acid pH and microtubule integrity.

4.**MORPHOLOGIC VARIATIONS OF THE PITUITARY PARS INTERMEDIA OF VISCHACHA DURING LONG AND SHORT PHOTOPERIOD**

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The viscacha (*Lagostomus maximus maximus*) is a rodent of nocturnal habits and photoperiod-depend seasonal reproduction. The pituitary pars intermedia (PI) of *Lagostomus* shows a developed parenchyma, with granulated and non-granulated (folliculo-stellate; FS) cells, typical follicular structures and scanty blood vessels. The aim of this work was to study the morphology of PI in adult male viscachas during long (summer) and short (winter) photoperiod. Histochemical (PAS), immunohistochemical (antibodies anti- α -MSH, anti-ACTH, anti-S-100 and anti-GFAP), and morphometric (area percentage, cells and follicular number, major colloidal diameter, major cellular and nuclear diameters to immunolabeling cells) studies were carried out. The FS cells show cytoplasmic and nuclear staining for anti-S-100 and cytoplasmic staining for anti-GFAP. These cells are distributed throughout the parenchyma and they are originating PAS-positive follicles. There are granulated cells near follicles and limiting with the pars nervosa, where there are important blood vessels. Most parameters show a significant decrease during short photoperiod. Our results demonstrate that the PI parenchyma of adult male viscachas is composed by granulated and FS cells closely associated. The PI morphology and histoarchitecture varies according to the different photoperiods.

5.

ANTIFUNGAL ACTIVITY OF ESSENTIAL OIL FROM *Satureja parvifolia*

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The aerial parts *Satureja parvifolia*, known popularly as “muñamuña” are used in San Juan, province, Argentina in infusion to treat gastric troubles, stomach aches, hemorrhages and as stimulant. No reports related to the antimicrobial activity and chemical analysis of the essential oil constituents from *Satureja parvifolia* collected in San Juan has been found in the literature. The essential oil of the aerial parts of *S. parvifolia* was examined by GC and GC-MS and evaluated for its antimicrobial properties by the agar dilution method against dermatophytic fungi, filamentous fungi and yeasts. Fresh aerial parts (300 g) collected in San Juan Province, were subjected to hydro distillation for 2 h in a modified Clevenger apparatus. The oil yield was 0.8% (2.4 ml). Essential oil was active against dermatophytes *Trichopyton mentagrophytes* and *Trubrum* (MIC = 125 µg/mL). It is important to keep in mind that these two fungi are responsible for approximately 80-93% of chronic and recurrent non-scalp skin infections in human beings. *Microsporum gypseum* (MIC=32.5 µg/mL) was the most susceptible dermatophytes species assayed.

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6.

CHEMICAL COMPOSITION AND ANTIMICROBIAL ACTIVITY OF ESSENTIAL OIL FROM *Tagetes mendocina*

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Previous chemical work on Argentinian *Tagetes mendocina* (Asteraceae) has been reported on chemotaxonomy, distribution, antibacterial and free radical scavengers activity. No reports related to the chemical analysis and antimicrobial activity of the essential oil constituents from *T. mendocina* collected in San Juan has been found in the literature. The essential oil of the aerial parts of *T. mendocina* was examined by GC and GC-MS and evaluated for its antimicrobial properties by the agar dilution method against dermatophytic fungi, filamentous fungi, yeasts and bacteria. Fresh aerial parts collected in San Juan Province (500g) were subjected to hydro distillation for 4 hour in a Clevenger-type apparatus. The oil yield was 0.66% (3.3 mL). Main essential oil constituents were identified representing 97.3% of the total. (E) β ocimene, (Z)-tagetone, (E)- tagetone, (Z) ocimenone, α pinene and (E) ocimenone were found to be the major components. The oil showed MIC values of 250-500 µg/mL against *Microsporum gypseum* C1152000, *Trichophyton rubrum* C1132000 and *Trichophyton mentagrophytes* ATCC9972. On the other hand essential oil showed a capacity of inhibiting eleven both sensitive and resistant strains including *Staphylococcus aureus* methicillin-sensitive ATCC 29213, *S. aureus* methicillin-resistant ATCC 43300, *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853 with MIC between 500 and 1000µg/ml.

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7.

ANURANS ASSOCIATED TO PLANT COMMUNITIES IN URBAN WATER BODIES OF SAN LUIS CITY (ARGENTINA)

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Anurans use water bodies and surrounding habitats with hydrophyte plants as refuge, reproduction, feeding and rest sites. The aim of this study was to explore the relation between composition and structure of plant communities and anuran abundance and diversity, in urban water bodies of San Luis city (Argentina). Anuran and plant communities were sampled simultaneously from October to March. Three anuran species were registered: *Chlaenius arenarum*, *Leptodactylus mystacinus* and *Odontophrynus americanus*. Higher richness and reproductive activity of anurans were associated to plant communities with high floristic and structural diversity and high proportion of native taxa. Larval populations were more abundant in water bodies with submerged vegetation and/or occurrence of pleustophyte species not colonizing the entire water surface. Vegetation present in water bodies inhabited by anurans included, predominantly, communities of the classes *Lemnetea*, *Potametea*, *Bidentetea tripartitae*, *Phragmito-Magnocaricetea Polygono-Poetea annuae y Stellarietea mediae*. Patterns detected are related to particular site characteristics and alterations produced by anthropization in urban environments.

8.

ALPHA-MACROGLOBULINE (α M) PLASMATIC CONCENTRATION: A SIMPLIFIED MODEL OF ITS REGULATION

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Introduction: Among the proteins which belong to the family of the macroglobulin, the alpha-macroglobulin (α M) is a normal antiprotease of the serum whose inactivation by trypsin determines structural changes that accelerate its uptake by tissue receptors.

Objective: In the present work an iterative method is used to model the change of the plasmatic concentration since the control of its plasmatic concentration is unknown.

Methodology: Let $d(\alpha M)/dt = f(\alpha M) - g(\alpha M)$ where f is the secretion rate and g is the inactivation rate of αM in the plasma. In order to analyze the dynamics of the process functions f and g are linearized at the basal concentration with slopes k_1 and k_2 . As regards the dynamics of the process, the balance is reached when $f(\alpha M) = g(\alpha M)$.

Results: If $k_1 < k_2$ the system becomes stable and allows to determine if the final concentration of αM is greater, lower or equal to the basal value. Otherwise, case the system becomes unstable.

9.

VITELLOGENESIS AND FOLLICULAR REGRESSION IN *DIPETALOGASTER MAXIMA*, A VECTOR OF CHAGAS' DISEASE

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In insects, reproduction entails an accurate compromise between factors promoting vitellogenesis and those causing follicular regression. In Chagas' disease vectors, these aspects have been poorly investigated. In this work, we have analyzed the vitellogenic phase of *D. maxima* as well as morphological and cellular changes occurring in ovarian tissue during follicular regression. For the study, we have used western-blot, ELISA and immunohistochemical techniques. In addition, electron and fluorescence microscopy were employed to assess cellular changes. Results showed that: [a] vitellogenin protein expression in fat body and the levels in hemolymph were maximal between days 4-20 post-feeding; [b] during the vitellogenic phase, ovarian tissue showed a typical asynchronous development and some differential deposition of the protein according to the size of the follicles; [c] ovarioles entering in regression (from day 15 post-vitellogenesis) were poorly developed, and follicles presented nurse cells with different degree of vacuolization. In degenerating follicles, death of follicular cells occurred by apoptosis and necrosis. We conclude that in this vector, follicular atresia is complex. It can serve to promote resorption of some oocytes in order to sustain the development of younger follicles during the second vitellogenic cycle.

10.

IN VITRO ANTIOXIDANT ACTIVITY IN METHANOL EXTRACT OF *Sechium edule* (Jacq) Sw

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INTRODUCTION: *Sechium edule* is a neotropical species that a valuable although little explored alimentary resource. The vegetables produce great quality of chemical fractions, that activity as antioxidants in order to control the oxidative stress. Therefore, it is necessary to investigate *in vitro*, these properties before considering it food with antioxidant functions.

OBJECTIVE: the main aim of the present work.

METHODS: Extraction of total phenols (Singleton VL *et al.*, 1965) DPPH assay (Brand – Williams *et al.*, 1995), Scavenging activity against nitric oxide (NO test), (Marcocci L *et al.*, 1994). Samples: flour of fruit (f), seeds(s) and pulp(p).

RESULTS: methanol extract was found to be effective in scavenging DPPH to express how inhibition percentage (%): 88,87 (f); 89,13 (s) and 90,42 (p). Inhibition NO %: 47,51 (f); 55,62(s) and 50,01 (p)

DISCUSSION: Previous studies related with the nutritional composition by means of biological experiments claim of the good quality of this vegetable. The obtained findings encourage us to continue investigating the antioxidant properties of *Sechium edule* in order to recommend it as a food with functional characteristics.

11.

MOLECULAR CHARACTERIZATION AND CRYSTAL MORPHOLOGY OF INDIGENOUS ENTOMOPHATOGENIC *Bacillus* STRAINS AGAINST *Spodoptera frugiperda*

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Introduction: *Bacillus thuringiensis* (*Bt*) are the microorganisms most widely used in biological control of insect pest. These bacteria produce entomopathogenic crystals (Cry) during sporulation showing different morphologies according to *Bt* strains.

Objectives: To determine the morphology of crystals produced by three native *Bt* strains active against larvae of *Spodoptera frugiperda* (*Sf*) and to characterize the microorganisms molecularly.

Materials and methods: Crystals were studied by scanning electron microscopy. Sporulated cells were removed from the agarized medium, washed and placed on a micro slide. The 16S rDNA gene was amplified from genomic DNA by PCR.

Results and conclusions: Morphological observations showed that the three strains contained bipyramidal crystals of two different sizes. This feature may resemble those against lepidopteran targets. The 16S rDNA analysis identifies the isolations as *B. thuringiensis* subspecies. The sequences were deposited in the Gene Bank, access N°: RT EF638795, LSM EF638796 and LQ EF638798. These results complement the knowledge on these native pathogenic strains with promissory characteristics for *Sf* biological control.

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12.

CILIARY NEUROTROPHIC FACTOR (CNTF) STIMULATES CEPHALIC NEURAL CREST CELLS MIGRATION

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The molecular bases that regulate the oriented cell migration are little known, even if chemotactic signals have been proposed as a possible mechanism of modulation. Mesencephalic neural crest cells (NCC) of vertebrate embryos emigrate from the neural tube in a dorso-lateral way and change direction in cephalic sense towards the optic vesicle, where they differentiate in neurons and glia of the ciliary ganglion. Results of our laboratory showed that the chemokine Stromal Cell Derived Factor-1 (SDF-1) and the trophic factor Neurotrophin-3 (NT-3) induce NCC chemotaxis, whereas Ciliary Neurotrophic Factor (CNTF) may be another candidate involved in the above mentioned mechanism. In the present work, NCC exposed to *in vitro* concentration gradients of CNTF and registered by real-time video-microscopy for 6 hours, showed a significant increase of the curvilinear and linear distances travelled, as well as cellular speed, dependent on CNTF concentration. Nevertheless, chemotactic response was not observed, expressed neither as chemotactic index nor proportion of orientated cells. The parameters of cellular area and perimeter showed an increase dependent on CNTF concentration, indicating possible changes of the cell-substratum interaction during the migration, without affecting the shape factor. These results suggest that the chemokinetic effect of CNTF play a role during NCC colonization of the optic vesicle, in a multifactorial system of orientation that involves, at least, SDF-1 and NT-3 signals.

13.***IN VITRO PORCINE CUMULUS EXPANSION AND OOCYTE MATURATION***

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We have previously observed that cumulus-oocyte complex (COC) quality is related to both nuclear and cytoplasmic maturation. The aim was to evaluate oocyte meiotic and cytoplasmic maturation according to cumulus expansion features. COCs recovered from slaughtered gilts were selected according to cumulus characteristics in classes: A₁ complete-multilayer-dark and A₂ complete-multilayer-light. COCs were matured in medium 199 for 48h and then classified in fully expanded, partially expanded or spontaneous partially denuded. Oocytes were used to evaluate nuclear maturation (metaphase II) or incubated in TBMm with fresh spermatozoa during 18h. Cytoplasmic maturation was evaluated by sperm head decondensation and/or pronuclear formation. In A₁ and A₂ no differences were found between the percentages of fully expanded and partially expanded COCs, but spontaneous partially denuded were lower ($p<0.05$). There were no differences in the percentages of nuclear maturation among cumulus expansion types in both classes. In class A₁ no differences were observed in the percentages of cytoplasmic maturation among types of cumulus expansion, but in class A₂ spontaneous partially denuded oocytes showed lower cytoplasmic maturation respect to fully expanded ones ($p<0.05$). Oocyte nuclear maturation would be independent of cumulus expansion. Cumulus quality seems to be the main factor affecting cytoplasmic maturation instead of degree of cumulus expansion.

14.***PHOTODYNAMIC INACTIVATION IN YEAST***

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Photodynamic inactivation (PDI) is a novel therapeutic modality in the fungal infections treatment. It involves the pre-treatment with a sensitizer, which it accumulates in microbial cells. The exposure to visible light leads to the generation of singlet oxygen (${}^1\text{O}_2$), which produces photodamage and destruction of cells. In this work was studied the photodynamic properties and the PDI of *Candida albicans* sensitized by porphyrins with different patterns of substitution. The cultures were treating with different concentration of sensitizer [1-5 μM] by 30 min at 37°C in dark. Under these conditions, three sensitizers were innocuous in dark. The cationic porphyrins (A₃B³⁺ and TMAP⁴⁺) show an incorporation of $\sim 3.4 \pm 0.2$ nmol/10⁶ cells. After irradiation, the cell viability was depending of the concentration used. When the cultures are irradiated for 15 min, A₃B³⁺ and TMAP⁴⁺ (5 μM) produce a diminution of ~ 4 log ($>99.995\%$), while negligible effect is found using TPPS⁴. Fluorescence microscopy studies allow observing the distribution of sensitizers in the cells. A₃B³⁺ and TMAP⁴⁺ are localizing with irregular distribution in all cell, whereas the fluorescence of TPPS⁴ is very weak according to its low cellular binding. The studies indicate that cationic porphyrins have potential applications as phototherapeutic agents in yeast inactivation by PDI.

15.***BETULINA, METABOLITE ISOLATED FROM *Colliguaja integerrima* WITH DIURETIC ACTIVITY***

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Colliguaja integerrima (Euphorbiaceae), popularly known as “coliguaya”, is a plant that grows in western Argentina, from La Rioja to Santa Cruz. Plant species of the same genus are known to have diuretic activity (Del Vitto *et al.*, 1997). Betulina was isolated as major constituent from aerial parts of *Colliguaja integerrima*. This study was designed to determine the diuretic activity of the *C. integerrima* and betulina in Wistar rats, using the Lipschitz method (1943).

The treated rats received 5% infusion (*p. o.*) of the aerial part, methanolic extract (250 mg/kg, *p. o.*) or betulina (100 mg/kg) of *C. integerrima* or furosemide as standard drug (10 mg/kg, *p. o.*). The control group received only the NaCl isotonic solution (50 ml/kg, *p. o.*). Urinary volumetric excretion, and urinary levels of Na⁺ and K⁺ were measured in 3 hours diuresis. The urinary concentration of sodium and potassium were determined by flame atomic emission spectroscopy-FAES. All values were expressed as the mean \pm SEM. Student's *t*-test was performed to evaluate the statistical differences between the control and the experimental samples. The lots treated with betulina and infusion of *C. integerrima* showed diuretic activity ($p<0.05$ vs. control). The *Colliguaja integerrima* effect could be due, in part, to the presence of betulina in the infusion. This was also demonstrated by capillary electrophoresis studies (Álvarez ME *et al.*, 2005).

16.***EFFECTS OF EXTREME PHOTOPERIODS ON THE SECRETORY PRODUCT AND PRESENCE OF α -SUBUNIT IN SPECIFIC CELLS OF THE PARS TUBERALIS (PT) IN THE RAT***

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The α -subunit is a component of the glycoproteic hormones and could be part of the secretion of the specific cells of the PT. These cells have abundant receptors for melatonin (mel), suggesting their participation in the endocrine activity related to the photoperiod. The aim of the present study was to evaluate the presence of the α -subunit in the rat PT in connection with the identification of the secretory product with a specific antibody. Three groups of mature males Wistar rats were formed (n=12): Group 1: completes darkness, Group 2: continuous illumination, Group 3 (12 L / 12 D). After 5 weeks the PTs were collected, fixed 48 h in Bouin and being processed for routine histology. The peroxidase-antiperoxidase method was carried out with a specific antiserum (FB02) and another anti α -subunit. With FB02 in the group 1 and 3 group the secretory product was restricted to the paranuclear region. Immunoreaction was not detected in the group 2. When the samples were immunostained with anti α -subunit in the 3 groups, a diffuse immunoreaction was observed in the cytoplasm. The present results could indicate that the secretory product recognized by FB02 follows a behaviour related with the secretion of mel. On the other hand, the α -subunit was present in all the groups, indicating that it would be part of a compound that is not the recognized by the FB02 antibody. Further studies will be conducted to check it.

17.**POTENTIAL USE OF PEROXIDASES FOR 2,4-DICHLOROPHENOL BIOREMEDIATION**

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Peroxidases (Px) are enzymes that have been applied in the removal of phenolic compounds like 2,4-dichlorophenol (2,4-DCP), which is a highly toxic environmental pollutant. The aim of this study was to perform a biochemical characterization and a kinetic study of 2,4-DCP removal by using different fraction of Px, obtained from tobacco wild type (wt) and transgenic hairy roots which express two basic Px from tomato (TPX1 and TPX2). The different Px fractions (total crude extract (TCE), soluble Px (SP) and ionically bound to cell wall Px (IBP)) were obtained using buffers with low and high ionic strength. Pxs were analyzed by isoelectric focusing and the zymograms showed the presence of different basic, neutral and acidic isoenzymes. The optimal conditions for 2,4-DCP removal were established for each fraction of Px. The optimal pH for the activity of Px groups obtained from tobacco wt hairy roots was 6 while for transgenic hairy roots the optimal pH was 7. The optimal H₂O₂ concentration was 0.2 mM. The Px present in TCE and IBP of the transgenic hairy roots, showed lower values of Km and Vmax than those from wt hairy root extracts. These kinetics parameters were similar for the SP from wt and transgenic hairy roots. The higher catalytic efficiency was obtained for IBP fraction of transgenic hairy roots. These results suggest that IBP fraction could be used for bioremediation of 2,4-DCP from aqueous solutions.

18.**EFFECT OF BIOFERTILIZATION WITH *Azospirillum* AND UREA, IN SUGAR MORPHOLOGIC CANE PARAMETERS CV. RA. 87-3, IN BELLA VISTA, TUCUMÁN**

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It is known that *Azospirillum brasiliense* produces fitohormone and fixes N₂. Inoculation studies were made in sugar cane culture plants, cv. RA 87-3 to determine the effect of biofertilization with *Azospirillum* sp., urea and combination of both in morphologic parameters, in Bella Vista, between 2006/07. Experimental design was: randomized block with 4 treatments: T1: Control. T2: Urea 90 (UF.ha⁻¹). T3: *Azospirillum* sp. 10⁸ x 3 doses. T4: Urea 60 + *Azospirillum* sp. x 10⁸ x 3 doses, and 4 repetitions. It was determined: length and diameter of stem of 4 plants by parcel, in primary stems, secondary and tertiary, in 5 dates of the cycle of culture; wide length and of leaves from the leaf plus one according to nomenclature of Kuijper, to the lowest green leaf. With the data of wide length and of leaves, plus a coefficient, the Individual Foliar Area (IFA) of each plant was obtained. In order to obtain total Foliar Area (TFA) the total number of stems of each parcel was taken. It was made ANOVA and Test of Tukey (*p*=0.05). Were not founded differences in length and diameter of stem. It was detected significant differences in TFA (cm²) in 5th date of observation (T3: 212,20 respect to the control (156,80). It is inferred that *Azospirillum* sp. it showed to promotional effect of the growth when stimulating the development of the foliar area in the RA 87-3 sugar cane cultivar.

19.**THE FUNCTION OF THE UTERO TUBAL JUNCTION AS SPERM RESERVOIR IN LAMA**

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In *Lama glama* ovulation occurs approximately 36 hours after mating, hereby it would be crucial to the fertilization success the maintenance of the sperm viability by the establishment of sperm reservoirs. We have studied the sperm reservoirs formation by sperm adhesion to the oviductal mucosa. The oviducts of adult lama females were obtained at 6, 18, 24, 28 and 34 hours post mating. Utero Tubal Junction (UTJ) and caudal isthmus were studied by light microscopy, electron microscopy and Scanning electron microscopy. Sperm were observed adhered to the UTJ epithelial cells at 6, 18, 24 and 28 h pos-mating. Sperm were trapped in a viscous substance and patchy distributed. This substance was absent in the oviducts of non mated females, suggesting a seminal origin. Later, the sperm would detach from the UTJ as they were not observed 34 hours after mating. No sperm were found adhered to the Isthmus cells in all the samples. The sperm interaction was also studied by *in vitro* methods, cocultivating sperm and oviductal vesicles (OV). The sperm binding index (BI) was calculated as the number of sperm adhered/0.5 mm² of OV. The BI of UTJ was found to be higher than Isthmus (*P*<0.0001, *t* test). We conclude that the UTJ could form a sperm reservoir in lama, and seminal substances could be involved in its formation. The existence of a functional sperm reservoir in lama would explain why even 28 h after semen deposition the sperm fertilizing ability is maintained.

20.**PRELIMINARY STUDY OF ORAL INORGANIC ARSENIC (As) ADMINISTRATION IN EXPERIMENTAL ANIMALS**

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Arsenic is a water contaminant in certain areas of the world. Its toxic and cancerous effects on human beings have been proved, with an unsuitable evidence in experimental models. The aim of this work was the study of the toxic effects of chronic consumption of As in Balb-c mice drinking water. 24 8-week mice were chosen and divided into 4 lots: A) As III dissolved in drinking water (150 ppm); B) As V idem (200 ppm); C) Well water (0,1 ppm); D) Free from As drinking water. Throughout the 23 treatment weeks clinical monitoring, body weight control and water input consumption were carried out. A decrease in body weight, corresponded to 41%, 44% and 50% for lots A, B and C respectively in relation to group D. Water consumption was markedly less in lot A, slightly lower in lot B, and slightly higher in lot C (63%, 91% and 116% respectively in relation to the control group). This preliminary data would indicate that the marked weight reduction in lots A, B and C, would not be directly related to the volume of the drunk water but to the presence of the different forms of As contained in it, with no evident clinical changes shown. Further studies will be able to establish As effects on various target organs.

21.**EFFECT OF TRYPANOCIDAL DRUGS ON LIPIDS AND FATTY ACIDS COMPOSITION OF *T. cruzi****Armentano S, Woelke M, Garcia M.**Departamento de Biología Molecular, Fac Cs Exactas Quim Nat, Univ Nac Río Cuarto, Río Cuarto 5800 Córdoba. E-mail: mgarcia@exa.unrc.edu.ar*

Changes in the membrane lipidic composition of *T. cruzi* are decisive factors in its invasion ability toward the host. We have studied lipids and fatty acids composition of *T. cruzi* under ketoconazole (K) effect. Acetate 1-¹⁴C was added to the media and K was added 24 hours later. Protozoans were harvested after five days of growth and then total lipids (TL) were extracted. Phospholipids (PL) and neutral lipids (NL) were resolved by thin-layer chromatography (TLC) and quantified by measuring the radioactivity in a liquid scintillation counter. Fatty acid methyl esters were obtained, resolved by AgNO₃ TLC and the radioactivity was measured in a liquid scintillation counter. The fatty acid pattern was obtained by gas chromatography (GC).

We found that free sterols and sterol esters synthesis decreased 65% and 75% respectively in cultures under K effect. Ketoconazole caused an increase (19%) in the percentage of labelling diunsaturated fatty acids of the TL. In agreement with this, we observed a decrease of 14% in the oleic acid level when the fatty acid composition was determined. These results are indicatives that K could affect the desaturase activities and so modify the *T. cruzi* invasion ability. Thus, our results could contribute to go into depth the research of new therapies for Chagas' disease control.

22.**CHARACTERIZATION OF HMG-CoA LYASE ISOFORM CODIFIED BY HMGCLL1 GENE***Arnedo M¹, Pie-Juste J¹, Campetelli A², Menao-Guilén S¹, Monesterolo N², Santander V², Casale C².**¹Dto. Fisiol. Animal. Fac. Med., Univ. de Zaragoza. ²Dto. Biol. Molecular, FCEFQN. U.N.R.C. E-mail: mariaar82@hotmail.com*

Acetoacetyl-CoA thiolase and 3-hydroxy-3-methylglutaryl-CoA (HMG-CoA) synthase which comprise the HMG-CoA-generating system(s) for hepatic cholesterologenesis and ketogenesis exhibit dual mitochondrial and cytoplasmic localization. In contrast, HMG-CoA lyase (HL), an enzyme unique to the "HMG-CoA cycle" of ketogenesis, appears to be localized in the mitochondrion. The identity of enzymes involved in these processes and their regulation remain unclear. Recently, a HMG-CoA lyase new gene (HMGCLL1) has been cloned in our laboratory. Kinetic analysis showed that the proteic product of HMGCLL1 has a similar behavior to that found in cytosolic fraction of mouse testicle, while HL has a similar kinetic behavior to that found in the same tissue mitochondrion. To determine the sub-cellular localization of HMGCLL1 protein, it was fused to GFP and expressed in HEK 293 cells. Confocal images showed that HMGCLL1 protein colocalizes with microtubules and calreticulin, an endoplasmic reticulum (ER) membrane protein. Biochemical experiments support the idea that HMGCLL1 protein interacts with ER. These results suggest that the new HMG-CoA lyase, product of HMGCLL1 gene, is a cytoplasmic protein that interacts with ER and that ketone bodies could be sintetized in the cytoplasm.

23.**GLYCOGEN STORAGE SITES IN *Chasmagnathus granulatus* UPON HYPERREGULATION: DIFFERENTIAL POSTINGESTA RESPONSE***Artillo R¹, Pinoni SA^{1,2}, Asaro A¹, López Mañanes AA^{1,2}.**¹Dpto Biología, FCEyN, UNMDP, Funes 3250 (B7602AYJ) Mar del Plata, ²CONICET, E-mail: mananes@mdp.edu.ar*

Glycogen (G) storage sites in crustacean decapods vary depending on species, physiological status and stress condition. The aim of this work was to identify G storage tissues of *C. granulatus* at different times post ingesta upon hyperregulation conditions. Adult male crabs acclimated for 14 days at 10‰ salinity were starved for 5 days. After starvation (t_0) crabs were fed. At t_0 and up to 92 h post ingesta G (mg glucose x g tissue⁻¹) was quantified in homogenates from chela muscle (M), hepatopancreas (HP) and anterior gills (AG) by hydrolysis with α -amyloglucosidase (Sigma Chemicals) (0.2ml/ml) for 2,5 h at 55°C. Released glucose from G was determined with a commercial kit (Wiener-Lab AA). Five independent experiments were carried out. ANOVA was used as statistical analysis ($p<0.05$). At t_0 G content was: HP=2.9±1.1, M=32.3±5.0, AG=20.6±1.3. At 23 and 92 h G content increased in GA (34.8±3.5 and 3850±3.3, respectively). At 92 hs G content increased in HP(11.3±2.6). No changes occurred in either M or HP. Our results show that in *C. granulatus* anterior gills constitute G storage sites after ingesta.

24.**INHIBITION OF JUNIN VIRUS MULTIPLICATION BY RNA INTERFERENCE***Artuso MC, García CC, Damonte EB.**Laboratorio de Virología, Depto. Qca. Biológica, Fac. Cs. Exactas y Naturales, Univ. de Buenos Aires. E-mail: caro.artuso@gmail.com*

Junin Virus (JUNV), the ethiological agent of the Argentine hemorrhagic fever, has a single-stranded RNA genome with ambisense expression which encodes five proteins. In previous studies we have demonstrated that the Z arenavirus protein represents an attractive target for antiviral therapy. In this report we study a new alternative therapeutic mechanism where different small interference RNAs are used against the mRNA of Z (Z-siRNAs) to inhibit JUNV multiplication. Their antiviral activity was evaluated by transfecting Vero cells with Z-siRNAs and following infection with JUNV. At 24 h p.i, supernatants were harvested for viral titration and cells were processed concomitantly for RNA extraction and qRT-PCR. Results showed an efficient inhibition of viral replication, analyzed by using specific primers against Z, and a reduced viral yield in those cells treated with Z-siRNAs. Moreover, expression of recombinant Z protein was studied by indirect immunofluorescence, and it was observed a decrease of cell number expressing the viral antigen when cells were cotransfected with Z-siRNAs and pcDNA-Z. These results demonstrated that Z silencing by siRNAs inhibits JUNV multiplication.

25.**RELATIONSHIP BETWEEN T3 AND PROGESTERONE DURING SUMMER AND WINTER IN SOWS**

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In the Intensive Systems of Outdoors Breeding one of the problems is reproductive performance decrease during summer, because the animals are exposed to high temperature, which is impossible controlled. The objective of this work was to determine and to compare reproductive performance and the serum hormone levels of triiodothyronine and progesterone between winter and summer. The serum levels of triiodothyronine and progesterone and the reproductive parameters of the breeding herd were measured during the whole experimental period.

The conception index decreased during summer, with increased irregular return rates. Also there was a decreased in the serum levels of triiodothyronine and progesterone mainly in the sows with 20 days of pregnancy.

There was a positive correlation between T3 and progesterone in the sows with 20 days of pregnancy during the winter.

It could be concluded that the decrease in the serum levels of T3 was related with a decrease in the serum levels of progesterone during summer, which would be related to embryonic mortality and the disruption of early pregnancy as well as the increase in the irregular returns ratio and a decrease in the conception index.

26.**CALCIUM REGULATES THE ION BALANCE DETERMINING SPERM FUNCTION**

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Calcium is absolutely required for sperm, being necessary for motility, capacitation, acrosome reaction (AR) and interaction with the oocyte. Strontium can replace Ca^{2+} in several of those aspects but not in the AR. Recently, we observed that the difference between these two ions is related to an influx of Na^+ . In the present work, we analyzed which transport systems could be involved in this event. Hamster sperm were incubated in regular or modified TALP (with Sr^{2+} instead of Ca^{2+}) for 3 hs and then treated with different agents. We evaluated the spontaneous or zona pellucida (ZP) induced AR by light microscopy on motile cells. The addition of 1 uM Amiloride (Am), an inhibitor of the epithelial Na^+ channel (ENaC), to sperm incubated in Sr^{2+} allowed spontaneous AR to take place (Sr: 16±1*, Sr+Am: 71±4, Ca: 78±4, *: p<0.001). The presence of this agent during sperm co-incubation with oocytes raised AR on the ZP to normal values (Ca: 93±7, Sr: 21±13*, Sr+Am: 77±7, *: p<0.05). The addition of NH_4Cl (5 mM) increased AR (Sr: 23±6, Sr+ NH_4Cl : 58±1, Ca: 60±12, *: p<0.01). We also analyzed the participation of other Na^+ transport systems. These results suggest that, different to what happens with Ca^{2+} , the ENaC would not be inactivated when sperm are incubated in Sr^{2+} , revealing the participation of Ca^{2+} in the regulation of the sperm ion balance.

27.**CHALKBROOD CONTROL WITH *Aloysia triphylla* ESSENTIAL OIL**

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Chalkbrood is an invasive mycosis affecting exclusively the *Apis mellifera* (honeybee) larvae growth. The disease is produced by the Ascomycete fungus *Ascospaera apis*. This fungus has a worldwide distribution, in Argentina can be found in all the areas where the apiculture was developed. Pharmacological treatments have not been effective against this mycosis disease. The use of natural products such as the essential oils from plants represents a therapy without the own negative repercussions of synthetic compounds such as residual and resistance effects. The oil of *Aloysia triphylla* was obtained by hydrodistillation and analyzed by GC and GC/MS. The main components were: limonene (48.7%), geranial (11.2%) and neral (5.2%). Fungicidal activity of the oil was evaluated *in vitro* against four strains of *A. apis* isolated of two apiaries located in San Luis and two in Buenos Aires, both places were naturally infected. Isolation of the fungus was made in MY20 medium. The oil was tested by dilution method against *A. apis*. The oil concentrations assayed were at 1,000; 500 and 250 ppm ($\mu\text{L/L}$) in MY20 culture medium. Was incubated at 30°C, and measuring fungicidal activity after 14 days. Three repetitions and a control for each strain were tested. The Minimal Fungicidal Concentration (MFC) for the *A. triphylla* oil, was 500 ppm for all the tested *A. apis* strains.

28.**ELECTROPHORETIC PROFILE OF CELL WALL PROTEIN OF *Candida* spp ISOLATED FROM PATIENTS WITH ORAL CANDIDIASIS**

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Oral candidiasis is a common widespread infection in humans. It is known that components of the fungal cell wall actively participate in the adhesion of *Candida* to oral tissues previously to colonization and infection. To investigate if the set of proteins of the cell wall is specific in each *Candida* species, the electrophoretic pattern of collection strains (*C. albicans*, *C. tropicalis*, *C. glabrata*, *C. dubliniensis* and *C. krusei*) were determined and compared with those of *Candida* isolated from patients. Proteins were extracted with SDS in reducing medium or by ultrasound and the electrophoresis was carried out in 15% acrylamide gels. Gels were stained with Coomassie Brilliant Blue, and digitalized and analyzed using the Image Pro Plus 4.0 program. Although the electrophoretic profiles of collection strains showed different protein patterns between 90 and 15 kDa, a common domain from 25 to 40 kDa was observed. The electrophoretic patterns of *Candida* isolated from patients were similar to those of *C. albicans*, *C. tropicalis*, and *C. glabrata*, in agreement with the identification by conventional methods. Results suggest that protein profiles of fungal cell wall could be a complementary tool for identification of species of *Candida* isolated from patients with oral candidiasis.

29.**EFFECTIVENESS OF THE ERBIUM LASER YAG IN ENDODONCIA**

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The application of the laser in endodoncia takes implicit in the conduit treatment to let to free he himself of detritus and dentinario mud soon to come to its clogging. Was chosen laser Yag the Erbium, due to its characteristics. Extracted dental pieces were selected just that did not have verified anomalies clinical and radiográficamente. After selected they were divided in four groups, to first was made the extraction of the fillet to pulpar, washing to him and drying, to the second group was made to him the same plus the mechanical cleaning of the conduit with the use of the suitable material and to the third and fourth group equal maneuver was done to him that the previous one but was applied the laser to them with different powers previous marked, and with escoplo and hammer the root of all the pieces with a dry blow fractured. The pieces were observed in the electron microscope of sweeping. One verified that to greater power applied with laser Yag the Erbium greater amount of dentinario mud was removed. Therefore the use of greater power with the laser of Yag Erbium applied to the conduit obtains a greater effectiveness in the treatment.

31.**ENDOCRIN STIMULUS IN MESENTERIC GANGLION MODIFIES THE ANDROSTENEDIONE AND NITRITES RELEASE IN THE RAT OVARY IN OESTRUS STAGE**

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To obtain more exact knowledge of the influence of the steroid in peripheral innervation the objective of this work was to elucidate if addition of Estradiol (E_2) and its antagonist Tamoxifen (Txf) in Superior Mesenteric Ganglion (SMG) modifies androstenedione (A_2) and nitrites (soluble metabolite of nitric oxide) ovarian release in a standardized *ex-vivo* sistem, GMS-Ovarian Nervous Plexus-Ovary, from oestrus rats. Incubation was made with/without presence of E_2 (10^{-6} M) and Txf (10^{-8} M) in the ganglion compartment. A_2 release was measured by RIA at 15, 30, 60 and 120 min and nitrites were measured as well by Griess method. Krebs Ringer buffer pH 7.4 at 37°C was used like incubation medium in metabolic bath. Student test was applied with significance of $p<0.05$. E_2 increased release of A_2 at all times ($p<0.001$), while nitrites decreased ($p<0.05$) with respect to controls. Txf decreased A_2 at 15, 30 ($p<0.001$) and 60 ($p<0.05$) min and nitrites increased at 15, 30 and 60 min ($p<0.001$) with respect to controls. These results show that stimulus in peripheral nervous sistem would be involved in ovarian fisiology trough E_2 ganglionic receptors via ovarian nervous plexus.

30.**MICROSCOPIC ALTERATIONS ON ENAMEL TISSUE DUE TO CHEMICAL TRAUMATISMS SEEN THROUGH SCANNING ELECTRONIC MICROSCOPY**

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Traumatisms in teeth can have physical or chemical origin. Chemical traumas cause destruction of enamel surface in depth. Therefore, there is a greater vulnerability to biofilm action.

Method: acid engraving with gel were carried out on one hundred teeth for 15 seconds. Results were observed through SEM after an aurification process on enamel surface.

Objectives: To prove the existence of chemical traumas on enamel surface due to chemical agents.

Results: The following results were recorded: on 63% of teeth: between 10 and 13 um, on 17% between 13 and 19 um, on 15% between 19 and 25 um; on 4% between 25 and 30 um; on 1% sin alterations.

Discussion: Any chemical traumas caused on enamel creates suitable conditions for biofilm retention and makes it susceptible to possible acquired damage.

32.**ANTIBACTERIAL AND ANTIFUNGAL PROPERTIES OF ESSENTIAL OILS OF ALOYSIA TRIPHYLLA**

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A.triphylla (cedrón) is an aromatic shrub that wildly grows from the north to the center of Argentine country and is cultivated from Mexico to South América. It is popularly used as digestive, anti-inflammatory, analgesic and antipyretic. It is considered in the national and international market because of the Objectives: To study the antimicrobial activity (AA) of the EO of *A.triphylla* collected from different places.: EO were obtained by hydrodistillation methods from vegetables collected in plantations located in Río 1°, La Paz, Las Viñas, Paraguay, Mendoza and San Luis. The AA was evaluated with the disc diffusion technique and the broth microdilution method, using bacterial and yeasts strains. The Minimum Bactericidal and Fungicidal Concentration were determined (MBC/MFC)). All the EOs were more active against Gram positive bacteria, while Gram negative ones were inhibited by half of the EOs. The yeasts were inhibited by all the EOs. The oils from Cba (La Paz and Rio 1°) showed the most effective AA. The best Minimum Inhibitory Concentration (MIC) values were obtained with La Paz EO on *S.aureus* and *S.epidermidis* (MIC=28,12mg/ml), for *B.cereus*, *M.luteus*, *Hansenula sp* and *Rhodotorula sp* (MIC=7,03mg/ml). It inhibited all the Gram negative bacteria, with the exception of *P.aeruginosa*. La Paz EO was bactericidal for *S.aureus* and fungicidal for *C.albicans* while the rest of EOs was bacteriostatic and fungistatic.

33.**VALIDATION OF A LYOPHYLIZATOR DESIGNED AT LABORATORY SCALE**

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Lyophylization equipment was built and designed at laboratory scale in order to conserve small samples of foods and biological ones which are sensitive to temperature by reducing their aqueous activity. The lyophylization is a process which principle is the sublimation of ice of the frozen product. Basically the equipment consists in a hermetic camera where the frozen product is put and vacuous is made in order to reach a pressure below the one corresponding to the ice at freezing temperature. The product is maintained below the eutectic temperature and the necessary heat is supplied for the ice sublimation, (primary dehydratation) and in order to eliminate the resting water binding to the product (second phase of the process). For the equipment validation, curves of dryness were obtained (percentage of humidity versus time). Samples of fresh papaya, pig blood, fresh papaya latex, and yolk of egg and pear homogenate were utilized. The percentage of initial humidity of the samples was determined by the AACC44-15A(1993) method, and then the dryness curves were obtained. From the results we can conclude that in all the cases the dryness curves were sigmoid and the humidity reduction of the product was greater than 98%, allowing food to be conserved for a long time. The lyophylized samples were then hydrated. Their values of humidity, properties and aspect were like the fresh ones.

34.**PAPAIN ENZYME ACTIVITY**

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This study of the kinetics breaks of the peptide connection of the N-benzoil-arginine-p-nitroanilide (BAPNA) molecule catalyzed by papain. This protein belongs to the group of thiol proteases enzymes. The specific interaction with the BAPNA substrate is produced through the residue of phenylalanine. The hydrolysis of this peptide connection generates, among other reaction products, yellow p-nitro aniline. This allows following espektrophotometrically the progress of the reaction, through the speed of formation of this product. The increment of absorbency of the dissolution was measure in relation to time and the maximum absorption wavelength of the p nitro aniline, 400 nm, for different initial concentrations of BAPNA. Fresh latex from papaya was utilized as well as latex from a fig tree as a source of papain. The Biuret method was used to determine the total protein which results 3 mg/ml for fig tree latex and 10 mg/ml for papaya latex. It was possible to obtain an initial speed of reaction of 0.0024 abs/s, 0.0020 abs/s, 0.0016 abs/s, 0.0010 abs/s, and 0.0004 abs/s for a BAPNA concentration of 0.015M, 0.012M, 0.009M, 0.006M, and 0.003M respectively. The dependence of the speed reaction with the concentration of BAPNA follows the kinetics suggested by Michaelis Menten. It is concluded that, in effect, the papain contained in the latex of papaya and the fig tree catalyze the hydrolysis of BAPNA, observing more activity in the papain of the latex of papaya.

35.**MICROBIOLOGICAL QUALITY OF RAW MILK FROM DAIRY FARMS OF THE REGION VILLA MARÍA (CÓRDOBA)**

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Milk is a biological product rich in carbohydrates, fats, proteins minerals vitamins and oligoelements. Since its pH is almost neutral milk is an adequate medium for the multiplication of most bacteria. Their presence causes the alteration of milk properties and they can also act as agents responsible for diseases. The present microorganisms reflect the hygienic-sanitary conditions of the dairy rodeos. The aim of this work was to determine the presence of indicators and pathogens in samples of milk from refrigerated tanks from 26 dairy farms of the dairy region of Villa María. The counting of aerobic mesophilic, psychrotroph and viable thermophilic (FIL 100 B:1991 bacteria was performed. The counting of total coliforms (FIL 73 A:198) and positive coagulase staphylococci (FIL 145:1990), and the isolation of *Escherichia coli* (ICMSF 1983), were also performed. The orders of aerobic mesophilic and psychrotroph bacteria were $3,0 \times 10^3$ - 9×10^6 and 7×10^2 - $1,1 \times 10^7$ ufc/ml, respectively. Coliform bacteria oscillated between 3×10^2 and $3,6 \times 10^6$ ufc/ml. No thermophilic bacteria were isolated and *E.coli* was found in only one simple. The number of catalase-positive Gram positive cocci varied between 3×10^2 y $4,2 \times 10^5$ ufc/ml. In this study, hygienic quality of 70% of the samples was unsatisfactory. This fact can be attributed to an inadequate hygienic-sanitary handling of the dairy farms studied in the region.

36.**STRESS, BLOOD COAGULATION AND VITAMIN E IMMOBILIZED RATS**

Bianco M, Bensi M, Niebylski A, Scoppa G, Gauna HF.

It is well known the relationship between stress and cardiovascular risk on the other hand. Energetic homeostasis in the stress generates an increase in the production of pro- oxidants molecules. When oxidant defence is surpassed this leads to oxidative stress. The aim of this work was investigate the effects of IMO stress and vitamin E supplementation on blood coagulation. Adult male Wistar rats into three groups were divided: 1) With soybean oil (O), 2) vitamin E (V) and a third control group without supplementation (Ss). Each group was divided in controls (C) and stressed (S) rats the last group was submitted to mobilization (IMO) 2 h per day during 14 days. Blood samples were taken and hemostatic parameters were evaluated. Liver was removed and lipid peroxidation was determinated by TBARs method. Short coagulation time was observed in Ss and O rats with chronic and acute stress ($p < 0.005$) compared with their controls. No differences were observed for partially activated thromboplastine (PPAT) due to the administration of oil or vitamine. In chronic stress all stressed animals showed elevated fibrinogen values compared with controls ($p < 0.005$). MDA high values were observed in all stressed rats (oil group $p=0.0002$), (vitamin group $p=0.001$), but the increases were lesser in vitamin E group than other stressed groups. These parameters indicate an hipercoagulability associated to stress which was prevented by vitamin E supplementation. This would indicate the participation of oxidative stress in the thrombogenic effect by immobilization and consequently a major cardiovascular risk.

37.**EFFECT OF AROMATASE INHIBITORS (AI) ON VEGF AND PGE₂ LEVELS IN THE PERITONEAL FLUID (PF) IN A MURINE MODEL OF ENDOMETRIOSIS (EDT)***Bilotas M¹, Meresman G¹, Suelo C², Barañao RI¹.*¹*IBYME,* ²*CEGYR.*

It has been demonstrated that Aromatase P450 enzyme is expressed in the eutopic and ectopic endometrium of patients with EDT and that VEGF and PGE₂ levels are increased in the PF of these patients. Recently it has been proposed that VEGF and PGE₂ would be involved in a positive feedback loop that would promote the local estrogen synthesis produced by Aromatase P450 inside the endometriotic lesions (EL). The AI, Letrozole (L) and Anastrozole (A), are being evaluated as a therapeutic alternative to EDT treatment.

The aim of this work was to evaluate the effect of AI on the EL growth and the VEGF and PGE₂ levels in the PF in a murine model of EDT. After four weeks from EDT induction, mice were treated with a diary s.c. injection of A, L or saline (Control, C) during another four weeks. At the end of the treatment mice were sacrificed and the lesions were counted and measured. The PF was collected and assayed for VEGF and PGE₂ levels by ELISA and RIA respectively. The treatment did not cause EL regression but diminished EL size significantly ($p<0.05$ vs. C). As well, A and L diminished VEGF levels in the PF ($p<0.05$ vs. C). Nevertheless, only the treatment with L caused a significant decrease on PGE₂ levels in PF ($p<0.001$) whereas A did not cause any significant change. Our results suggest that AI has a beneficial effect on EL growth and supports further investigation of AI as a possible treatment for EDT.

38.**STRESS AND AGING: EFFECTS ON OXIDATIVE STRESS, PITUITARY-ADRENAL AXIS (HPA) AND BLOOD PRESSURE***Binotti S, Boccolini A, Puebla M, Bianco M, Bensi N, Gauna H, Niebylski A.**Fisiología Animal, FCEFQyN-UNRC, Río Cuarto (Cba). E-mail: aniebylski@exa.unrc.edu.ar*

Stress has a marked influence on the energetic homeostasis, free radicals (FR) production and blood pressure (BP). It is known that FR production is higher during aging, effect that could increase with stress. The aim of this work was to investigate the effects of acute immobilization (IMO) on oxidative stress, HPA axis and BP in old rats. Eight months old Wistar male rats were used, divided in control (C) and stress (S) groups. The last one was exposed to 1 hour of IMO. Corticosterone (COR), γ -glutamyl transferase (γ -GT) and plasmatic glucose were quantified. The malondialdehyde (MDA) in the liver as a cellular oxidative stress indicator was evaluated. Systolic (SBP), diastolic (DBP) and mean blood pressures (MBP) in the carotid artery by an invasive method were registered. The IMO increased the levels of COR ($p=0.000037$), γ -GT ($p=0.001$), MDA ($p=0.000001$) and plasmatic glucose ($p=0.05$). SBP and MBP increased in response to stress ($p=0.008$ y $p=0.005$ respectively), without changes in DBP. The stress-induced COR increase was lesser and BP increase was higher in old rats than young animals. The stressed rats showed highest γ -GT and MDA values than control rats. However, MDA values were higher in olds than young animals in both groups (C and S). This results show a different stress response in old than young rats.

39.**EXPRESSION OF CYTOKINES ASSOCIATED WITH YERSINIA ENTEROCOLITICA SYCH- INDUCED IMMUNOGLOBULIN A PRODUCTION***Blanco HM, Guzmán AMS, Di Genaro MS.**Immunology, FQBF, National University of San Luis. E-mail: sdigena@unsl.edu.ar*

The *Yersinia* outer protein (Yop) H is a virulence factor that requires for its translocation a chaperone SycH. We have demonstrated that *Yersinia enterocolitica* deficient in YopH (*sycH*-) is highly attenuated and induce Immunoglobulin (Ig) A production. The cytokine TGF- β induces switch to IgA class, and TNF- α decreases IgA secretion. The objective was to investigate TGF- β and TNF- α mRNA expression in *Y. enterocolitica* *sycH*- infected mice. C57BL/6 mice were orally infected with *Y. enterocolitica* O:8 wild-type (WAP) or *sycH*-. IgA levels in feces and sera were studied by ELISA, and the number of IgA- secreting spleen cells was studied by ELISPOT. TGF- β and TNF- α mRNA expression in Peyer's patches (PP) and mesenteric lymph nodes (MLN) was studied by RT-PCR. Seven and 14 days after infection, an increase of *Yersinia*-specific IgA and the number of IgA-secreting spleen cells was detected in *sycH*- infected mice ($p<0.05$). Fourteen days after infection, TGF- β mRNA expression was increased, and TNF- α mRNA decreased in PP and MLN compared with WAP infection. We concluded that IgA induction by *sycH*- strain may be associated with the expression of cytokines associated with the switch to IgA class, such as TGF- β , and with a decrease of pro-inflammatory cytokines such as TNF- α .

40.**MOLECULAR SCREENING IN THE HFE GENE IN PATIENTS WITH ALTERATIONS IN THE IRON METABOLISM AND IN THEIR RELATED FAMILY***Blariza MJ¹, Bonafé M¹, Montes C², Rossi N², Moreno Barral J¹, Soria N¹.**¹Cátedra de B y BM. FCM. UNC. ²Laboratorio de Citogenética Médica. Hospital de Niños. Córdoba. E-mail: maijo013@hotmail.com*

Hereditary hemochromatosis (HH) is characterized by an increment in the intestinal absorption of iron. This genetic disorder is usually inherited in an autosomal recessive form with an estimated prevalence in Caucasians of 1 case per 400 individuals. Two mutations in the HFE gene (C282Y and H63D), are the most frequent in HH. The aim of this study was to detect the C282Y and H63D mutations in the HFE gene in our population.

We studied 30 patients (17 males and 13 females) with iron overload, and/or alcoholic liver cirrhosis with iron overload and/or patients family. In all cases, consultations with Medical Genetics Service were done. DNA was extracted from peripheral blood and PCR-AE and PCR-RFLP were used for the C282Y and H63D screening.

We did not find these mutations in the patients with alcoholic liver cirrhosis and iron overload. We found mutations in 24 patients (80%). From positive patients 51,6% carried C282Y mutation (38,7 % in heterozygous form; 12,9 % in homozygous form). The H63D mutation was found in heterozygous state (28,4%). The results are according with the prevalence of C282Y mutation as the most frequent in Caucasians.

41.**CHRONIC SOCIAL STRESS: NEUROENDOCRINE RESPONSE, ENERGETIC BALANCE AND RENAL EXCRETION**

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The exposition to chronic social stress (CSS) seems to be related with many systemic and mental disorders. The objective of this work was investigate the neuroendocrine changes, Na^+ renal handling and energetic balance in rats submitted to CSS. Male Wistar rats were submitted to six consecutive sessions of CSS, using the resident-intruder paradigm. Plasmatic corticosterone (CORT) and creatinine (CREAT) and urine levels of epinephrine (E), norepinephrine (NE), dopamine (DOPA), Na^+ , K^+ and CREAT were measured. Daily food intake (FI) and body weight gain (BWG) were evaluated. The feed efficiency (FE) and CREAT clearance (ClCREAT) were calculated. CORT ($p<0.01$), NE ($p<0.01$) and DOPA ($p<0.05$) values increased in stressed animals. Antinatriuresis ($p<0.01$) and an increased K^+/Na^+ ratio ($p<0.01$) also were observed in this group. No differences were found in ClCREAT. Besides stressed animals showed larger BWG ($p<0.05$), FE ($p<0.01$) and FI ($p<0.05$). Hypothalamic-pituitary-adrenal and sympathoadrenal axis activations could lead to the lower Na^+ excretion founded in stressed animals. The energetic balance and Na^+ renal changes could be related to the greater incidence of the obesity, diabetes and cardiovascular diseases in human populations submitted to a non-traumatic stress.

43.**THE ACTION OF CHROMIUM (CR VI) ON ERYTHROCYTE RHEOLOGY**

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Once absorbed, Chromium (Cr) is immediately taken up by red blood cells (RBCs), with an effect on the rheological properties of these cells still unknown. The *in vivo* effect of Cr on shape and aggregability of human erythrocytes was studied. RBSc (obtained from healthy donors with previous stated consentment) were incubated in Cr solutions at 50 and 100 mg/l (n=16). Both RBCs properties were analyzed: shape (through microscopy) and aggregability in dextran 500 solution (by optical method determining s_0/n_0 [the final shape of the aggregates] and $2k_2n_0$ [initial rate of the process]). Statistical analysis: "t" Student (paired groups) *a*: $p<0.0001$.

	shape	s_0/n_0	$2k_2n_0$
Control	Discocytes	$1,89 \pm 0,003$	$1,60 \pm 0,05$
Cr 50 mg/l	Stomatocytes II	$1,86 \pm 0,006^a$	$1,22 \pm 0,07^a$
Cr100mg/l	Stomatocytes II	$1,86 \pm 0,007^a$	$1,15 \pm 0,07^a$

Cr increases the negative curvature of the membrane, leading to a loss of aggregability, an main rheological factor in microcirculation. This diminution contributes to the release of O_2 to the tissues with the subsequent synthesis of free radicals, which constitutes a probable cause of Cr's carcinogenic effect.

42.**POSITIONAL EFFECT ON COMPETITION BETWEEN INDIGENOUS VS. INOCULATED BRADYRHIZOBIA FOR PEANUT NODULATION**

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Selected *Bradyrhizobium* sp. strains inoculated on peanut seeds often fail to occupy a significant proportion of nodules when a competitor rhizobial population is already established in soil. Such failure may result either from a genetic or physiological advantage of the adapted soil population over the introduced inoculant, or from a positional advantage, *i.e.*, the soil population occupies the soil profile where the roots will penetrate, whereas the inoculant remains concentrated around the seeds. We studied competition among native rhizobia and a *Bradyrhizobium* sp. strain inoculant. The seed-inoculated strain formed minor amount of nodules on peanuts with respect to bradyrhizobia inoculated in-furrow, indicating the positional advantage of soil populations or the bacteria inoculated in-furrow for nodulation. We subsequently assessed the contribution of this positional effect using a laboratory model in which a rhizobial population is stabilized in sterile vermiculite. Our results demonstrate the importance of bradyrhizobial distribution, and/or movement into soil, for strain competitiveness and nodulation in soil-grown peanuts.

44.**GENOMIC REGIONS RESPONSIBLE FOR RESISTANCE TO MAL DE RÍO CUARTO DISEASE**

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Microsatellites (SSR) markers have been used extensively to map quantitative traits loci (QTL). A discrete discriminant analysis (DA) was carried out with a maize recombinant inbred lines (RIL) population to identify the chromosomal regions responsible for resistance to Mal de Río Cuarto (MRC) disease. Recently, MRC has increased in importance being the more serious viral disease of maize in Argentina. The RIL population, consisting of 144 lines, was developed from a cross between a susceptible inbred line, M017, and a partially resistant inbred line, BLS14. The population was genotyped with 40 SSR markers, and was also evaluated for traits related to MRC disease at two environments in the endemic disease area. For each trait evaluated, the RIL were assigned to two extreme phenotypic groups. Markers DA-selected produced high levels of correct classification, indicating an association between SSR and phenotypic traits, which allows to deduce genomic regions responsible for MRC resistance. Across environments, the number of SSR DA-selected per trait ranged from one to ten. Several SSR associated with resistance to MRC were clustered in a few chromosomes. Results indicate that these primers could be promising in molecular marker mapping for MRC resistance and consequently, marker-assisted selection.

45.

CHARACTERIZATION OF APOPTOSIS IN THE ADRENAL CORTEX OF RATS SUBJECTED TO GESTATIONAL CHRONIC STRESS

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Previous results of this laboratory indicate that immobilization stress during pregnancy produces an increase of apoptosis in the maternal adrenal cortex, related to the rise of corticosterone plasmatic levels. We aimed at determining the signalling route involved in apoptosis, and at demonstrating the structural change occurred in this process by means of electron microscopy (EM). Wistar rats (250-300g) kept at usual laboratory conditions were employed. Pregnant rats were separated into control (C) and stressed (S) groups, and sacrificed by decapitation at 21 days of gestation. Adrenal glands were processed using the conventional histological techniques for immunocytochemistry and EM. Histological sections were marked with anti-Bcl₂ and anti-DR₄ antibodies, revealed with DAB and contrasted with haematoxylin. The results were analyzed semiquantitatively. A remarkable reactivity to DR₄ was observed in the adrenal cortex, with a more extended distribution in S. Bcl₂ marking was very scarce in both groups, with no differences between them. These results show that the apoptosis observed in the adrenal cortex of pregnant rats is produced by activation of the extrinsic route.

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46.

CORTISOL-INDUCED CHANGES IN THE APOPTOSIS (AP) OF PERIPHERAL BLOOD MONONUCLEAR CELLS (PBMC) FROM TUBERCULOSIS PATIENTS AND THEIR ASSOCIATION WITH THE *IN VITRO* IFN-γ PRODUCTION

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Cell-mediated immune responses (CMI) along with TNF-α and IFN-γ production are required for protection in tuberculosis (TB). In studies on immunoendocrine interactions, cortisol (Gc) 10⁻⁶M was shown to inhibit *in vitro* CMI to sonicate *M. tuberculosis* (Tso, 8 µg/ml), this effect not being reversed by dehydroepiandrosterone -DHEA- (10⁻⁷-10⁻⁹M). We have now investigated the effect of Gc and DHEA on the AP (flow cytometry, Annexin V/IP 24h- and 4-day hypodiploidy) of PBMC (10⁶ cells/ml) from TB patients and its relation with IFN-γ production *in vitro* (ELISA, pg/ml). Results at 24 h (% AP, median-rank, similar to 4-day data) were as follows: unstimulated cultures, TB 25(22-37) n:17; Controls -Co-19(17-24) n:15 p<0.01]; Tso-stimulated cultures, TB 28.5(23-35); Co 19(17-27) p<0.01]; Gc-induced AP, was more evident in Co, Tso 19(17-27), Tso+Gc 31(26-36), p<0.01. **IFN-γ Tso-stimulated cultures**, TB 1335(616-2282); Co 646(413-905) p<0.01; Tso stimulation+Gc, TB 351(92-1017); Co 225(91-358), p=0.05; IFN-γ levels in Tso+Gc cultures from TB patients correlated with % AP (r:-0.62, n=13, p<0.05). DHEA failed to reverse Gc effects. While Gc increased *in vitro* AP from PBMC, only in TB was this effect associated to a decreased IFN-γ production.

47.

EVALUATION OF EFFECTS OF NITROGEN, PHOSPHORUS AND POTASSIUM ON the SPECIFIC GRAVITY IN POTATO (*Solanum tuberosum* L. Spunta) OF TAFÍ del VALLE, TUCUMÁN

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The potato culture requires amounts significant of nutrients to obtain higher yield compatible with a rational handling to obtain tubercles with industrial quality, without causing negative impacts in environment. The effect of nitrogen (N), phosphorus (P) was evaluated and potassium (K) on the specific gravity in Potato cv. Spunta, produced in Tafí del Valle, to 2200n.s.n.m in Tucumán, during 2006/07. The experimental design was Randomized Blocks with 7 treatments of fertilization with NPK in units of fertilización.ha⁻¹ and 5 repetitions: 1. Control; 2. 100N; 3. 100N: 90P: 150K; 4. 100N: 150P: 150K; 5. 30N: 90P: 150K; 6. 130N: 150P: 150P; 7. 130N: 160P. After harvested, the tubercles were weighed in air and in water and its specific gravity by each treatment were determined. ANVA and Test of Tukey (*p*=0.05) were realized. Significant differences of specific gravity in 1 (36.0) with respect to 2 (13.5) and 4 were detected (12.947); there were intermediate values in 6 (29.057), 7 (20.027), 5 (19.443) and 3 (16.507). Treatments 1; 6; 7 and 5 surpassed the threshold of acceptance of 18% established by the industry. This is inferred that the culture responds in form differentiated to the fertilization with NPK, influences qualitative aspects required by the industry, in as much, its adjustment reduces negative environmental effects.

48.

EFFECT OF TIME VARIATION IN WATERBIRDS COUNTING

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It is very well-known that many factors affect the activity of the birds. The study of the factors that slant the birds' counts should be evaluated a priori in all monitoring programmes. Little information exists in bibliography about the effect that the hours of the day have on the counts of aquatic birds. In this work how the time of day affects the total number of species and abundance of water birds was examined. Counts of waterbirds were conducted in two lagoons (La Olla and Rivero) situated to the south of the Province of Córdoba, Argentina. The counts began half an hour before the dawn and they concluded half an hour after the evening. The counts of the number of individuals of each present species were made every 30 minutes. No significant differences were found in the values of total number of species for the two lagoons among the morning, midday and late counts (Kruskal-Wallis Test). The abundance values didn't register statistically significant evidences in Lag. La Olla, but they did in Lag. Rivero; this is due to this lagoon is sleeping place for some species of birds that group in big numbers in hours of the afternoon. This work suggests that the counts of aquatic birds are comparable among places independently of the hour of the day in which they are carried out, having always present different behavioral pattern of each species under study.

49.**RENAL ELIMINATION OF P-AMINOHIPPURATE (PAH) IN RATS WITH DIFFERENT STAGES OF EXTRAHEPATIC CHOLESTASIS***Brandoni A, Torres AM.**Área Farmacología. Fac. Cs. Bioq. y Farm. U.N.Rosario. CONICET. E-mail: abrandon@fbioy.unr.edu.ar*

Alterations in renal elimination of organic anions were described in different models of cholestasis in rats. The aim of this work was to study the influence of the duration of extrahepatic cholestasis in PAH renal elimination in male adult Wistar rats with bile duct obstruction for 21 h (L_{21} , n=4) and 72 h (L_{72} , n=5). A Sham group (S, n=8) was also processed. Excreted (E.L.) and secreted (S.L.) loads ($\mu\text{g}/\text{min}/100\text{g b.w.}$) of PAH were determined by clearance techniques. Abundances (%) of OAT1 and OAT3 (organic anion transporters involved in PAH renal secretion) were evaluated in basolateral membranes by Western Blotting. Results (ANOVA-Newman Keuls test, $p<0.05$: a vs. S; b vs. L_{21} ; c vs. L_{72}). E.L.: S= 205±7; L_{21} = 219±8 c; L_{72} = 175±10 a,b. S.L.: S= 164±6; L_{21} = 185±7 a,c; L_{72} = 140±6 a,b. OAT1: S= 100±6; L_{21} = 139±11 a,c; L_{72} = 71±9 a,b. OAT3: S= 100±4; L_{21} = 92±17 c; L_{72} = 198±14 a,b. In the early phase of extrahepatic cholestasis, the observed up-regulation of OAT1 expression would explain in part the increase in PAH renal secretion, in order to compensate the failure in hepatic elimination of organic anions. A decreased in PAH renal secretion was detected in rats with extrahepatic cholestasis of 72 h. This fact would be explained by the observed decrease in OAT1 protein expression; the increase in OAT3 abundance did not compensate the lower renal secretion of PAH in these rats. The present study puts in evidence the critical role of OAT1 in PAH renal elimination in rats with extrahepatic cholestasis.

50.**SEmen BACTERIOLOGICAL QUALITY IN *Apis mellifera****Brazzola M¹, Catena M¹, Capel AM¹, Palacio A², Collins A³, Andere C¹. ¹FCVet. UNCPBA. ²Unidad Integrada INTA FCA. UNMdP. ³Bee Research Laboratory, USDA. E-mail: amcapel@arnet.com.ar*

Apis mellifera shows great genetic variability as a result of its capability to adapt to different environments. Variability preservation is the goal of the National Genetic Program (MeGA), which employs a closed breeding system to preserve the materials achieved through instrumental insemination. The objective of this research was to evaluate semen bacteriological quality due to the impact in the Program. Samples from ten colonies were evaluated. Enriched culture media were done under different atmospheres and the colony forming units were registered. Identification was done through biochemical routine tests. There were isolated *Acinetobacter calcoaceticus*, *Staphylococcus epidermidis*, *S. hominis*, *S. cohnii* subsp. II, *S. spp.*, *Bacillus cereus*, *B. coagulans*, *B. subtilis*, *B. spp.* *B. micoides*, *B. coagularis*, *Flavimona oryzihabitans*, *Listeria spp.*, *Arcanobacterium haemolyticum*, *Corynebacterium spp.*, *Streptococcus mutans*, *Alcaligenes faecalis*, *Acidovorax delafieldii*, *Aerococcus viridans*, *Bordetella pertussis*. Although the majority of these microorganisms has been reported in other *Apis mellifera* organs, there is not enough reference in the semen of this specie.

51.**INFLUENCE OF ESTRADIOL AND TAMOXIFEN IN SUPERIOR MESENTERIC GANGLION IN 3 β - AND 20 α - HIDROXISTEROID DEHYDROGENASE ENZYMES ACTIVITY IN RAT OVARY***Bronzi D, Vega OA, Capone C, Martinez C, Escudero C, Sosa Z, Rastrilla AM.**Lab. Biol. de la Reprod. (LABIR). FQB y F. UNSL. San Luis, Argentina. E-mail: zsosa@unsl.edu.ar*

Part of the ovarian innervation is constituted by the ovarian nervous plexus (ONP) which fibers are originated at the superior mesenteric ganglion (SMG). The objective of this work was to investigate if the presence of one steroid and its antagonist in SMG, in a standardized *ex-vivo* system, modify through ONP progesterone (P) ovarian release and the activity of 3 β - and 20 α - hidroxisteroid dehydrogenase (3 β - HSD, 20 α - HSD) at the oestrous stage in rat. These systems are then incubated with/without E₂ (10⁻⁶ M) and Txf (10⁻⁸ M) presence in the ganglion compartment. In all cases a specially designed compartment is used with Krebs Ringer solution, pH 7.4 in a 37°C metabolic bath. Student test was applied with significance of $p<0.05$. These results show that when E₂ was added in the ganglion compartment stimulated P release at 15, 30 and 60 min ($p<0.001$). Txf was lower than the control group at 15 min ($p<0.05$) and 60 min ($p<0.001$). Besides E₂ increased 3 β - HSD activity ($p<0.05$) but did not make changes in 20 α - HSD activity and Txf presence did not make changes. Physiologically these results show that presence of ganglionic receptors for E₂ and evidence physiological relevance of peripheral nervous system neurones, like the SMG ones, in control of ovarian functions.

52.**LACK OF KNOWLEDGE ABOUT ESTIMULANT BEVERAGES PURE OR MIXED***Busmail LL, Cesolari JAM, Calvi BJ, Martínez C, Taborda M. Facultad de Cs. Médicas (UNR), IUNIR, UNSAM. E-mail: lbusmail@argentina.com*

Stimulant beverages are non alcoholic, and are considered as functional food since they induce vitality in physical and mental strain. This type of beverages is not recommended to be used combined with alcoholic ones, but to supply nutrients. Objectives: to demonstrate that young people consume them without knowing their components and the actual effect of them when added to alcoholic beverages. Material and Methods. A group of university students (b=234; 150 females, 81 males) were studied, applying a questionnaire collecting the following data on these beverages: 1. information about their components and effects, 2. information regarding the recommendation of not mixing them with alcoholic beverages. Results: 101 out of the total, both sexes, 80.5% consume them mixed with alcohol and 19.5% alone, without knowing the actual effects and their components. They know that they raise the mood and increase intellectual performance, ignoring voluntarily the effects when combined with alcohol. The average consumption is 2-4 cans daily according to marks. Combined with alcohol they consume a can per drink, and since they could take more than 3 drinks per night considering that this practice is frequently in Friday and Saturday, the consumption is 6 cans or more in 24 h. Each can with a liquid content of 250 cm³ and 35 mg /100ml of caffeine provides 87.5 mg. per drink. Conclusions: These substances were created to drink without any addition, in optimal quantities, to obtain a desired effect. Population Lack of knowledge states that mix of these substances or their consumption excess produce severe clinical events, such as dehydration, confusion, muscle weakness, lung congestion, and even cardiac arrest.

53.**INVOLVEMENT OF EPITHELIAL CADHERIN (ECad) IN BOVINE GAMETE INTERACTION**

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Introduction: Ecad is a homotypical calcium-dependent adhesion molecule. Its presence has been previously described in bovine spermatozoa and cumulus-oocyte complexes (COCs) in regions involved in gamete interaction. **Objective:** To evaluate the participation of Ecad in sperm-COC interaction during fertilization. **Methodology:** The ability a specific anti Ecad antibody (DECMA-1, Sigma, directed against adhesion domains) to block gamete interaction was evaluated in (1) *in vitro* fertilization (IVF) assays using COCs or denuded oocytes and (2) zona pellucida (ZP)-spermatozoa binding assays; the percentage of fertilized oocytes (oocyte penetrated by a spermatozoon) and the number of spermatozoa bound to the ZP were evaluated, respectively. **Results:** Anti Ecad inhibited IVF of COCs ($69 \pm 10\%$ vs $90 \pm 25\%$; mean \pm SDM, anti Ecad vs IgG control, $p < 0.05$) and oocytes ($57 \pm 15\%$ vs $89 \pm 10\%$; $p < 0.05$). Part of the inhibitory effect was caused by a blockage in sperm-ZP interaction (17 ± 7 vs 34 ± 11 sperm/ZP; $p < 0.0001$). **Conclusions:** The results suggest the participation of Ecad in the molecular mechanisms of gamete interaction in the bovine model.

54.**LOCATION, ISOLATION AND CHARACTERIZATION OF THIOL PROTEINS DURING SPERM CELL MATURATION**

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Sperm thiol oxidation is involved in sperm motility, capacitation, acrosome reaction and fertilization. Changes in thiol oxidation, to disulfide bonds, in sperm cell proteins are verified during epididymal trip. Using different methodology we attempt to characterize the thiol reach protein. First, by sonication and ultracentrifugation in sucrose gradient, we fractionated and isolated spermatozoo head and tails. Then compared mature vs. immature sperm cell to establish which specific proteins support SH-SS changes. Second different protocol to solubilize cell proteins were used. Some samples were incubated with detergents (0.1% Triton X100, DTT 32 mM and SDS 4%) and soluble vs non-soluble proteins were analyzed. All samples were incubated with monobromobimane (mbb) that allow to stain thiol proteins. Other method was used to localize in sperm cell or in the fraction above mention using the mbb but observed under fluorescence microscopy and were also studied by electron microscopy to know the subcellular origin of the proteins. The thiol proteins were separated by SDS-PAGE and determinated the molecular weight. Proteins about 27 kDa display a great fluorescence as well as the principal piece of the sperm. Complementary at TEM this fraction show elements corresponding to sperm tail. Additional effort will be down to further characterize an isolated one by one of the mbb-SH-SS-proteins.

55.**STUDY OF THE DISTRIBUTION OF THE OXTRAOSEUS IRRIGATION OF THE HALLUX (PART 2)**

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Analyze and describe the different anatomical patterns of the vessels that nourish the Hallux, allows to category the more fragile and exposed to vascular damage zones.

It has been studied 14 cadaverical feet of adults. These were canalized and injected with Latex RV620 **tined** with red, using catheter N° 35, in the tibial anterior, posterior and peronea artery, using manual pressure. Later, they were kept in cold for 24 hs to solidify the latex and they were conserved by the MAR II technique (Museo Anatomía de Rosario).

The vitality of the Hallux depends on an arterial system whose elements are: Metatarsal Dorsal Artery, Metatarsal Plantar Artery, and the superficial branch of the Metatarsal Plantar Artery (branch of the Tibial Posterior Artery).

It is of extreme importance the knowledge of the vascular patterns in order to avoid damages in reconstructive surgeries. Chevron and Trehowian osteotomy, plantar **colgajos** and other pathologies with avascular necrosis of the first metatarsal. Completing the previous studies and enlarging the casuistic, it was conclude that minor collaterals were found, but with similar patterns formerly established. From the previously exposed, we reconfirmed that respecting the different anatomical patterns of irrigation it should be **reparado** in the lateral, medial and plantar faces of the Hallux to diminish the incidence of aseptic necrosis of the metatarsal head.

56.**REGULATION OF ATPases BY INTERACTION WITH ACETYLATED TUBULIN**

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(Na⁺,K⁺)-, H⁺- and Ca²⁺- ATPases are integral membrane proteins that play important roles in the exchange of ions and nutrients between exterior and interior of the cell. Involvement of these ATP-hydrolysing enzymes in signal transduction pathways has increased the interest for their study. The activity of these ATPases can be regulated by several specific effectors. This work summarizes the information regarding the regulation of the mentioned ATPases by a common effector, acetylated tubulin. This tubulin isotype interacts with these ATPases, and inhibits their enzymatic activities. The presence of an acetyl group on lysine 40 of α -tubulin is a requirement for the interaction. The stimulation of the ATPases activity by the specific effectors involves the dissociation of the tubulin/ATPase complexes. In cultured cells, acetylated tubulin that is associated with ATPases is apparently constituent of microtubules. Stabilization of microtubules by taxol is an impediment for association/dissociation of the complexes. Membrane ATPases could function as anchorage sites for microtubules.

57.**GENERATION AND CHARACTERIZATION OF A TRANSGENIC MOUSE TISSUE SPECIFIC FOR TRPC6***Canales C^{1,2}, Krall P^{1,2}, Walz K¹.**¹Centro de Estudios Científicos, CECS, Valdivia, Chile.**²Universidad Austral de Chile. E-mail: cesarcanales@cecs.cl*

The TRPC6 channel (Transient receptor potential channel 6) belongs to the TRPC family and it is a protein with six transmembrane domains, activated by Di-acilglicerol (DAG) that is related with the calcium transport towards the cytoplasm. This channel is present in different organs and tissues in human as well as in mouse, displaying an homology of 87% and 93%, at cDNA and aminoacids level, respectively. Recently, mutations in *Trpc6* have been described in patients with Focal and Segmental Glomerulosclerosis (FSGS), a pathology with dominant autosomic inheritance. The goal of this work was the generation of a transgenic mouse that over express the wild type form of *Trpc6* in the podocytes and its later phenotypic characterization. We have generated a construct with the full length *Trpc6* cDNA with the addition of a hemagglutinin (HA) tag. This constructed was cloned into an expression vector, downstream of the human podocin promoter (pNPHS2). After corroboration of *in vitro* expression, molecular weight and subcellular localization of the protein, the construct was microinjected to generate transgenic mice. Three of the resulting transgenic lines were systematically analyzed to evaluate copy number integration of the transgene, expression of the transgene and phenotypic presentation. Preliminary characterization shown increased of proteinuria and abnormal morphology of the kidneys in agreement with a renal pathology derived by the over expression of *Trpc6* in podocytes.

58.**DETERMINATION OF MITOTIC INDEX IN MERISTEMS OF *Allium cepa****Cangiano MA, Correche ER, Enriz RD.**Facultad de Química, Bioquímica y Farmacia. Universidad Nacional de San Luis. E-mail: ercor@unsl.edu.ar*

The time and distribution of different status of cellular population might be evaluated from the cellular cycle. Cells follow a cyclic rhythm for the division. Thus, it is possible to calculate: the percentage of dividing cells, the division phase, as well as the division time of each phase. The mitotic index (MI) is a parameter indicating the frequency of cell division and the velocity of the meristems grow.

The aim of this work is to determine the hour of the day in which the MI has the highest value. Thus, it is possible to give further information on the cytology of *Allium cepa*, eliminating random factors and using non subjective criteria to obtain the samples. The present study was carried out on root meristem of *Allium cepa*. Root samples were fixed, stained and squashed, following the routine techniques. The samples were obtained using 3 h intervals during a final period of 24 h. An average of 4500 meristematic cells was analyzed and then the MI of the population was evaluated. The analysis of these results indicated that the highest MI was obtained at 10:00 am. (MI=8.89%) and the lowest value was obtained at 18:00 pm (MI= 3.64%).

This study allows establish that the MI is a significant data giving the potential proliferation of cells and provide an excellent guide to obtain the samples in those hours during the day with better probabilities for the cell division.

59.**PHOSPHATIDIC ACID PRODUCES CELL EXPANSION IN POTATO STOLONS***Cantoro R, Cenzano AM, Abdala G.**Dpto. de Ciencias Naturales. Facultad de Ciencias Exactas, Físico-Químicas y Naturales. UNRC. Río Cuarto. Córdoba. E-mail: renacantoro@hotmail.com*

Jasmonic acid (JA) and its derivates stimulate potato tuber formation through an increase in central mother cells area of stolon's apical meristem. JA effect on tuberization could be mediated by phospholipid changes and second messengers production. Cenzano *et al.* (2007) demonstrated that exogenous JA produces phosphatidic acid (PA) accumulation in the second stage of tuberization. In this work we propose to evaluate PA effect on cell expansion of first and second stage of stolon to tuber transition. For this purpose, two analysis zones located in the apical meristem were defined. Liposomes containing 5 and 10 µM PA were made for tissue stimulation. Stolons were prepared for microscopic observation and cell area measurement. A statistically significant increase of cell area was caused by 5 µM PA in Zones I and II of first stage and 10 µM PA treatment produced the same effect in Zone I of second stage. These results allow us to conclude that PA may act as a lipid messenger during cell expansion process conduced to tuber formation. Moreover, the increase in cell area of apical and sub-apical zones of first stage stolons suggests that the signal is "triggered" in this stage allowing the posterior tuber development.

60.**SEmen EVALUATION OF *Apis mellifera* IN THE IMPROVEMENT GENETIC PROGRAM***Capel AM¹, Palacio MA², Catena M¹, Martínez A², Collins A³, Rodríguez EM¹, Andere C¹.**¹FCV-UNCPBA. ²UI INTA-FCA. UNMdP. ³Bee Research Laboratory, USDA. E-mail: amcapel@arnet.com.ar*

In our country, genetic diversity showed by *Apis mellifera* is employed for the selection work that is at present being performed by the Improvement Genetic Program (MeGA) of the Apicultural National Project (PROAPI). The instrumental queen insemination with quality semen in a closed breeding program insures improved genetic materials. As there are no references concerning semen quality evaluation previous to its employment in insemination, this set a starting point for the present work: to estimate semen quality to be used in the Program. There were evaluated drones from 14 colonies, collected in individual boxes. Kiev diluents were used, motility was described by visual observation, sperm concentration was measured by Newbauer counting chamber and viability was observed using a fluorescent staining. Motility was similar in all the colonies. Sperm concentration (million/µl) was 1.11×10^6 to 9.27×10^6 . Sperm viability (% live) by colony varied from 72 to 98%. The analyzed results were similar to the ones found by other authors and appropriate to obtain a successful insemination. Other researches are being done to evaluate different preservation methodologies.

61.**PRENATAL TREATMENT WITH ACE INHIBITORS AFFECTS AT₁ RECEPTOR EXPRESSION DURING DEVELOPMENT IN RAT LUNG**

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Components of the renin-angiotensin system (RAS), including angiotensin converting enzyme (ACE) and Angiotensin (Ang) II receptors are expressed in lung endothelial and epithelial cells. The aim of the present work was to investigate prenatal ACE inhibition influences in expression of Ang II receptors (AT₁ and AT₂) in postnatal lung tissue development. Mini-osmotic pumps with enalapril, captopril or saline solution, were implanted in pregnant Wistar rats during the last week of pregnancy. The expression of both Ang II receptors (AT₁ and AT₂) was analysed by RT-PCR. Pup's lungs at four different ages: PND0, PND8, PND15 and PND30 were evaluated. There was not expression of AT₂ receptor. However, we found expression of AT₁ receptor, incrementing during the ages of development, with a maximum value at PND15 (ANOVA, P<0.01). Semiquantitative assessment indicated significant differences (ANOVA, P<0.05) at PND0 and PND8 with pharmacology treatment, respect to controls. Inmuno-fluorescence analyses proved localization of AT₁ receptor in vascular and alveolar tissues. We conclude that AT₁ receptor expression in rat lung is affected by treatment with ACE inhibitors, interfering with functional integrity of the RAS.

62.**PULEGONE, MENTHONE AND LIMONENE ISOLATED OF ESSENTIAL OIL OF *MINTHOSTACHYS VERTICILLATA* EFFECTS ON HUMAN CELLS**

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The PHA and ConA mitogens are able to stimulate to ($\alpha/\beta\gamma\delta$) CD4⁺ and CD8⁺ T cells leading to its polyclonal activation. Epling has demonstrated anti-allergic and lymphoproliferative properties *in vitro* of essential oil (EO) of *Minthostachys verticillata* (Griseb.). Objective: The *in vitro* effects of pure compounds isolated from the EO of *M verticillata*: pulegona (P), menthone (M) and limonene (L), like polyclonals activators on human lymphocytes, were investigated. **Material and Methods:** The cells, isolated from peripheral blood, were cultured in independent assay and stimulated with PHA, ConA, P, M or L. In addition co-stimulated cultures with: P/M, P/L, M/L and P/M/L in percentage similar to that are these compounds in the vegetable (63% of P, 16% of M and 2% of L) were realized. The cultures were making by MTT colorimetric assay. **Results and Discussion:** The combination of 3 pure compounds was effective to clonal expansion of lymphocytes like PHA o ConA (p=ns). These results revealed their mitogenic capacity such being able to be used *in vitro* to evaluate the functionality of the cellular immunity.

63.**GENERATION AND CHARACTERIZATION OF RAI1 POINT MUTATIONS ASSOCIATED TO SMITH-MAGENIS SYNDROME**

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Smith-Magenis Syndrome (SMS) has an approximated incidence of 1/25,000 births and its clinical presentation includes craniofacial abnormalities, obesity, behavioral and sleep anomalies and mental delay. This syndrome is associated to a deletion of a genomic interval in the chromosome 17p11.2 that includes 23 genes. Recently it was demonstrated that mutations in the Retinoic Acid Induced 1 gene (*RAI1*), one of the 23 deleted genes, are associate with most of the pathological characteristics before mentioned, indicating that *RAI1* is the responsible gene for great part of SMS clinical manifestations. Although the exact function of the protein is still unknown, *RAI1* could comprise the transcription machinery and would be involved in nervous system development. In order to determine the molecular mechanisms affected in *RAI1* mutants human *RAI1* cDNA was obtained, which was analyzed *in vitro* for the first time. We generated two point mutations described in patients with SMS, using site-directed mutagenesis, in human and murine *RAI1* cDNA. Their subcellular location and its molecular weight were verified, in addition, the transcriptional activity of wild and mutant proteins was measured, not finding differences between the wild type and the mutant forms of the murine protein.

64.**EFFECTS OF LIPID PROFILE AND SERUM FIBRINOGEN OVER THE BLOOD VISCOSITY IN HEALTHY MENOPAUSAL WOMEN**

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The increase of blood viscosity (η_s) could accelerate the atherothrombosis due to fall of blood flow and the extension time of residence of atherogenic particles near the endothelium. The objective of this work was to study the effect of lipid profile and serum fibrinogen (F) over η_s in healthy menopausal women with normal hematocrit, globulin and albumin. The η_s was measured with rotacional viscosimeter cone - plate, the F by gravimetric method and the serum lipids (HDL-C, triglycerides (TCG), LDL-C, total cholesterol (C) and no HDL cholesterol) by colorimetric enzymatic method and the ratios (LDL-C/HDL-C, total cholesterol/HDL-C) in 40 patients previous written consent of them. We observed a significant positive correlation between F and η_s ($r=0,47$ and $p=0,0028$), LDL-C and η_s ($r=0,40$ and $p=0,012$), LDL-C/HDL-C and η_s ($r=0,52$ and $p=0,006$), cholesterol no HDL and η_s ($r=0,41$ and $p=0,0078$), cholesterol total/HDL-C and η_s ($r=0,47$ and $p=0,0022$). Positive correlation but no significant between C and η_s ($r=0,28$ and $p=0,07$), TCG and η_s ($r=0,02$ and $p=0,89$). Negative and significant correlation between HDL-C and η_s ($r=-0,33$ and $p=0,03$). The serum lipids and the relative proportions as well as serum fibrinogen influence in part over η_s . Nowadays, fibrinogen and serum lipids are considered cardiovascular risk biomarkers.

65.**INFILTRATION RATES IN FEEDLOT SOILS**

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The feedlot is a system used for finishing cattle that concentrates a large number of animals in a small area. In this system the pollution of soils and ground and underground water, as a consequence of manure accumulation and effluents movement, constitutes the most important environmental risk. Therefore, our aim was to characterize the infiltration rate of two physiographic points (high and lower) settled in the yards of the feedlot after four months of rest and to compare it with a control. The external and internal characterization of the soils and the infiltration rate was measured by a double ring infiltrometer. The soils presented an upper horizon (Ap) with a laminate structure of considerable thickness originated by trample, and a compacted A2 horizon (massive with high apparent density). This study was able to show that the infiltration rate is significantly higher in the control than in the feedlot yards. The infiltration curves presented potential adjustment of the type $I=K \cdot t^m$, in which r^2 values are higher than 0.73. The m value is significantly high between control and feedlot yards. The control presents m values over 0.6, which shows the stability of its soil structure. The K value is significant for the low yards of the feedlot. We conclude that there are not significant differences between the infiltration rates among the physiographic positions settled in the feedlot yards, but there is with regard to control. The practical consequence of these results is that the infiltration in the feedlot yards is very low so, perhaps, *in situ* contamination is not a consequence of lixiviation processes.

66.**OVIDUCTAL CALCIUM DYNAMICS DURING THE REPRODUCTIVE CYCLE OF *Pomacea canaliculata* (Gastropoda, Prosobranchia)**

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In *Pomacea canaliculata*, aerial egg deposition is associated with the presence of a calcareous shell that protects embryo from desiccation. The main calcium deposit involved in the egg-shell formation is the glandular complex of the pallial oviduct. The aim of this work is to analyze the variations in contents and distribution of oviductal calcium during the annual reproductive cycle. Samples were conventionally processed for transmission electronic microscopy.

During the reproductive period (September-March), an intense tissue calcification, calciferites which reach their maximal volume, and calcific particles between cell folds can be seen. Associated with these observations, there is an evident calcium mobilization towards the ductal lumen and calcified cell extrusion. In the post-reproductive period (April), scarce remnant calciferites and empty ductal lumens can be observed. In the pre-reproductive period (May-August), occur the crystallization of nuclei and an increase in number of size of calciferites. The extracellular space between the labyrinthic cells and the ductal lumens are lacking of calcific material. Conclusions: Deposits of oviductal calcium exhibits cyclic variations in relation to the reproductive activity. This cation is mobilized and enters massively the ductal lumen for the formation of the egg-shell in the reproductive period. Calcific reserves decline notoriously; and basal labyrinth and ductal lumens are left empty in the post-reproductive period. Calcium storage activates, there is calciferite formation and growth, but calcium does not enter ductal light in pre-reproductive period.

67.**SPERMATIC STORAGE ORGANS IN THE GASTROPOD*****Pomacea canaliculata*: STRUCTURAL INDICATORS OF FUNCTIONAL ACTIVITY**

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Pomacea canaliculata female's, that mate and egg-lays several times during the reproductive season, have the capacity to preserve viable sperms for long periods of time.

The aim of this work is to analyze the functional activity of the copulatrix bursa and seminal receptacle on the basis of its ultrastructural characteristics. Samples were conventionally processed for transmission electron microscopy.

The copulatrix bursa would be the primary sperm storage organ. Its location in the terminal end of the spermatic channel and the presence of a microvilliar epithelium constitute morphological indicators of its participation in the reception and concentration of the ejaculate. The secondary sperm storage organ would be the seminal receptacle that exhibits a glandular region with sacs lined by ciliated and secretory cells and a microvilliar region which stores euphyrene sperms and apyrene parasperrms. Glandular region secretions would favor spermatic survival and the provision of a suitable ambient for the fertilization of oocytes coming from the renal oviduct. The thick muscular cover surrounding the seminal receptacle would drive the entrance of fertilized oocytes to the shell glandular duct. Multiple coupling, spermatic storage in sacciform organs and the sperm prolonged viability would promote clutch multiple paternity, thus facilitating oocyte insemination with sperm coming from different males.

68.**TESTICULAR ANDROSTENEDIONE RELEASE MODULATED BY THE AUTONOMIC NERVUS SYSTEM**

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The male gonads receive nerves from the autonomic ganglia. They converge into the testis along two pathways, the Superior Spermatic Nerves (SSN) and the Inferior Spermatic Nerves (ISN). The SSN arrive to the testis from the Superior Mesenteric Ganglia (SMG) along the spermatic artery, whereas the ISN originate from the Inferior Mesenteric Ganglia (IMG). The objective of this work was to explore if the addition of the neurotransmitters acetylcholine (Ach) and noradrenaline (NA) into the IMG modifies the androstenedione (A₂) release from the testis of adult rats. For that, the already standardized *ex vitro* system (IMG-ISN-testis) was used. Methods: After the surgical operations, the right testis interconnected by the spermatic nerve with the ganglionic plexus was transferred into two separate containers, one for the testis and the other for the ganglion. Ach or NA to a final concentration 10⁻⁶ M, were administered in the ganglionic compartment (experimental). Control samples were obtained without the use of neurotransmitters. The incubation medium placed in each metabolic bath was Krebs-Ringer buffer, pH 7.4, at 37°C. Samples were obtained at 15, 30, 60, 90, 120 and 180 min. The released A₂ was measured by RIA. Results: (ng/mg testis/ml; mean ± SEM). With the addition of Ach in the IMG compartment, the A₂ significantly increases ($P \leq 0.001$). While, by the addition of NA, A₂ diminishes ($P \leq 0.001$) at all the times. Conclusions: Results indicate that the autonomic nervous stimulation modified the testicular hormonal release.

69.

ASSOCIATION OF ROOEX (*Rottboelia exaltata* L. f.) WITH OTHER SPECIES OF WEEDS IN TUCUMÁN SUGARCANE CROPS

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ROOEX is established in the sugarcane crop by means of a population explosion for initiating the process of invasion accompanying with other associated species. The objective was to determine the weed species associated with *Rottboelia* in interference with the sugarcane crop. The essays and samples were realized from 2004 to 2006 in a radio of 600 km², in the localities of: Manantial, Lules, Fronterita and El Bracho. SA methodology of evaluation of weeds in extensive areas and a model for measuring the associative value was employed. For that were analyzed three plots for locality with five determinations into and out of the infested sugarcane crop. The results showed a Direct Association with: *Cynodon dactylon*, *Sorghum halepense*, *Digitaria sanguinalis*, *Euphorbia heterophylla*, *Cyperus rothundus*, *Amaranthus spinosus*, *Ageratum conizoides*, *Talinum paniculatum*, *Alternanthera philoxeroides* and others. Indirect Association with: *Digitaria insularis*, *Ambrosia tenuifolia*, *Eleusine indica* and others. It is concluded that there are two types of associations between ROOEX and the rest of the weeds, ones of the direct type into the crop and others of indirect type outside the crop that are of different order and can reach to a third associative order.

71.

PHYTOMASS AND DENSITY OF *Rottboelia exaltata* L.f. IN INITIAL INVASIÓN OF SUGARCANE cv LCP 85-384 FOR FOUR LOCALITIES OF TUCUMÁN

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R. exaltata is a weed which is in the establishment stage in the Tucumán sugarcane crops. The objective of this essay was quantified the phytomass and density of the weed for relating the losses with the grades of infestation. Plots of 4 m² were sampled of in infested areas of four localities using three replications (Lules, Fronterita, El Cevilar, Esquina). The employed method was destructive. Plants were cut, their number, the fresh weight was determined and it was obtained the dry biomass in stove until constant weight. The production of the cv LCP 85-384 was obtained for sampling of 2 m of central furrow of the plot and the yield analysis was made over the same canes with three replications. The obtained values were extra poled to hectare. Parametric statistical analyses were realized. Results: In high infestation of ROOEX there are 24,30 pl/m² which produce 248 g/m² of phytomass and 44,12% of losses of cane (3,95 kg/m²) and 39,23 % of losses of sugar (0,450 kg/m²). In low infestation there are 4,25 pl/m² which produce 61g/m² of phytomass and losses of 11,19% of cane (0,892 kg/m²) and 9,80% of sugar (0,461 kg/m²). It is concluded that these values establish important competitive characteristics that permit to understand the prejudicial effects of the weed over the sugarcane cultivar essayed.

70.

DEMOGRAPHY AND INFESTATION POTENTIAL OF *Talinum paniculatum* (Jacq.) Gaertn. IN SUGARCANE CROP cv LCP 85-384

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T. paniculatum (TALPT) is a weed of hard control with common herbicides and it is spread with mulching use as a technique of sugarcane culture. The objective was to study the evolution of populations and to determine their potentiality as an infesting weed of the crop. It was worked at El Naranjito locality (Burruyacu - Tucumán) with cv LCP 85-384 between 2005-2006; seven plots were marked of 192 m² in an area of 30 ha and it was measured total plants on 20 December, alive and dead plants on 20 February and survival plants on 20 March. Non-parametric statistical analyses were made. The demographic parameters were: a) Brute rate of mortality: TBM = 665,50 %; b) Brute rate of natality: TBN= 334,50 %; c) Survival over 4589 pl/ha: Sv = 209,58 %; d) Probability of surviving: PSv= 0,0209 %. The individuals with reproductive capacity (ICR) were 921,28pl, the real reproductive capacity (RCR) of 663,32pl and the infestation potential (PI) of 1069,87 m²/pl. There is a marked non equilibrium between births and deaths that characterize the invasion state owing to the intense mechanical management and permanent chemical control of the sugarcane crop. The low survival expresses the fighting condition with the environmental and anthrop factors for setting inside the crop.

72.

SPATIAL AGGRESSIVENESS INDEX (Iea) FOR *Rottboelia exaltata* L.f. (ROOEX) IN SUGARCANE CROPS OF TUCUMÁN (ARGENTINA)

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R. exaltata is considered as the worst weed of sugarcane in the sugar zones of the world. In the productive zone of Tucumán its invasion is recently. It is pretended to determine the index of spatial aggressiveness (Iea) for establishing the incidence over the colonized or invaded territory. This work was realized during 2006 in Tucumán (Argentina) in c.v. LCP 85-384 of sugarcane crops. The Iea in the localities of Manantial, Lules, Fronterita, García Fernández and El Bracho was obtained employing a model (Chaila et al., 2005) which interrelates the height of the original plant, its dry biomass and the surface that occupying with its descendant in the last three years relating with the number, its mean height and its dry average biomass. Samples were made for determining production and sugars. The results of the Iea were: 2,431 (El Manantial); 7,428 (Lules); 1,277 (Fronterita); 6,571 (García Fernández) and 0,822 (El Bracho). The losses found for the different localities were: 26,44 a 44,12% of cane and 21,31 to 39,23% of sugar. It is concluded that the analyzed species had differential spatial aggressiveness indexes depending on the antiqueness of the invasion and the management conditions of the sugarcane crops.

73.**IMMUNOLOGICAL INFERTILITY IN HUMAN REPRODUCTION**

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Introduction: The anti-sperm antibodies (AAE) are the main cause of immunological infertility, can be in the membrane of the sperms, free in seminal plasma (PS), in serum of the man, of the woman and in cervical mucus (isoantibodies)

Aim: It was to study the presence and location of the AAE.

Methods: There was realized a retrospective study of 210 couples who consulted by infertility in The Service of Human Reproduction of the Hospital School "Eva Perón" of Gr. Baigorria from May, 2005 until May, 2007. The presence of the AAE was evaluated in serum of the couple semen and cervical mucus with the test of mixed agglutination TACII (Gatti V, 1998). To all the samples of semen they there was realized espermograma according to procedure WHO (1999).

Results: Of 210 studied couples, 59 (28, 10%) was presenting AAE in someone of 5 samples 21 (35,6%) in alone sperms, 15 (25,4%) in seminal plasma, 6 (10,2%) in serum of the man and 3 (5,1%) in serum of the woman and 4 (6,8%) in cervical mucus and 10 (16,9%) in more than one sample. The AAE not always are in sperms and yes in others biological fluids.

Conclusion: It is important for the study of the infertile couple to realize the determination of AAE in different samples not to lose valuable information and to optimize the therapeutic conduct to continuing.

74.**ZOONOTIC DISEASES THAT AFFECT THE HUMAN HEALTH AND ANIMAL IN THE DEPARTMENT OF SECOND RIVER (2005-2006)**

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The epidemiologic study of zoonotic diseases offers complete information on presentation and maintenance of the same ones in relation to the socioeconomic and cultural context. At moment, there are not previous data, so a Program between the UNRC and the Municipality of city Villa del Rosario (Department Second River) was elaborated. An organic and systematic boarding for the serological study of Brucellosis, Leptospirosis, Chagas and Toxoplasmosis was realized. One hundred individuals and 50 canine were analyzed. From a blood sample by person/animal the following techniques were carried out: Huddlesson, SAT, 2-ME, IDAG (Brucellosis); MAT (Leptospirosis); HAI, IFI (Chagas) and IFI (Toxoplasmosis). Epidemiologists surveys were made and analyzed with Software Epi Info 6.01. The prevalences found in humans were: 0%, 19%, 6% and 62.8% respectively and in canines 8% for Brucellosis, 56% Leptospira and 4% Chagas. The possession of cats of cats was not risk factor for Toxoplasmosis OR=1.67, p=0.43 and Rr=1,33. For Leptospirosis 54% of children lived with some canine serological positive, indicating the potential source of infection. The transference of these results allowed to obtain the diagnosis situation that will be the base of future strategies of prevention and control.

75.**ANATOMIC DESCRIPTION OF VISCACHA (*L...*). PROSTATE GLANDS. SEASONAL HISTOCHEMICAL STUDY**

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The objective of present work is to describe the anatomy of the prostate (P) of the viscacha. This wild rodent has nocturnal habits and exhibits a seasonal reproductive cycle, with a maximum gonadal activity in summer and a minimum activity in winter. The anatomic study was performed in glands fixed with glutaraldehyde (5%). In the histological and histochemical study, the samples obtained from both summer and winter periods were processed by means of conventional techniques and stained with H-E, PAS-Alcian blue (pH 2.5), van Gieson, Tric. of Masson and acid orcein. The P is an ochre-colored gland located at ventral position in relation to the urethra. It is constituted of two partially lateralized lobes, covered by connective tissue and linked by an isthmus. The cranial prostatic region is related to the urinary bladder, seminal vesicles and deferent ducts. Histologically, in relation to the urethra, the P is divided into two zones: central (ZC) and peripheric (ZP). **Active Period:** the ZC epithelium is mainly formed by tall cubic cells and the lumen is PAS (++). In ZP the epithelium is low cubic and the lumen is PAS (+). **Regression Period:** The ZC has a lower cylindrical epithelium with a more intense luminal secretion PAS +. The ZP shows low cubic cells and intense luminal secretion PAS +. The size of the adenomas, the quantity of smooth muscular fibers and the sanguine irrigation are higher in the active period. In conclusion, the P of the viscacha exhibits morphologic and histochemical changes along the reproductive cycle. Probably, they may be conditioned by different hormonal activity in both summer and winter periods.

76.**MELATONIN ADMINISTRATION TO MOTHERS SUPPRESS THE EFFECT OF THE PRENATAL VARIED CHRONIC STRESS ON LDH AND MDH CIRCADIAN RHYTHMS OF RATS TESTIS**

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In mammals, the circadian system in development is synchronized by circadian signals transferred by the mother during gestation. The aim of this study was to know if the maternal synchronization (MS) of the malate (MDH) and lactate (LDH) dehydrogenases circadian rhythms (CR) in pups testis, could be affected by prenatal varied chronic stress (VCS) and the effect of melatonin. MDH and LDH activities were determined in homogenates of testes of 28-days-old pups born from mothers treated with VCS from 10th to 20th day of pregnancy. A group of VCS mothers was injected with melatonin during the last 5 days of pregnancy. Mothers and pups were maintained in constant dark during all the experiment. Enzymatic circadian rhythms was not observed in testes of pups born from mothers submitted to VCS. Maternal melatonin treatment restored the CR of MDH and LDH activities (p<0,05). These results suggest that stressful situations endured by the mother during pregnancy impair the MS of the studied rhythms as a consequence of the absent of maternal melatonin rhythm, the main entrainment signal of the fetal suprachiasmatic nuclei.

Financial support: SECyT- UNC, CONICET.

77.

Azospirillum brasilense PROMOTES TERPENE CYCLASE ACTIVITY UNDER STRESS BY NaCl

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Azospirillum brasilense are plant growth promoter bacteria (PGPB) that improve yield in cereals and others species. The beneficial effects of PGPB are attributed to production of terpenic phytohormones like ABA. ABA production by *A. brasilense* in chemically-defined media was doubled when 100 mM NaCl was added to the medium. In stressed plants FPP is cyclized by sesquiterpene-cyclases (TPS) to produce phytoalexins. The aim of this work was to evaluate sesquiterpene-cyclase activity in *A. brasilense* cultures with NaCl. Bacteria were cultured in NFB medium fed with ^3H -FPP. Addition of 100 mM NaCl enhanced 40-fold the proportion of radioactivity related with terpene cyclase activity. These results suggest that *A. brasilense* elicitate synthesis of sesquiterpene phytoalexin-like compounds under NaCl stress conditions.

79.

MATERNAL MELATONIN RESTORES THE EFFECTS OF PRENATAL CONSTANT LIGHT ON MDH AND LDH CIRCADIAN RHYTHMS OF RAT TESTIS

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In rodents, exposure to chronic constant light (LL) disrupts the overt rhythms by uncoupling the synchrony between the individual oscillators of the suprachiasmatic nuclei. During gestation, the fetus circadian rhythms (CR) develop in synchrony with those of their mother, which in turn are synchronized with the environmental photoperiod. This work analyzes the effect of LL, during a stage of the embryonic period, on the maternal rhythmic signals transmission to fetal circadian system in development, and the melatonin effect. Malate (MDH) and lactate (LDH) dehydrogenases activities showed disrupted CR in testes of 28-days-old rats pups born from mothers submitted to LL from 10th to 20th day of pregnancy, and kept in constant darkness since birth. In contrast, a daily dose of melatonin to LL mothers, during the last 5 days of pregnancy, restored the CR of MDH and LDH. These results suggest a maternal entrainment impairment due to LL action over the fetal circadian system development. It is also emphasized the pineal gland melatonin restoring the function on the studied CR.

Financial support: SECyT-UNC, CONICET.

78.

MATERNAL MELATONIN ATTENUATES THE DEMASCULINIZED COPULATORY BEHAVIOR OF PRENATALLY CONSTANT LIGHT EXPOSED MALE RATS

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In mammals, adverse conditions endured by the mother during pregnancy entail behavioral alterations, affect the rhythmic functioning of the hypothalamic-pituitary-adrenal axis and cause incomplete masculinization of the central nervous system (CNS). In this regard, great significance is acquired by maternal synchronization of the fetal circadian system, regulatory entity of the neuroendocrine system. During gestation fetal circadian rhythms develop in synchrony with those of the mother. Exposure to chronic constant light (LL) disrupts the circadian system, by uncoupling the synchrony between the independent oscillators of the suprachiasmatic nucleus. The influence of LL and melatonin treatment on pregnant mothers and its effects on male adult offspring sexual behavior were investigated. Adult rats born from mothers exposed to LL from the 10th to the 20th day of gestation, showed an increase in their mount, intromission and ejaculation latency, and a decrease in their number, compared to the controls ($p<0,01$). Prenatal LL could impair the copulatory behavior, persisting this effect into adult life. Melatonin maternal treatment during the last 5 days of gestation, significantly attenuated those effects. The LL influence was interpreted as a disruption of the fetal circadian system maternal synchronization, affecting the CNS temporal maturation.

Financial support: Secyt-UNC, CONICET.

80.

MELATONIN ADMINISTRATION TO MOTHERS EXPOSED TO ADVERSE CHRONIC CONDITIONS RESTORES THE ANXIOUS BEHAVIOR IN PUPS

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This laboratory demonstrated that stressful treatment of pregnant rats impairs the maternal synchronization of enzymes circadian rhythms and sexual behavior of the offspring. The aim of this study was to investigate if varied chronic stress (VCS) or constant light (LL) during gestation can affect the postnatal anxiety-related behavior, and the possible effect of melatonin (Mel). In pups of 60 days-old born from rat mothers submitted to VCS or LL, from the 10th to the 20th day of pregnancy, was studied the anxious behavior using the elevated plus-maze. In a group of VCS or LL mothers, melatonin was administered during the last 5 days of pregnancy. Pups from mothers not treated were used as control group. It was observed that prenatal VCS or LL increases the anxiety-like behavior since rats spent significantly more time in open arms ($p<0.05$). After melatonin injections to the treated mothers, pups showed a lower anxiety level similar to that of the control animals. Our results show an increased anxiety behavior in pups from mothers exposed to LL during gestation, reassert the deleterious effects of prenatal stress, and suggest a restitution of the basal anxiety level by melatonin, the main entrainment signal of the fetal circadian clock.

Financial support: SECyT- UNC, CONICET.

81.**ISOLATION OF PSEUDOMONAS FROM MAIZE ROOTS**

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Introduction: Rhizobacteria beneficial to plants are often referred as “Plant-Growth-Promoting Rhizobacteria” (PGPR). Several direct or indirect mechanisms have been postulated to explain how they stimulate plant growth (Lugtenberg *et al.*, 2004). *Pseudomonas* spp are among the bacteria that can be considered PGPR. The objective of the present study was to isolate *Pseudomonas* with PGPR ability from maize roots.

Methods: Bacterial strains were isolated from the bulk soil, rhizosphere and endorhizosphere of maize plants grown in fields of Cordoba Province, in King B and Gould's S1 medium. Gram stain and biochemical tests were realized. Isolates were analyzed for their ability to solubilize phosphate, produce siderophore, indole acetic acid (IAA), extracellular enzymes and inhibit fungi phytopathogens. **Results:** A greater number of isolates was obtained from rhizosphere than endorhizosphere and bulk soil in both King B and Gould's S1 medium. Plant growth promotion mechanisms were analyzed in isolates that showed phenotypic characteristics of *Pseudomonas* genus. We observed that some strains have biocontrol and plant growth promotion potential; For example, DKR39 and DKR41 strains were able to solubilize phosphate, synthesize siderophores, proteases and inhibit *in vitro* *Rhizoctonia solani*. MGR1 synthesize siderophores, cellulases, solubilize phosphate and inhibit phytopathogens.

82.**EFFECTS OF CHRONIC STRESS ON PLASMATICS LIPIDS COMPOSITION IN RATS**

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Stress-induced metabolic changes lead to a increase in plasmatic lipids which would contribute to atherosclerosis and cardiovascular diseases. We studied the pattern and concentration of plasmatic fatty acids (FA) and phospholipids (PL) in rats under immobilization stress (IMO). Male Wistar rats were submitted to IMO (2h/day/7 days). The first day (acute stress, AS) and the last day (chronic stress, CS) of IMO, blood samples were obtained and total lipids (TL) were extracted. The TL were methylated and FA methyl esters (FAMES) were identified by GC. The PL were separated and identified by TLC and the quantization was made by colorimetric method. An increase of PL, sphingomyelin (SM) and phospahtidylglycerol (PG) fraction and a decrease of myristic acids were found in AS. In CS, an increase in SM, myristic and palmitoleic acids were observed. The IMO-induced PL increase could predispose to development of atherosclerotic lesions since when they are oxidized they trigger inflammation that damage blood vessel walls and accelerates the formation of fatty plaques. On the other hand, the changes in the plasmatic FA composition could be related to pathologies such as atherosclerosis, hypertension and cardiovascular disease.

83.**ENZYMATIC ACTIVITIES INVOLVED IN ENERGY GENERATION IN THE CAPACITATION OF CRYOPRESERVED BOVINE SPERMATOZOA**

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Heparin (H) glycosaminoglycan induces bovine sperm capacitation and a respiratory burst. The aim of our work was to determine the variation of metabolism pathways involved in energy generation in heparin-capacitated spermatozoa through malate dehydrogenase NAD(P), isocitrate dehydrogenase NAD(P), lactate dehydrogenase (LDH), alanine and aspartate aminotransferases (ALT and AST), and creatine kinase (CPK) activities. Enzymes activities were registered spectrophotometrically. Chlortetracycline technique was used to evaluate capacitation and samples were capacitated by H. CPK activity decreased (50%) in capacitation with H ($P<0.05$). Enzyme activities ratio obtained for IDH NADP/NAD and MDH NADP/NAD were 2:1 and 3:1 respectively in controls samples. In H-capacitation MDH NADP maintained the same level activity of the control ($P>0.05$). H-induction decreased activities of ALT, AST and LDH ($P<0.05$). Capacitation percentages were increased by H respect to the control ($P<0.05$) and no significant differences were observed in sperm viability. In H capacitation amino acid catabolism and enzymatic activities involved in oxidative substrates sources and energy generation decrease, especially LDH and CPK, suggesting differential participation in the energy generation.

84.**DETERMINATION OF AZOREDUCTASE ACTIVITY IN *Clostridium perfringens* STRAINS ISOLATED FROM FOOD**

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Clostridium perfringens is an anaerobic bacterium present in normal intestinal flora that under certain conditions is known to cause food poisoning and gaseous gangrene in humans, among other pathologies. In humans, food poisoning is produced by the presence of an enterotoxin. It is known that some enzymes produced by the intestinal flora -such as glycosidase, β -glucuronidase, azoreductase and nitroreductase- are associated to conversion of pre-carcinogens to carcinogens, which would strengthen the virulence factors in these bacteria. The aim of this work was to determine azoreductase activity in *Clostridium perfringens* strains isolated from food in San Luis -Argentina, and to contrast enterotoxigenic and non-enterotoxigenic strains. Azoreductase activity was assayed in culture supernatants by reduction of Direct Blue 15 dye, an azo dye that turns from blue to colorless when reduced. Enzymatic activity was determined using the time needed for dye decolorization. All assayed strains showed azoreductase activity; enterotoxigenic strains showed highest activity with lowest dye decolorization time. The presence of azoreductase in *Clostridium perfringens* indicates high virulence; and high pathogenicity of enterotoxigenic strains correlates to their higher azoreductase activity.

85.**EXTRACELLULAR PROTEASES PRODUCED BY ENTEROTOXIGENIC AND NON-ENTEROTOXIGENIC STRAINS OF *Clostridium perfringens* ISOLATED FROM FOOD**

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Clostridium perfringens is an anaerobic Gram-positive bacterium that causes intoxications in animals and humans. In humans, it generates food poisoning due to the presence of an enterotoxin. In addition, several other toxins and enzymes acting as virulence factors are also produced by this bacterium. Production of proteases is related to its pathogenicity. The aim of the present work was to determine protease activity in enterotoxigenic strains (cpe+) and non-enterotoxigenic strains (cpe-) isolated from food from San Luis, Argentina. Thirteen cpe- and fifteen cpe+ strains were used. Bacteria were cultivated in thioglycollate broth at 37 °C during 6 hrs in anaerobiosis. Cell-free supernatants were obtained by centrifugation at 10,000 xg - 10 min, and protease activity determination (Dominguez and Cejudo Method - modified) was performed using 1% azocasein and absorbance measurements were taken at 366 nm after 1 hr incubation. Protease activity was expressed as Units/ml and specific activity as Units/mg of protein. Protein concentration in culture supernatants was determined using the Lowry Method. While cpe- strains showed a specific activity mean of 0.22 U/mg of protein, cpe+ strains showed a specific activity mean of 0.30 U/mg of protein. There is a quantitative difference between the cpe- and cpe+ strains, which can be correlated to the higher pathogenicity of enterotoxigenic strains.

86.***Penicillium* spp. AS SOURCE FOR PRODUCTION OF POTENTIAL COMPOUNDS FOR BIOCONTROL OF *Erwinia carotovora***

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During the course of our screening for active compounds with potential use in biocontrol of phytopatogen bacteria *Erwinia carotovora*, ethyl acetate extracts (EAE) from *Penicillium canescens* *P. citrinum* *P melinii*, *P verrucosum*, *P. roqueforti*, *P. jensenii*, were assayed *in vitro* by agar dilution method. The highest activity was presented by EAE extracts from *P canescens*, *P citrinum* and *P melinii*, with MICs between 5 to 30 µg/ml. A bioassay-guided study by agar dilution method as well as bioautography assay was undertaken to isolate and identify metabolites from EAE extract from *P melinii*. Active extract was successively purified by Sephadex LH-20 and silica gel column to affording the strongest antibacterial sub-fraction, which was analysed by GC-MS, NMR ¹H and ¹³C. The main fatty acids stearic, linoleic, oleic and palmitic acid were identified. Unsaturated linoleic acid showed the strongest activity toward *Erwinia carotovora* with a MIC = 50 µg/ml. Recently has been reported that fatty acid function as the key ingredient of antimicrobial food additives. This work report the potential use of *P canescens*, *P melinii*, *Penicillium citrinum* as source of production of long-chain unsaturated fatty acid for biocontrol of *Erwinia carotovora*.

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87.**CITOTOXIC ACTIVITY OF THE ESSENTIAL OILS OF *ALOYSIA TRIPHYLLA* ON *ARTEMIA SALINA***

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It has been given recent attention to determine the toxicity of extracts and biologically active compounds isolated from aromatic plant species used in herbal medicine. Consequently, in order to find non toxic substances to be used in medicine without side effects, new techniques are being performed. The brine shrimp (*Artemia salina*) assay is a convenient preliminary toxicity test, since it is highly sensitive to a variety of chemical substances. Objetive: to evaluate the toxicity of *A. triphylla* essential oils (EO) on *A. salina*. Materials and Methods: *A. triphylla*'s EO's were screened for toxicity with larvae of *A. salina* (brine shrimp) as described by Franssen *et al.*, (1997). Finney's probit analysis was used to determine the 50% lethal concentration (LC₅₀) of EO's. Toxicity of EO's to *A. salina* is poorly described. It was considered values of LC₅₀>1000 µg/ml non-toxic. The EO's from Rio 1° and Las Viñas were non-toxic on *A. salina* (LC₅₀ = 1279 µg/ml and 3580 µg/ml, respectively). The rest of EO's samples showed different toxicity values. The EO's without toxicity were composed by α-thujone that is considered a neurotoxic compound. It could be suggested that the components included in the EO's are interacting chemically between them and neutralize the toxic effects of thujone in the oil. The chemical composition of the EO's could determine the toxicity of them and non toxic ones should be taken into consideration for therapeutical formulations.

88.**EVALUATION OF ACTIVITY OF PHENOLIC NATURAL COMPOUNDS, CARVACROL AND EUGENOL, ON GABA_A RECEPTOR**

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GABA_A receptor has different recognition sites to specific ligands (GABA) and other allosteric modulators as benzodiazepines, barbiturates, neurosteroids, etc. Many natural products have been assayed by our group as possible modulators of this receptor. One of them, thymol, demonstrated an important activity as positive modulator and GABA agonist. In the present work, it was evaluated the activity of two natural products on GABA_A receptor through the evaluation of the ³H-flunitrazepam binding in sinaptosomal membranes from chicks forebrain. The assayed compounds, carvacrol and eugenol, share in their structure the presence of a phenolic ring. The results showed a positive effect on ligand binding induced by both compounds, with EC₅₀ values of 157_{Carv} y 171_{Eug} µM and maximal stimulation of 160_{Carv} y 143_{Eug} %. Since these phenolic compounds are highly lipophilic, we analyzed the non-specific ligand binding, which showed non-significant changes respect to control. Concluding, the results indicate an agonist/positive modulatory-like activity for both compounds. Additional studies tending to confirm this activity (evaluation of their effects on ³H-flunitrazepam binding in the presence of known GABA antagonist and determination of chloride uptake) are being investigated by our laboratory.

89.**EFFECT OF DEHYDROLEUCODINE IN THE PROLIFERATION OF HeLa CELLS***Costantino VV, López LA.**Laboratory of cytoskeleton and cell cycle. Instituto de Histología y Embriología. Facultad de Ciencias Médicas. Universidad Nacional de Cuyo. Mendoza. E-mail: vcostantino@fcm.unco.edu.ar*

Dehydroleucodine (DhL) is a sesquiterpenic lactone purified from the medicinal plant *Artemisia douglasiana*. Previous results of this laboratory showed that DhL delays the proliferation of meristematic cells of *Allium cepa* roots and vascular smooth muscle cells. DhL arrested the cells in G2 phase. In this work we analyzed if DhL inhibit the proliferation of tumor cells. Cultured HeLa cells were used in the assay. 1×10^4 HeLa cells cultured in quiescence for 24 h, were stimulated with 10 % of SFB in the presence of 0-4 μM of DhL by 96 h. The growth index (IC \pm ESM) were assayed and the results were analyzed by the test of the Variance. After 96 h of culture the control (0 μM of DhL) showed an IC of $11,8 \pm 0,48$, with 1 μM DhL $8,3 \pm 0,52$, with 2 μM $8,03 \pm 0,21$, with 3 μM $6,3 \pm 0,74$, and with 4 μM $2,3 \pm 0,09$. The statistical analysis indicated significant difference between the studied groups. In base of this results it is clear that DhL is very effective in inhibiting the proliferation of tumor cells.

90.**OVIDUCTAL CALBINDIN D 28 KD, CALMODULIN AND Ca^{2+} PUMP DURING THE SEXUAL CYCLE OF *Bufo arenarum****Crespo CA, Ramos I, Medina MF, Cisint S, Cruz López ME, Fernández SN.**Dpto. Biol. del Desarrollo INSIBIO (CONICET-UNT). Chacabuco 461. 4000- Tucumán. E-mail: ccrespo@fbqf.unt.edu.ar*

Ultrastructural and biochemical studies, performed in our laboratory, demonstrated for the first time that in anuran, the epithelial (ESC) and glandular (GSC) secretory cells of the oviductal pars convoluta (PC) contain Ca^{2+} deposits and active Ca-ATPasa pump. Both of them show variations according with the period of the sexual cycle. Likewise it was demonstrated that Ca^{2+} , also present in the jelly coats organized around the oocytes during their transit by the PC, plays an important role in the gametic interaction. The aim of the present study was to identify and localize, by indirect immunohistochemical techniques, the presence of calbindin D 28 KD (Cb), calmodulin (Cm) y Ca^{2+} pump at different regions of the PC throughout the sexual cycle.

Before ovulation, in the first zone of the PC, only some ESC and GSC evinced colocalization of the three proteins. In the rest of the oviduct Ca^{2+} pump is present only at the ESC membranes, while both Cm and Cb predominate in the apical zone of the ESC. During the ovulatory period the first zone of the PC show all the proteins analyzed localized at the luminal level of the ESC and GSC. In the rest of the duct the reactivity predominates at the apical region of the ESC. At the basal region of the GSC it could observe only Ca^{2+} pump reaction. After ovulation the proteins studied are limited at the ciliated cells and basal region of the ESC. The evinced polarity and the colocalization of Cm, Cb and calcium pump suggest the active transport of Ca^{2+} to the PC lumen for its incorporation to the jelly coat.

91.**STUDY OF APOPTOTIC WAYS IN PORCINE PLACENTAS***Cristofolini A, Merkis C, Zubeldía D, Vaquer V, Barroso F, Lloret M, Hechem B, Schleef N, Koncurat M.**Área de Microscopía Electrónica. FAyV. Universidad Nacional de Río Cuarto. Río Cuarto. Córdoba.**E-mail: acristofolini@ayv.unrc.edu.ar*

The apoptosis is a fundamental physiological process, that it plays an important role in the cellular remodeling as much in the embryonic development as of placenta in most of the mammals. The aim of the present work was to determine the expression of the receptors of death FAS B-10, FAS C-20, FAS ZB4 and DR-4 and the presence of the mitochondrial regulating protein Bcl-2; in addition the technique of TUNEL like apoptosis control was used. Histological slides of ± 4 mm of porcine placenta of $\pm 30, 55, 70$ and 114 days of gestation were used. For the detection of the cell death receptors and the regulator Bcl-2 commercial antibodies and for the TUNEL technique the ApopTag® equipment were used. From the beginning and to the 70 days of gestation the expression levels of the membrane receptors FAS and DR-4 in the epithelia of the villi, were negative. As of day 70 and even the end of the gestation, receptors FAS expressed an abundant expression. The expression of the mitochondrial regulator Bcl-2 in the villi was negative. At the beginning of gestation the apoptosis would be by the intrinsic way, whereas as of day 70 of gestation this phenomenon would occur through the ways of FAS receptors.

92.**HISTOLOGICAL AND HISTOCHEMISTRY ANALYSIS OF THE ORPINGTON HEN OVIDUCT***Cruz López ME*, Crespo CA, Cisint S, Ramos I, Fernández SN.***Fac. Cs. Nat. e IML y Fac. Bioq., Qca y Farm, UNT.*

As other birds, the reproductive system of the Orpington hen is formed only by one ovary and oviduct. By means histological and histochemical techniques it was analyzed the morphology and contents of the oviduct during the laying period. The oviduct wall is formed by four tunics: mucous, submucous, muscular and serose. Along this duct the mucous organization changes from a simple cylindric ciliated epithelium at the beginning of the infundibulum, to pseudostratified epithelium constituted by ciliated and caliciform cells which contain acid and neuter glyconjugates, in the rest of the oviduct. The connective chorion present serous eosinophilic tubular glands, except in the vagina. At the magnum level the glandular cells show variation in form and eosinophilic content amount and some of them present a refringent and crystalline material. The proximal isthmus exhibits serous glands containing eosinophilic granules not present in other sectors of this oviductal segment, the glands lack of the cytoplasmic contents. The submucous layer does not show any variation along the duct.

The smooth muscular tunic exhibits isolated fascicles disposed in different direction in the infundibular ostium while in the rest of the oviduct this tunic is organized in two layers which are more developed in the vagina. The serous layer shows a simple epithelium resting on lax connective tissue. The variability of contents which exhibit the epithelium coat and the tubular glands are in close relation with the diverse material which contribute to form the egg envelopes.

93.

MORPHOMETRIC DESCRIPTION OF LYMNAEA SNAILS FROM BEAGLE CHANNEL, TIERRA DEL FUEGO
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Studies related to Lymnaeid snails from southern Argentina are very scarce, in spite of its importance in the transmission of fascioliasis. Our objective is to describe morphologically a population from Tierra del Fuego and thus characterize it more fully. On the margins of Lasifashaj river, 83 *Lymnaea* snails were collected. Their identification was done according to the keys of Castellanos *et al.* In the laboratory, using a calibrated microscope, we measured (in mm.) the following parameters: Shell length (SL), Aperture Length (AL), Last Spire Length (LSL), Aperture Width (AW), and Shell Width (SW)

	LS	AL	LSL	AW	SW
Mean	8,11	4,56	6,84	3,26	4,60
SD	1,73	0,92	1,46	0,69	0,96

These results are similar to those of *L. diaphana*, species that is distributed all over Patagonia, including Tierra del Fuego. This information will allow, together with future morpho-anatomic and molecular studies, characterize the populations of Lymnaeid snails present in Patagonia.

94.

ULTRASTRUCTURAL CHARACTERIZATION OF CIRCULATING HEMOCYTES OF *Pomacea canaliculata*
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Hemocytes constitute the cellular component of the defense system of invertebrates. The types of circulating hemocytes has not yet been established in this species, and this is the first morphological description of the circulating cells of *P. canaliculata*. Hemolymph was obtained by heart puncture, treated with EDTA to avoid agglutination of the hemocytes, centrifuged and fixed with Karnovsky's fluid. Hemocytes showed nuclei with abundant chromatin clumps, many of them lining the nuclear membrane. A well developed rough endoplasmic reticulum was observed, that was made of extended but not stacked cisternae, and that were lined by orderly arranged ribosomes. The Golgi apparatus can be rarely observed. Most cells were found emitting short and filamentous pseudopodia. The scarce mitochondria are ovoid and they have tubular cristae. Two hemocytes' populations can be distinguished on the basis of their cytoplasmic granules: (1) the "granulocytes", with an eccentric, kidney-shaped nucleus, club-shaped granules that were both abundant and electron dense, and a moderately developed smooth endoplasmic reticulum; and (2) the more abundant "hyalinocytes", with a spherical nucleus, abundant smooth endoplasmic reticulum, and a few cytoplasmic granules of moderate electron density.

95.

EFFECTS OF RESTRICTED FEEDING ON GROWTH, FEED EFFICIENCY AND CIRCULATING CONCENTRATIONS OF IGF-I IN ANGLO NUBIAN DOES AND ITS CROSSES BY SAANEN
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The aim of the experiment was to evaluate the effect of intake restriction on liveweight gain, relative growth rate and IGF-I concentration in does. Sixteen 4 month old does were assigned by breed to two groups: Anglo Nubian (AN) 9.26 ± 0.28 (n=8) and AN x Saanen (CRO) 9.78 ± 0.47 kg (n=8). Feed were composed with alfalfa hay chopped (70 %) and ground corn (30 %). Energy was 2.4 Mcal/kgDM and CP 16 %. Feed was offered in individual cages in order to control daily intake. Does were fed at ad libitum level of intake and then subjected to a restricted feeding (70 % ad lib) expressed as BW % in both groups, for a 92 days period. Once a week blood was collected. IGF-I concentrations were quantified by ELISA (DSL®). "t" Student test was used to compare means. CRO does were heavier 17.41 ± 1.93 vs. 13.61 ± 1.48 kg and had a greater weight gain 38.61 ± 9.93 vs. 20.39 ± 7.16 g/d than AN does ($P<0.01$). Relative growth were consistently higher ($P<0.01$) in CRO than in AN does. IGF-I concentrations were higher in CRO. At the end of restriction period values were 291 ± 136 and 126 ± 26 ng/ml for CRO and AN does respectively ($P<0.05$). This study suggests that crosses does had a better adaptative response to feed restriction than pure ones and changes in serum concentrations of IGF-I may contribute to growth differences in the restricted feeding observed among biotypes.

96.

RISK AND PROTECTIVE FACTORS FOR NON MELANOMA SKIN CANCER DEVELOPMENT
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A case-control study was carried out in Rosario, Argentina, to evaluate Basal Cells (BCC) and Squamous Cells Carcinomas (SCC) risk factors. Twenty-two cases (mean age: 65.1 ± 15.1) and 35 controls (64.5 ± 12.3). 31 tumors were diagnosed (BCC:24, SCC:7). 45% of cases and 42.7% of controls had sunburns antecedents. 72.7% of cases and 37.1% of controls had actinic keratosis, and 27.3% of cases and no controls had more than 10 naevi. Skin cancer family history was found in 22.7% of cases and 5.9% of controls. High consumptions of Vitamin A was found in 18.2% of cases and 28.6% of controls, Vitamin E in 18.2% and 34.3%, and Vitamin C in 27.3% and 31.4%. Six cases (27.3%) and 10 controls (28.6%) were smokers. Mean habit duration were 32.2 ± 9.1 years for cases and 24 ± 3.8 for controls. When adjusting a multiple logistic regression model to estimate odd ratios -controlled for the confounding effect of the studied variables- the possibility to have BCC or SCC was higher in patients with 10 or more naevi ($p=0.03$), actinic keratosis ($p=0.05$), vacational sun exposure ($p=0.04$), and smaller in those high vitamin C ingestion ($p=0.04$). Even though sun exposure is the most important risk factor for Non Melanoma Skin Cancer, the risk could be increased or reduced by the effect of other variables.

97.

PATH COEFFICIENT ANALYSIS FOR COMPONENTS OF SEVERITY DEGREE IN “MAL DE RÍO CUARTO” DISEASE

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Path coefficient analysis allows dissecting the correlation between two variables into effects, direct and indirect, establishing between them a cause-effect relation. This analysis was carried out with the objective to interpret genotypic correlations between severity degree, caused by Mal de Río Cuarto disease (MRC), and seven related traits. One hundred and twenty recombinant inbreed lines, derived from a cross between Mo17 and BLS14, a susceptible and a tolerant inbreed line respectively, were evaluated during the 2006/2007 summer seasons at the Río Cuarto National University. The analysis showed that internode and empty ear presented a positive and significant direct effect on severity degree. Plant height exerted a notable indirect effect not only via leaf width and leaf length, but also via internode, in spite of its low direct effect. Tassel presented a negative and significant direct effect even though the correlation with severity degree was positive. Tassel, empty ear, leaf length and leaf width had an important indirect effect via internode. Consequently, internode, plant height, tassel and empty ear are effective traits to determine severity degree in breeding and selecting plant programs related to MRC.

99.

EFFECT OF THEOPHYLLINE ON AMYLASE ACTIVITY SECRETED FROM MURINE SUBMANDIBULAR GLANDS

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Bitter compounds are known to elicit specific effects on the gastrointestinal tract and exocrine glands. We have studied the effect of theophylline (TEO) on the salivary amylase activity of submandibular BALB/c glands (SMG). SMG were isolated and treated ex vivo to evaluate the activity of secreted amylase (mg maltose/min.g.wet tissue). TEO inhibited amylase activity in a dose-dependent manner, being 10^{-8} M the maximum inhibitory concentration (TEO: 0.08 ± 0.01 ; Basal: 0.180 ± 0.004 ; p<0.01). We investigated the role of the phospholipase C (PLC)-nitric oxide synthase (NOS) pathway, treating GSM with the PLC inhibitor, NCDC (10^{-6} M) (0.110 ± 0.01 ; n=3) or the NOS inhibitor, L-NMMA (10^{-4} M) (0.106 ± 0.01 ; n=3) previously to TEO (10^{-8} M) and both reverted the effect of the bitter compound. Neither the muscarinic blocker atropine (10^{-5} M) (0.120 ± 0.001 ; n=3) nor the β -adrenergic antagonist propranolol (10^{-6} M) (0.120 ± 0.007 ; n=3) modified the action of TEO on amylase secretion. We conclude that TEO inhibits the release of amylase, involving the PLC-NOS pathway, without participation of muscarinic or β -adrenergic receptors.

98.

UREAPLASMA UREALYTICUM AND MYCOPLASMA HOMINIS SENSITIVITY TO LACTOCIN L23

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Ureaplasma urealyticum (Uu) and *Mycoplasma hominis* (Mh) are important pathogen microorganisms associated with urethritis, cervicitis and cystitis. The objective of the present work was to study the correlation between the sensitivity to lactocin L23 and the sensitivity to antibiotics of Uu and Mh, both isolated from sexually active women. At the present time, lactobacilli constitute an alternative as preventive and biotherapeutic agents of these pathologies. The samples from endocervix were processed with the MYCOFAST® Evol 2 equipment. The antimicrobial activity was performed through an adaptation of the commercial technique. Uu was detected in 59,5% of the 190 studied samples, Mh in the 7,6% and both Uu + Mh in 32,9%. All of the strains were inhibited by lactocin L23. 95% of the strains of Uu were inhibited with a MIC of lactocin of 80-320 UA/ml, and 78% of Mh's with 320 UA/ml. A pronounced resistance to roxitromycin was observed in Mh and Uu + Mh in the assay of susceptibility to antibiotics. In conclusion, these results show the inhibitory activity of lactocin L23 upon urogenital mycoplasmas, because it presented inhibitory activity even when diluted; this is a remarkable fact keeping in mind the resistance to antibiotics of clinical use presented by these microorganisms.

100.

EFFECT OF AQUEOUS EXTRACTS OF *LARREA DIVARICATA* (JARILLA) ON NITRIC OXIDE PRODUCTION ON MURINE MACROPHAGES CO-CULTURED WITH *CLOSTRIDIUM* Sp.

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Clostridium chauvoei and *Clostridium septicum* are anaerobic and sporulated bacteria. The former produces blackleg in cattle, sheep and other ruminants, and later, causes in humans gas gangrene. *L. divaricata* is a folk medicinal plant that stimulates the activation of murine macrophages (MØ), and the production of nitric oxide. NO may induce apoptosis. The aim of this work was to assay the effect on NO production and viability of decoction (D) and infusion (I) of jarilla (0.2 mg/ml) on MØ co-cultured with *Clostridium chauvoei* and *Clostridium septicum*. The nitric oxide production was determined by griess reaction and the cell viability was tested by the MTT assay. Both were tested 1.5 hours post incubation. D and I increased the production of NO with *C. chauvoei* (p≤0.01). Instead, only D increased significantly the NO production when *C. septicum* was co-cultured (p≤0.02). The cellular viability was not modified by the treatment with D, I and bacteria. These results showed that the aqueous extracts of *L. divaricata* induce the NO production in co-cultures of MØ and *C. chauvoei* or *C. septicum*. As the cellular viability remains unchanged and that there are an increase in the level of NO it is probable that these microorganisms can be eliminated.

101.**PRIMARY HEALTH CARE AND SOCIAL SECURITY IN SAN LUIS: PREVALENT DIAGNOSES***De Pauw M, Rapisarda M, Pelzer L, Valsecia M, Calderón C.**Farmacología UNSL, Farmacología UNNE.**E-mail: ccal@unsl.edu.ar*

Our aim was to determine and to compare age, sex and prevalent diagnoses of concurrent patients at 2 hospitals and a social security (SS) of San Luis. An observational, cross-sectional and retrospective study was carried out. The data from the Cerro de la Cruz (CC) and del Sur (HS) Hospitals, and a (SS) were collected for 1 month; the diagnoses were classified according to classification ICD-10 and the prevalent pathologies were studied. Statistic: difference of proportions. Results (% for CC, HS, OS, respectively): Age (years): 0-14: 38.2, 36.5, 10.5; >14: 57.9, 62.3, 89.5. Sex: F: 62.1, 57.6, 63.2; M: 37.9, 42.5, 36.8. Diagnoses: respiratory (J): 24.1, 35.7, 16.7; digestive (K): 11.6, 8.4, 20.9; to osteomuscular (M): 9.1, 8.1, 14.5; genitourinary (N): 8.2, 4.4, 9.6; infectious and parasitic (AyB): 7.7, 6.2, 1.0; circulatory (I): 7.7, 4.8, 35.4; behavior upheavals (F): 0.6, 0.3, 11.7; endocrines (E): 1.8, 6.2, 21.6. Prevalent pathologies: Diabetes (DBT): 1.4, 5.1, 9.4 ($p<0.0001$) Hypertension (H): 6.3, 4.8, 24.5 (CC vs HS $p<0.04$; CC, HS vs OS $p<0.0001$). Arthritis (A): 0.5, 0.8, 3.7; Osteoporosis (OP): 0, 0, 2.2; Menopause (M): 0, 0.1, 1.7. Anxiety (An): 0.2, 0.1, 6.2. Depression (D): 0.3, 0.2, 3.3 (OS vs CC, HS $p<0.0001$ for H, A, OP, M, An, D). The population was mainly female in all the cases, in the SS there was a greater adult attention, prevalence of chronic diseases (H, DBT and A) and consulting about less important symptoms like the M. In hospitals the prevalent pathologies were acute ones, as well as the respiratory illnesses.

102.**NEW CONTRIBUTION ABOUT LIPIDS CARRIERS IN HUMAN SALIVA***Defago MD, Garces NR, Valentich MA, Actis AB.**Instituto de Biología Celular. Fac. Cs. Médicas (Univ. Nac. Córdoba). E-mail: danieladefago@hotmail.com*

Introduction: saliva reflexes the physiological conditions of organism through determinations of hormones, immunoglobulins and drug monitoring, but there is not data referred to the presence of lipoproteins in human saliva. Objective: to determinate the presence of lipoproteins in human saliva. Method: human saliva samples of both sexes with ages between 16 and 52 years old were collected according to international standars. The determinations of apolipoproteins A and B were made on saliva supernatant obtained by centrifugation (800xgx10min), measured in autoanalyser through immunoturbidimetric test and expressed as average (mg/dL) +/- D.S. Results: the level of apolipoprotein B in women was 11.25 +/- 3.2 and in men 17 +/- 2.79. Apolipoprotein A was not detected in any sample. Discussion: the results found are the first data of existence of lipid carrier in saliva. It should be adjusted to protocol to enlarge essays and make correlations with serum apolipoproteins.

103.**DISACCHARIDASES IN THE SUBTERRANEAN RODENT *Ctenomys talarum*: RELATION TO ENERGETIC COST OF REPRODUCTION***del Valle JC¹, López Mañanes AA^{1,2}.**¹Dpto Biología, FCEyN, UNMDP, Funes 3250. (B7600) Mar del Plata, ²CONICET, E-mail: delvalle@mdp.edu.ar*

Pregnancy and lactancy are energetically expensive periods for *C. talarum*. The aim of this work was to determine the relationship between disaccharidases activities and the energy cost of reproduction. Sucrase and maltase activities were determined in small intestine of *C. talarum* captured in Mar de Cobo (Bs. As., Argentina). 13000xg supernatants were used as enzyme extract (homogenization medium: 0.15 M NaCl) (20 mL g⁻¹ of tissue). Activities were assayed by a colorimetric method (sucrase or maltase 28 mM in 0.1 M maleate/OHNa pH 6.5 at 37°C. Reproductive females exhibited lower sucrase and maltase activities than non-reproductive ones ($t=2.8$ df=8 $p<0.0021$). No differences were found between males in different reproductive status ($p>0.05$). In reproductive females maltase activity appeared to be accounted for the sucrase-isomaltase complex (maltase= -35.23 ± 7.74 x sucrase; $r=0.92$, $p<0.0025$), but it was independent of sucrase-isomaltase complex activity ($p>0.05$) in non-reproductive females, immature and mature males. Our results show the occurrence of qualitative changes of disaccharidases activities in females of *C. talarum* in relation to increased energy demands upon pregnancy and lactancy.

104.**SELECTION AND CHARACTERIZATION OF CHITINASE PRODUCER MICROORGANISMS***Defini CD, Martínez MA, Figueroa LIC de, Siñeriz F, Baigorí M. PROIMI-CONICET. Av. Belgrano y Pje. Caseros, 4000 Tucumán, ARGENTINA. E-mail: cdelfini47@hotmail.com*

The producing microorganisms of chitinase are of great interest in agriculture due to their potential contribution as biological control of pest such as fungi and insects.

In a previous work, 21 bacteria with chitinase activity were isolated from bat feces. In the present work, the best chitinase producers were selected and studied. Those isolates were cultivated in a rich medium (YEFD) until they reached an OD_{540nm} of 0.5, which was correlated to 10⁸ UFC mL⁻¹. 20 µl of each culture were inoculated in minimal media supplemented with colloidal chitine as the only carbon source. Those isolates which showed the biggest clear area were selected as the best producers. Its ability to degrade chitin was quantified by the p-dimethylaminobenzaldehyde method and characterized through 16S rDNA sequence amplification and analyses.

Results: 11 isolates over 21 were selected due to their chitinase activity by measuring clear areas in minimal media with chitin as the only carbon source. Quantitative assays showed 6 isolates as the best ones: one of them belonging to *Streptomyces* genus and the other ones belonging to *Bacillus* genus.

105.**DIFFERENT RESPONSE OF PREPUBERTAL AND FIRST PROESTROUS OVARY TO CHOLINERGIC INFLUENCES**

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The cholinergic effect on the coeliac ganglion (CG) has been described in the prepubertal ovary of rats; yet, it remains unstudied in the first proestrous of the estral cycle. Purpose: to elucidate the effect of Acetylcholine in the ganglion compartment in the ex-vivo CG-superior ovarian nerve-ovary system in rat on the ovarian release of Progesterone (P), androstenedione (A_2) and nitric oxide (NO) in the first proestrous and to compare it with the prepubertal ovary. The system was isolated and incubated in Krebs-Ringer-glucose buffer in a metabolic bath in the presence of Ach (10^{-6} M) in CG. Aliquots from the ovarian compartment were taken at 15, 30, 60 and 120 min. P and A_2 were measured by RIA, and NO, by Griess reaction. Statistical significance by Student's *t* test at $p < 0.05$. Results: Ach caused a significant increase in P and A_2 release at all the studied times ($p < 0.05$) in the first proestrous and a decrease at 30 days. NO showed similar values in the first proestrous and at 30 days, and the ganglion cholinergic stimulus increased liberation in both cases ($p < 0.01$ and $p < 0.001$, respectively). Conclusion: the ganglion cholinergic stimulus in the first proestrous favours the ovary maturation through the increase of the hormonal secretion partly due to the low liberation of NO in relation to the prepubertal stage.

106.**EXPRESSION AND FUNCTION OF RENAL OAT3 IN RATS WITH ACUTE RENAL FAILURE**

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OAT3 is a protein localized in the basolateral membrane of renal cells in proximal, distal and collector tubules and in the thick ascending limb of Henle's loop. OAT3 is primarily involved in the renal secretion of organic anions. The present study was carried out in order to evaluate the renal excretion of p-aminohippurate (PAH, prototypical organic anion, single dose of 30 mg/kg b.w., i.v.) and OAT3 expression in renal cortex (C) and medulla (M) by immunohistochemical techniques and Western blotting. Male Wistar adult rats were used throughout the study, divided in two groups: one with bilateral ischemic acute renal failure (ARF, $n = 4$) and other that underwent sham operation (Sham, $n = 4$). Statistical analysis of the results was performed using an unpaired *t*-test ($*P < 0.05$). The renal excretion of PAH was significantly diminished in ARF group ($\mu\text{g}/\text{min}/100 \text{ g b.w.}$; Sham = 33 ± 8 , ARF = $0.42 \pm 0.21^*$). Two bands with different molecular weight (130 and 77 kD) were detected by immunoblotting. The respective expression was as follows: 130 kD (C: Sham: 100 ± 7 , ARF: $258 \pm 17^*$; M: Sham: 100 ± 3 , ARF: $184 \pm 9^*$); 77 kD (C: Sham: 100 ± 4 , ARF: $78 \pm 5^*$; M: Sham: 100 ± 2 , ARF: $76 \pm 4^*$). These results were confirmed by immunohistochemistry. In the presence of ARF, a different regulation was observed for the two protein's bands both in renal cortex and medulla. The reduced expression of the 77 kD band would contribute, at least in part, with the decrease in the renal secretion of organic anions observed in ARF rats.

107.**MICROSATELLITES ASSOCIATED WITH TOLERANCE TO MAL DE RÍO CUARTO VIRAL DISEASE**

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In Argentina, Mal de Río Cuarto (MRC) is the most important viral disease of maize. The objectives of this investigation was to identify microsatellites *loci* linked to quantitative trait loci (QTL) contributing to resistance to MRC disease in maize. The materials consist of $F_{2:6}$ recombinant inbred lines (RILs) derived from crosses between Mo17, a public dent line susceptible, and BLS14, a flint line tolerant. The RILs were descendants from the early generation population in which MRC-QTL were first identified. DNA profiles were obtained using 40 microsatellites. The RILs were evaluated for MRC disease throughout four field experiments conducted in a maize crop region of Argentina, which is endemic for MRC disease. Based on single-factor analysis of variance (ANOVA), microsatellites *loci* located in chromosome 1 and 4 were associated with resistance to MRC. The microsatellites *loci* associated with resistance to MRC were highly consistent across environments. These microsatellites could be promising in molecular marker mapping for MRC resistance and consequently, marker-assisted selection.

108.**HISTOLOGICAL STUDY OF TISULAR LESIONS IN PREGNANT RATS INTOXICATED WITH CADMIUM**

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Cadmium (Cd) is a heavy metal of great environmental toxicological relevance. Several works were performed on the effects of low and repeated doses intoxication. However, few works deal with effects of Cd high doses. In our lab, we are studying alterations determined by single subcutaneous doses of Cd (10 mg/kg, as CdCl_2) in pregnant rats. We report lesions observed in liver, kidney, spleen and lung and the relationship with their Cd tisular concentrations in each organ. Groups of 6 rats intoxicated on days 4, 7, 10 and 15 of pregnancy were used. All were sacrificed on day 20 of gestation. The above mentioned organs were collected and stained with H/E. Cd concentration was determined by atomic absorption spectrophotometry. Most common lesions were: Liver: congestion and hydric complications. Lungs: emphysema or atelectasis, necrosis and cellular infiltration. Spleen: haemorrhages and white tissue loss. Kidneys: hydric complications and tubular and corpuscular necroses. Lesions were similar for all administration days. These results are in correlation with Cd tisular concentrations, which were similar in the experimental groups.

109.**PREVALENCE OF INFECTION AND RELATIVE FREQUENCY OF ISOLATION OF PATHOGENS ASSOCIATED TO MASTITIS IN DAIRY HERDS OF THE MILKY AREA IN VILLA MARÍA (CÓRDOBA)**

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During the last 15 years, a reduction in the bovine milk production as well as in the number of herds has been observed in Argentina. There are few studies about infection prevalence (IP) using large numbers of dairy herds (DH) in the central region of our country. The main of this study was to determine the IP in cows from DH of the milky area of Villa María (Córdoba) and also the relative frequency of isolation of pathogens involved. Composite milk samples were obtained from 858 cows, selected systematically at random from 20 DH during the period March-June. Bacterial identification was done using accredited methodology based on NMC standards. Bacterial growth was detected in 78% of the samples, which is significantly more than in previous studies. No growth was found in 15% of samples. Among the samples with bacterial growth, staphylococci was the most commonly isolated bacterial group (80%), followed by the streptococci (6%). Both microorganisms were found associated in 5% of the samples. Frequency of isolation of staphylococci was high compared that in other studies. Although this is a preliminary study, it will be the first survey of IP on such a large scale in Córdoba. At the end of the 2007, a total of the 50 DH will be studied and also the molecular bacterial identification will be carried out. Finally, it will contribute to have real knowledge of the infectious mastitis in DH of Villa María (Córdoba).

110.**HISTOPATHOLOGIC FINDINGS IN CLINICAL MUCOSAL SURFACE LEUKOPLAKIAS**

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On mucosal surface white lesion are described. Histologically we can found alterations that justify it conduct as forerunner lesions. The objective was to find in a sample (n=74) some changes that indicate dysplasia epithelial. Obtain new sections of cuts from register material for evaluate dysplasia epithelial parameters, and differentiate its in low levels, altered mature and differentiation conserve (hyper or parakeratosis, low hyperplasia of basal cells, big nucleus in all the epithelium- conservation of intercellular bridges). Moderate with a discreet alteration of the differentiation (hyperplasia of conserve not atypical mitosis). Serious with immature epithelium and bad differentiation (architecture very altered). In the series it has been found epithelial changes whit this distribution: Acanthopapillomatous (25.7%), Acanthosys (24.3%) hyperkeratosis (23%) papillomatous (14.8%) hiperchromic (4%) basal activity (2.7%) basal hyperplasia (2.7%), basal atypical (2.7), others (1.4%). The significant changes are similar with an alteration maduration but the differentiation is conserved, changes that definir low and moderate dysplasia. It infers that a clinical white lesion can present diverts madurations and differentiation in different levels. This diverts are relationship with the possibility of malignant transformation and with the prognostic.

Histotechnologist: Del Viso, S.

111.**DIMENSIONAL GROWTH STUDY OF EXPERIMENTAL FREE RANGE BROILER HYBRIDS**

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The growth of experimental hybrids of free range broilers (CP: Cornish x Plymouth Rock Barrado and CR: Cornish x Rhode Island Red) compared to a reference population (Campero INTA) was dimensionally characterized from a non-lineal fit of longitudinal data: body weight-age (BWA) and shank length-age (SLA) by means of Gompertz and logistic models respectively. The estimates of asymptotic size (ASI) and maturing rate (MAD) for each variable (ASIPES, MADPES, ASICAN and MADCAN) were analysed with the multivariate technique of principal components. The first component (PC1) explained 69% of the phenotypic variance and grouped birds by their size being CP and CR smaller than Campero INTA. The second component (PC2), which accounted for 20% of the variance, is a harmonic development component. The CR hybrid might show a more harmonic pattern by combining lower asymptotic values and higher maturity rates. The third component (PC3: 8% of the variance), is also a size component but it positively associates asymptotic values with maturity rates. PC1 and PC3 allowed for a higher maturity size assignation to Campero INTA, although the alternative hybrids show a growth pattern compatible with the expected slaughter weight at the aimed chronological age.

112.**TIPOLOGY OF THE SYNFLORESCENCE AND CLEISTOGAMY OF *CLITORIA CORDOBENSIS* (FABACEAE)**

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Clitoria cordobensis Burkart is an endemic perennial herbaceous plant of the mountain ranges of Cordoba. The objective of this work was to study the typology of the synflorescence and floral morphology. Studies at field were made during years 2003 to 2007 in four natural populations that grew in the Punilla Valley, Córdoba. Samples were taken for their later analysis in the laboratory. The plants have aerial orthotropic axes with little buds of innovation and with very few branches with voluble apexes and truncated polytelic synflorescence. The paraclade is reduced to its coflorescence, hypotagma is not developed, the axis is very short and presents between 4-6 flowers in axillaries fascicles. Chasmogamic and cleistogamic flowers were observed, these were reduced to a small calyx, did not possess corolla, the androecium was reduced to only three stamens, the position of the anther of the vexilar stamen coincided, and it leaned in the stigmatic surface, the style was short and curved. The ovarian development was manifested immediately, the style was not extended, stigma was separated of the anther by the growth of the fruit. According to the observed data the increase of cleistogamic flowers at the end of the flowering guarantees the production of fruits and seeds.

113.

TYPES OF ENAMEL IN THE INCISIVE TOOTH GROUP

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Koenigswald classified mammalian enamel microstructure according to complexity levels. The level enamel types refers to the relationship both among prisms themselves and to the amelodentinal boundary but it has not yet been applied in the dentistry field. The aim of this work was to identify the types of enamel in sequential sections, in both longitudinal and cross planes of crowns of permanent upper incisives in humans using a grinding technique for SEM. From each piece 2 hemi sections were obtained. They were embedded in resin, grinded, etched with acid, washed, metallized and observed under SEM. Micrographs were obtained from incisal, medial and cervical regions in longitudinal sections and from vestibular and palatine regions in cross sections x120. Afterwards, each hemi section was grinded 2mm and the steps of the technique were repeated for new observation. In longitudinal sections enamel with Hunter Schreger bands (HSB) was identified in the incisal, medial and cervical thirds, occupying the thickest portion of the enamel in the last two, which then decreased towards the proximal faces. In cross sections, shifts in prism orientation that do not present the typical band aspect were seen. In longitudinal and cross sections radial enamel was found on both the outer and inner surfaces. HSB enamel is the most representative in the tooth group studied and is a specialization of microstructure to resist masticatory forces.

114.

CONSUMPTION OF EDULCORANTS AND INDEX OF LEARNING IN AN EXPERIMENTAL MODEL

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Introduction: Many factors exist that influence in the acquisition and the recovery of the received sensorial information. **Objective:** To analyze the possible effect of the consumption of edulcorants in the development of cognitive capacities from the analysis of a learning index. **Material and Methods:** 3 groups of 10 mice of the stock Balb c, endobring, of 3 months, exposed to a same diet and different drinks H2O and sweetened drinks H2O with Saccharine or Stevia rebaudiana were studied. The space memory by means of the aquatic labyrinth of Morris studied (proximal and distal version). Two stages were considered: 1) acquisition (EA) and 2) recovery (ER). The Index of Learning was defined like: IAP=(1-TE/TM) with used time =TE and TM= maximum time allowed (180seg (EA); 120seg (ER)). We consider : 0<IAP<0,40 (Poor Learning); 0,40<IAP<0,60 (Regular learning); 0,60<IAP<0,80 (Good learning) and IAP≥ 0,80 (Excellent learning). Descriptive exploratory statistical techniques were applied. **Results:** Great within-group variability (IAP). In average they presented/displayed a Level of poor or regular learning, being G3 the one of better performance. **Discussion:** we observed a nonawaited effect; apparently the long term consumption of Stevia R. improves the cognitive capacities of the mouse.

115.

MICROANATOMY OF THE *Moina eugeniae* (CRUSTACEA, BRANQUIPODA, CLADOCERA)

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Cladocera live in continental waters. They're used in aquaculture as food-fish or food for other crustaceans. They have reproductive cycles characterized by partenogenesis. *Moina eugeniae* is the most abundant species of the zooplankton in southwest ponds from Buenos Aires province. The aim of this work is to study the histology of the cladocera by light microscopy in order to improve the anatomy and histology of this species. Samples were taken from the Calderón pond, next Bahía Blanca city (Buenos Aires province). Partenogenic females were fixed in formalin 4%. Tissue samples were cut 3μ thick and stained with haematoxylin and eosin. The gut showed a simple cuboidal epithelium with cells with apical villi and 1 or 2 nucleoli within the nucleous. Neuronal tissue and compound eyes were observed. The ovary was formed by fully vitellogenetic eggs. The appendages of the trunk, antennae and antennules had skeletal muscle. This is the first histological contribution for this species.

116.

COMPARATIVE ASSAYS OF CYTOTOXICITY INDUCED BY EXTRACTS OF *Verbascum thapsus*

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Introduction: The biotechnological application of *V. thapsus* in the phytomedicinal field is promissory to control the microbial infections, which demands to study its selectivity of action. **Objective:** To analyze the cytotoxic effect of 5 vegetal extracts (VE) obtained from *Verbascum thapsus* (Ambay). The extracts: n-hexanic (HE), chloroformic (CE), methanolic (ME), cold and warms aqueous extract (CAE and WAE), were obtained from leaves of the plant, by continuous and sequential agitation. The toxicity assays were carried out in plates of 96 wells; 0.2 mL/well of different concentrations of each VE [10-4000 μg/ml] were seeded on monolayer Vero cells. Controls wells without VE were included. The system was incubated to 37°C by 48 hs. The Maximum non Cytotoxic Concentration (MCNC) was determinate by daily examination at the optical microscopic. The CC50 was calculated by neutral red uptake method (NRU). Statistical analysis of lineal regression was made, for a coefficient of determination 0.90 (software Origin 7,5). Although by both techniques the toxic concentrations surpassed 1mg/ml, NRU showed to be more sensitive to evaluate toxicity on eukaryotic cells. A toxicity order was established: WAE < ME < CAE < HE < CE. Data stimulate further bioactivities studies with these extracts with the security that they not exerting cellular damage.

121.**IMMUNOHISTOCHEMICAL AND MORPHOMETRIC STUDY OF PITUITARY PARS DISTALIS THYROTROPHS OF MALE VISCACHA***Filippa V, Mohamed F.**Proyecto 22/Q603, Fac Qca, Bioqca y Fcia, Univ Nac San Luis CONICET. E-mail: vpfilipp@unsl.edu.ar*

The viscacha thyrotrophs (TSH cells) were immunohistochemically identified, and the morphometric parameters which are cellular activity indicators: immunopositive percentage area (%IA), cells percentage in PD (%PDC), number of cells per reference area (N°cell/RA), cell density (CD), cellular area (CA), and major cellular and nuclear diameters) were analyzed. Adult male viscachas captured in their natural habitat during the year, melatonin-administered and castrated were used. The thyrotrophs were localized in the ventro-medial sector, mainly in the PD cephalic extreme. They were oval and pyramidal, and their immunostaining intensity was heterogeneous. %IA, %PDC, N°cell/RA, CD, and CA exhibited a significant decrease in June-July in relation to February-March, and they were recovered in August-September. No morphometric variations of TSH cells were observed in melatonin-treated animals whereas a decrease of the %IA, %PDC, N°cell/RA, CD, was observed in castrated animals in relation to the intact animals. Our results show TSH cells morphometric variations along the year in agreement with the animal different physiological conditions during the reproductive cycle, and probably in response to the environmental signals changes. Melatonin does not have direct effect on the TSH cells. Castration modifies some thyrotrophs morphometric parameters, reinforcing the hypothesis that androgens affect the cells activity, as reported in other species.

122.**OXIDATIVE SUBSTRATES USE INVOLVED IN BOVINE SPERM CAPACITATION***Florentín A, Pérez Aguirreburualde MS, Fernández S, Córdoba M. Área de Química Biológica. Fac. de Cs Veterinarias. UBA. E-mail: mcordoba@fvet.uba.ar*

In bovine spermatozoa heparin (H) induces capacitation and lactate dehydrogenase activity decrease. The aim was to evaluate heparin capacitation through lactate (L) concentration variation in the medium and cellular respiration with different oxidative substrates. L and pyruvate (P) were used as oxidative substrates. Capacitation was evaluated by chlortetracycline and the viability by trypan blue stain. Data were analyzed by ANOVA and Tukey test ($P<0.05$). In heparin capacitated spermatozoa lactate concentration decrease in incubation media only with P/L ($0.36\pm0.03\times10^6$ mM/sp) respect to control at zero time ($0.49\pm0.01\times10^6$ mM/sp). When P was used as unique oxidative substrate there was not L production. Oxygen uptake was increased in the presence of P/L ($P<0.05$) respect to other treatments. The major capacitation percentage was induced by P or P/L respect to the value registered with L alone. H required substrates to induce capacitation. Viability was similar in all treatments, decreasing in the absence of substrates ($P<0.05$). In H capacitated spermatozoa, substrates are mainly sent to oxidative pathways dependent to mitochondrial function.

123.**ANIMAL MODEL OF OVALBUMIN ALLERGY: IMMUNE RESPONSE IN BRONCHIAL MUCOSA***Fontanella GH, Bassan ND, Vinuesa MA.**Cátedra de Histología y Embriología. Facultad de Ciencias Médicas. Universidad Nacional de Rosario.*

The functional interface between respiratory and gastrointestinal mucosa was recognized. In previous works we demonstrated that subcutaneous ovalbumin (OVA) administration produces high specific anti-OVA IgE serum titers and increases in the number of lymphocytes, mast cells and eosinophils in all the mucosa surfaces studied. To proceed with this study, we have now analyzed quantitative modifications of CD25 and class II positive cells (activation markers) and eosinophils from bronchial mucosa in response to oral (OC) or inhalatory (IC) challenge. Adult male New Zealand rabbits were divided in 4 groups (n=6, each), G1: control, G2: sensitized, G3: sensitized and OC G4: sensitized and IC. Samples were obtained 18 (IC) and 24 hours (OC) after challenge and processed for HE, cromotropo IIR (eosinophils) and immunohistochemistry (CD25 and class II). CD25 and class II positive cells were increased significantly in G3 and G4 respect the G1 and G2 recipient rabbits ($p<0.005$ vs. G1 and G2). The same was true when analyzing the eosinophil population ($p<0.005$ G4 vs. G1 and G2). The OC produced an increase of the activation cell markers expression from bronchial mucosa supporting the hypothesis of which the allergic illness involves a systemic immune response.

124.**THE SUPERIOR OVARIAN NERVE REGULATES THE STEROIDOGENIC ABILITY OF MACROPHAGES ON THE RAT OVARY***Forneris M, Davicino R, Micalizzi B, Oliveros L.**Departamento Bqca. y Cs. Biológicas. Facultad Qca., Bqca. y Farmacia. UNSL, 5700-San Luis. E-mail: mforne@unsl.edu.ar*

We have previously shown that ovarian steroidogenesis is modified by fluid from splenocyte cultures after Superior Ovarian Nerve section (SON-s). Also, we know that ovary and spleen are innervated by the SON through the celiac ganglion. Now, we study the effect of macrophage (Mφ) secretions on ovarian progesterone (P) and androstenedione (A) release of adult rats at diestrus 2. Splenic Mφ from rats with the SON sectioned 7 days before sacrifice and from control rats were cultured 24 h in DMEM. The supernatant of their culture liquids were used to stimulate control ovaries incubated for 3 h in metabolic bath to measure the P and A release (by RIA). Secretions of SON-s Mφ decreased the A release, without change in P. In these Mφ, the phagocytic activity (zymosan/NBT) did not change, but the lysosomal enzyme activity (alkaline phosphatase) decreased compared with control Mφ. The nitric oxide (NO) production (measured by Griess reaction) and that of TNF-alpha (by ELISA) decreased in SON-s Mφ in relation to control Mφ. This neuroimmuno-modulation at peripheral level mediated by the SON section modifies the lysosomal enzyme activity of splenic Mφ and their production of both, NO and TNF-alpha, which are involved in the ovarian steroidogenesis.

125.**MINIMUM INHIBITORY CONCENTRATION OF LEVOFLOXACINE ON ENTEROPATHOGENIC STRAINS OBTAINED FROM ANIMALS**

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Introduction: Levofloxacin (LFX) is a wide spectrum antimicrobial. Its human therapeutic properties and the kinetic in animals justify the study of its activity on pathogenic strains isolated from domestic animals. **Objective:** Determination of antimicrobial activity of LFX on strains isolated from domestic animals. **Materials and methods:** The Minimum Inhibitory Concentration (MIC) was performed by the agar dilution method proposed by the NCCLS against *E. coli* (N=14) and *Salmonella sp* (N=8) isolated of clinical cases of pecuaria establishments of the south of Córdoba. It used a reference strain of *E. coli* ATCC 25922. Concentrations ranging from 6,25 to 0,02 μ g/ml of LFX were incorporated to Mueller-Hinton agar. The bacterial density was 0,5 of McFarland scale and it was inoculated by a Steers-Foltz multi-inoculator. Plates were incubated 24 h, at 37°C. The strains able to growth at concentrations equal or bigger than 2 μ g/ml were considered resistant. **Results and discussion:** All *Salmonella sp*. and twelve *E. coli* strains tested were sensible to LFX while two *E. coli* strains were resistant. These results show that LFX is a therapeutic alternative to treat animal patologies caused by enterobacteria since MIC values are similar to the reported for human and lower to plasmatic and tissue levels of LFX in animals.

126.**CHARACTERIZATION OF THE LIPID TRANSFER TO ENTEROCYTES IN PANSTRONGYLUS MEGISTUS (HEMIPTERA: REDUVIIDAE)**

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In insects, the transfer of hemolymph lipids to tissues is a remarkable event accomplished by lipophorin (Lp), the major insect lipoprotein. This process allows the formation of essential lipid reserves, which in turn will be used in many physiological events. In this work, we have analyzed the mechanisms involved in the transfer of lipids from Lp to the midgut of *P. megistus*, an important vector of Chagas' disease. Lp was isolated from hemolymph of fifth instar nymphs by a KBr gradient. The enterocyte membranes were obtained by ultracentrifugation of midgut homogenates. Lp interaction with midgut membranes was characterized by solid-phase binding assays. Lp binding sites in enterocytes were localized by direct immunofluorescence. Lp-binding proteins at the membranes were studied by ligand blotting assays and visualized by western blotting. Results showed: [a] a clear dependence of the Lp binding to membranes with the pH. Lp binding also showed a kinetic of saturation compatible with a model for a single binding site, [b] the interaction of Lp with midgut cells mainly occurred at their basolateral region, which *in vivo* contacts with hemolymph; [c] at the membrane, Lp mainly interacted with a protein of 71 kDa. Taken together, these results increase our understanding about the metabolic pathways of lipids in insects as well as the physiological processes occurring in these vectors of Chagas' disease.

127.**METABOLIC SYNDROME IN ADULT HYPERTENSION PATIENTS FROM SAN LUIS**

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The aim of the study was to study the characteristics and frequency of the metabolic syndrome (MS) in adult hypertension population from San Luis. The study included patients from private cardiological institute during one year (2006). The MS is defined using the criteria proposed by ATP III (Adult Treatment Panel III) being included as MS variables: systolic and diastolic blood pressure and seric levels of glucose (G), triglycerides (TG) and HDL-cholesterol (HDL-c) and others. Patients were considered as having the MS when four or more characteristics showed abnormal values. From 280 patients, the 67.5% was female (50% >50 age-old) and 31.4% was male (19.6% >50 age-old). The HTA patients represented the 78.1% and without HTA 21.8%. HTA (66%) between 50-65 age-old, the most female (71.3%), and MS patients 12.1% (64.7% female). We consider HTA with systolic blood pressure \geq 130 mmHg and diastolic \geq 85 mmHg. The TG (mg/dl): HTA (156.4 \pm 5.6), MS (223.6 \pm 16.1) and control (95.8 \pm 5.3). HDLc (mg/dl): HTA (43.3 \pm 0.7), MS (39.3 \pm 0.9) and control (48.6 \pm 1.8). G(mg/dl): HTA (94.7 \pm 1.0), MS (110.2 \pm 4.2) and control (91.8 \pm 1.1). The statistical analysis of data shown significant differences (ANOVA, P<0.001). In conclusion, the high-risk nature and growing prevalence of the metabolic syndrome highlights the need to identify patients with these conditions and to treat them with a multitargeted approach therapy that includes blood pressure control.

128.**VARIABILITY IN THE COMPOSITION OF ESSENTIAL OILS OF ALOYSIA TRIPHyllA**

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Introduction: The essential oils (EOs) of *A. triphylla* showed differences in their terpenic composition. These differences determine chemotypes, which are defined by the content of neral, geranial, limonene, sabinene and transthujone. These reasons justify the study of the best conditions to cultivate this vegetable. **Objetive:** to study the chemical composition of the EOs of *A. triphylla* collected in different dates and places. **Materials and methods:** The EOs were obtained by hydrodistillation. The identification and quantification of the EOs were performed by gaseous chromatography and mass spectrometry. **Results and discussion:** the average yields in EOs obtaining was 0,43% (w/v). The main terpenic components identified were limonene, neral, geranial, spathulenol and cariophyllene oxide. There were variations in amount of the common and minor components. There could be defined four chemotypes: I: Río 1°, II: Las Viñas, III: Salta, IV: La Paz, Paraguay, Mendoza, San Luis and Experimental Bs. As. The proportion between the main components was proposed as a valid criterion for the identification of chemotypes. The EOs belonging to the same genotype and chemotype obtained in different dates from La Paz (Cba), showed differences in the proportion citral:limonene. So for *A. triphylla* it would not be a valid criterion to define chemotypes.

129.**C-KIT POSITIVE CELLS IN THE PORCINE STOMACH***Galotta JM¹, Márquez SG¹, Portianky EL², Barbeito CG³.*¹*F.Cs. Agrarias UCA; ²Instituto de Patología, ³Histología F.Cs. Veterinarias UNLP. E-mail: jorge_galotta@yahoo.com.ar*

C-kit is a stem cells growth factor receptor. It is expressed in the interstitial cells of Cajal (ICC) of the muscular layer of the gastrointestinal tract. Here we analyze the presence, distribution and morphometric parameters of c-kit positive cells in the stomach of pigs. Samples were obtained from the pyloric portion of the stomach of six months-old healthy pigs at abattoir. Tissues were processed for paraffin embedding. Immunohistochemistry was performed using the anti-c-kit CD 117 monoclonal antibody. LSAB® (DAKO) was applied as a detection method; diaminobencidine was used as a chromogen and haematoxylin was used for counterstaining. Morphometry was performed using digital image analysis (ImagePro Plus 5.1 - Media Cybernetics). Methylene blue staining also revealed the presence of mast cells between the muscle fibres. c-kit positive cells were found in the muscular wall of the stomach, in close contact with neurons of the myoenteric plexus. Cells had a fusiform aspect showing fine prolongations. The morphometric analysis data reported an area of $27.03 \pm 7.21 \mu\text{m}^2$; major axis: $9.76 \pm 2.31 \mu\text{m}$ and minor axis: $3.68 \pm 0.70 \mu\text{m}$. Immunohistochemistry, anatomical location and morphometric data led us to identify them as ICC.

130.**DEVELOPMENT OF THE MALE AND FEMALE GENITAL SYSTEM IN *Pomacea canaliculata* (AMPULLARIIDAE)***Gamarra-Luques C, Giraud-Billoud M, Castro-Vazquez A.**Laboratorio de Fisiología (IHEM-CONICET), FCM-UNCuyo, Mendoza. E-mail: cgamarra@fcm.unco.edu.ar*

The ampullariid genital system is formed by two sets of structures: (1) the gonoduct; and (2) the primordial copulatory apparatus, internally attached to the right mantle edge, and which is present in both sexes. How these primordia differentiate post-hatching in either the male or female structures (including the gonads) is unknown. Newly hatched (2 mm) juveniles have a sexually undifferentiated gonoduct and no copulatory primordium. The latter appears in 5 mm juveniles, when it is of similar size in all individuals. The proximal (visceral) part of the gonoduct differentiates in either the testis or the ovary in 10 mm juveniles, while the copulatory primordium stays of similar size in both sexes. In the 15 mm stage, the copulatory apparatus is definitely larger in males, and shows all the adult components, though not yet fully developed. In this stage also, the distal (pallial) gonoduct has differentiated either in the prostate/seminal vesicle continuum or in the uterine gland and vagina. All the adult structures are present in 20 mm snails, but the copulatory apparatus remains vestigial in females. Full gonoductal development is reached in 25 mm snails of both sexes, but the copulatory structures remain vestigial in females.

131.**CHRONIC STRESS ASSOCIATED TO INFLAMMATORY MARKERS***Gambini A, Bensi N, Niebyski A, Gauna HF.**Fisiología Animal. UNRC. Río Cuarto. Córdoba. E-mail: hgauna@exa.unrc.edu.ar*

There are evidences that stress response is integrated by neuroendocrine system, been related with cytokines produced by inflammatory process. The aim of this work was to evaluate the blood levels of inflammatory markers under chronic stress conditions (sc). Adult male wistar rats were used with two different stressors: immobilization (imo) and tube glass restraint vs their respective no stressed controls. Days 1 and 14 of experimental period, blood samples were obtained immediately after stress. The fibrinogen blood levels of chronic imo rats were elevated (509 ± 45 vs. 377 ± 35 mg/dl – p=0.000825). In leucogramme: blood leucocytes showed a significant decrease in stressed rats (3625 ± 850 vs. 6512.5 ± 1000 cel/ml p=0.001895) with lymphopenic effects in two stressed models. A significant increase of levels of ggt was demonstrate in sc rats (25.5 ± 3 vs. 20.6 ± 1 ui/dl p=0.030110). The effects of stress on different variables, showed indicous on inflammatory process as stress response of animal in this type of stress, opening a new view point, and inducing new expectatives in prevention and treatments.

132.**OXIDATIVE STRESS INDUCED BY LOW TEMPERATURES IN *Digitaria eriantha* CV. MEJORADA INTA***Garbero M¹, Zirulnik F², Molina A², Pérez-Chaca V², Pedranzani H¹.*¹*Lab. Fis. Veg. FICES. UNSL. Villa Mercedes. San Luis. ²Lab. Qca. Biol. PROIPRO 2-0304. UNSL. San Luis.**E-mail: mgarbero@fices.unsl.edu.ar ; garberom@yahoo.com.ar*

Digitaria eriantha cv. Sudafricana introduced in Argentine and *Digitaria eriantha* cv. Mejorada INTA diminished their parameters of production at 4°C. Biochemical studies in cultivate native Sudafricana showed variations in the antioxidant defense system and increase the thiobarbituric acid-reactive substances (TBARS). The aim of this study was to evaluate the effect of cold on oxidative stress parameters in *Digitaria eriantha* cv. Mejorada INTA. There were used plants with 10 cm of height, at 4°C for periods of 0 (control), 6, 24 and 72 h. Non-protein thiols (NPT), reduced glutathione (GSH), phytochelatins (PC) and TBARS were measured as parameters of antioxidant defense and oxidative stress. A significant increase of NPT levels at 24 and 72 h respect to control plants was observed. The contents of GSH and PC diminished slightly at 6 h of treatment and then they increased at 24 and 72 h, without significant variations. TBARS level did not change in any treatment. The increase of non-protein thiols would indicate activation of the non-enzymatic antioxidant system that protects the cells against the oxidative damage produced by low temperatures.

133.**GENETIC ORIGIN COMPARISON THROUGH MORPHOMETRY IN *Apis Mellifera***

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The objective of the Genetic Improvement Program (MEGA) from the Integrated Project of Apicultural development (PROAPI) is to select honeybees adapted to every region in the country. Morphometric analysis, together with other tools, is employed to characterize the genetic materials. The differences found among ecotypes of the country encouraged this work, whose objective was to use morphometric variables to compare colonies from different genetic origins. All the colonies were installed in an apiary from Rafaela, Santa Fe. Some packages from Mendoza (G1); others from Tucumán (G2) which employ PROAPI genetics, and another group of unidentified origin (G3), were compared to a population located in Balcarce (PROAPI) (G4). Eleven morphometric variables were measured: length and width of forewing and hind wing, different angles and length of wing veins, tibia, femur and metatarsi length, metatarsi width and proboscis length. A multifactorial distance analysis using euclidean distances showed that G1 resulted different from G4 ($P<0.05$); while G2 and G3 did not differ from G4 ($P>0.05$). Morphometric analysis is a tool that complements behavioural characterization and ecotype genetics. The differences between the colonies of a same genetic origin, multiplied in different environments would indicate an environmental effect over the morphometric variables.

134.**PROLIFERATIVE ACTIVITY IN HEPATOCYTES AND RENOCYTES OBTAINED FROM WEANING MICE INJECTED WITH TUMORAL CELLS**

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The presence of injected tumors modifies the intensity and temporal distribution of DNA synthesis (DNAs), and the mitotic activity (MA) of cellular populations in tumor bearing mice. The purpose of the present study was to analyze the effect of the SS1K hepatocellular carcinoma cells injection on, DNAs and MA of hepatocytes and renocytes. The mice (21 days old) were divided in two groups: 1) Control, and 2) Mice injected with 0.2 ml of tumoral suspension on the day 15 of life. The sacrifice of the animals was performed on the day 21 at 00:00 and 16:00 hours in both groups. The samples were processed as follows: one half with the immunohistochemistry technique using bromodeoxyuridine, and the other half with statmokinetic method. The DNAs and the MA indexes were determined for each animal studied and the $X \pm SE$ was established either for each lot and group. The data were analyzed with t Student Test. The results showed that, hepatocytes and renocytes, DNAs indexes were significantly lower in Group 2 than in the Control group (Group 1). It could be argue that the presence of carcinoma alters the proliferation process in the cellular populations studied. However, further investigations are necessary to study the complete circadian cycle to achieve a definitive conclusion.

135.**NOREPINEPHRINE MODIFIES THE EFFECT OF VIP AND NPY IN CELIAC GANGLION ON THE OVARIAN PROGESTERONE AND NITRIC OXIDE RELEASE**

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We have previously shown in Holtzman rat at Diestrus 2 that stimulation of celiac ganglion (CG) with Vasoactive Intestinal Peptide (VIP) or NPY in the ex vivo integrated system CG-superior ovarian nerve(SON)-ovary (O) incubated in Krebs Ringer buffer in a metabolic bath stimulates the progesterone (P) release from the ovary. In this work we study, in the same system, whether the addition of 10^{-6} M norepinephrine plus 50 ng/ml of VIP or NPY on the CG modify the ovarian P and nitric oxide (NO) release. Samples from the ovarian cuvette were taken at 30, 60, 120 and 180 min of incubation. Basal values were obtained without neuropeptides addition in GC. Statistical analysis by ANOVA, one way. The NE reversed the P increase induced by VIP or NPY alone. VIP decreased the NO release, and NPY did it not modify. However, the addition of NE together with each neuropeptide, increased the ON release from de O. NE regulates the ovarian P release in an inhibitory way, associated to an increase of NO.

136.**CHARACTERIZATION OF POLYPHENOLOXIDASE OF SETEMBER PEACH GROWN IN CUYO'S REGION**

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The enzyme polyphenoloxidase (PPO) of fruits catalyzes reactions involving phenolic compounds and molecular oxygen to produce brown pigments, decreasing the quality of fresh fruits, juices and other products. The objective of this work is to characterize and to control the PPO activity of Setember peach using a spectrophotometric method. Was defined one unit of PPO activity (U) as an increase in absorbance of 0.001 (408 nm) per minute and per millilitre of enzyme. At pH 6.5 (pH-activity optimum) and 25°C PPO showed activity using the following substrates: catechol: $K_m=2.51$ mM, $V_{max}=416$ U/min. mL E; 4-methyl catechol: $K_m=3.63$ mM, $V_{max}=506$ U/min.mL E and pyrogallol: $K_m=0.214$ mM, $V_{max}=225$ U/min. mL E. Thermal stability of peach PPO was investigated at various constant temperatures from 25°C to 60°C. After 10 minutes of incubation between 30°C and 60°C, the residual percentage activity decreased gradually from 85% to 0%, respectively. Heating PPO at 60°C for 5 minutes the residual activity is less than 5%. Ascorbic acid ($I_{50\%}=0.296$ mM) and sodium metabisulfite ($I_{50\%}=0.234$ mM) were evaluated as inhibitors. It can be concluded that PPO activity from Setember peach is sensitive to the temperature and to assayed inhibitors.

141.**BRANCHING PATTERN AND REPRODUCTIVE STRUCTURES IN PEANUT GENOTYPES***Giayetto O, Bessone A, Cerioni GA, Fernandez EM.**Department of Plant Production. Faculty of Agronomy and Veterinary. UNRC, Río Cuarto. Córdoba.**E-mail: ogiayetto@ayv.unrc.edu.ar*

Peanut genotypes (*Arachis hypogaea* L.) are characterized by different branching patterns and growth habit. The objective was to evaluate the relationship among branching pattern and reproductive structures distribution of different peanut genotypes. A study in a field conditions was carried out in 2005/06 under non-limited water conditions. Three genotypes were included: (1) Tegua, procumbent habit and alternate branching pattern; (2) LF 36, spreading bunch habit and alternate branching pattern; and (3) LF 19, erect habit and sequential branching pattern. At harvest time pods number and weight and mature seeds weight of each branches type per plant were quantified. Genotype (3) presented pod in main stem. Most number (68-77%) and weight (72-78%) of mature pods and seeds weight (73-78%) per plant were concentrated in cotyledonary and n+1 branch in all genotypes, but mainly in LF 36. This genotype had high percentage of seed >9 and 10 mm. Tegua and LF 36 had the most number and weight of reproductive structures in the periphery of plant following their own branching pattern. Meanwhile LF 19 doesn't have its reproductive structures near main stem and it had the highest immature pod percentage.

143.**THE PENIAL SHEATH OF *Pomacea canaliculata* (AMPULLARIIDAE). I. THE OUTER GLAND***Giraud-Billoud M, Gamarra-Luques C, Castro-Vazquez A.**Laboratorio de Fisiología (IHEM-CONICET) y Departamento de Morfología y Fisiología (FCM-UNCuyo).**E-mail: mgiraudbilloud@gmail.com*

The penial sheath of this snail is a complex structure that includes three glands, with elements probably involved in hormone secretion. We studied here the most conspicuous of them, the so-called "outer gland". It is orange-yellowish and is located at the sheath's base. Its thick secretory ducts open in a depression located at the front of the sheath's insertion on the mantle edge. Abundant mucus is secreted at this point during copulation. The glands' internal ducts of the gland, and the tubular alveoli in which these end, are lined by a complex cylindrical epithelium, in which goblet cells, ciliated cells and cylindrical secretory cells (loaded with thick granules) are interspersed. The basal part of many of the granular cells, as well as some of the goblet cells, protrude towards the submucosa forming a deep stratum which appears sub-epithelial, but whose cells, with their deeply located nuclei, are indeed above the basal membrane and have their apices attaining the lumina of secretory ducts. These granular cells have mitochondria with tubular cristae, and Reinke's type crystalloids, which are also found in esteroidogenic cells in chordates. The latter crystalloids are embedded in the microgranular matrix of the large granules.

142.**PHOSPHATIDYLINOSITOL- 3 KINASE ACTIVITY IN *T. cruzi* EPIMASTIGOTE FORMS***Gimenez AM, Gesumaria MC, Machado EE.**Dpto. Biología Molecular, FCEFQyN, UNRC.**E-mail: mgimenez@exa.unrc.edu.ar*

Phosphatidylinositol (PI) metabolism through phosphatidylinositol kinases (PI-K) activity plays a central role in different signaling pathways. In particular PI(3) phosphate (PI3P), the product of PI-3 kinase, is implicated in control of several cellular processes, including membrane trafficking, autophagy and cell signaling. The aim of this work was determinate the presence and activity of PI-3K in *Trypanosoma cruzi* epimastigote forms. Bioinformatic and phylogenetic analysis showed that parasite genome codificate at least two putative proteins with conserved domains of PI-3K. One of them, TcVps34p, would be homologue to yeast class III PI-3K, Vps34p, and the other protein shares high homology with class I PI-3Ks. Western Blot analysis on epimastigote membranes using an antibody against catalytic subunit of human PI-3K, yielded a single band of 110 kDa. This MW is in concordance with MW predicted for both putative proteins of *T. cruzi* after bioinformatic study and it is similar to PI-3Ks of other organisms. PI-3K activity was assayed using endogenous lipids as substrates for kinase assay with [γ^{32} -P] ATP and developed in TLC-borate system. Treatment of epimastigotes with wortmannin and LY294002 (PI-3K inhibitors in mammalian cells) decreased enzymatic activity in approx. 40%. These results suggest that *T. cruzi* epimastigotes express a functional protein with activity of PI-3K.

144.**THE PENIAL SHEATH OF *Pomacea canaliculata* (AMPULLARIIDAE). II. PROXIMAL AND DISTAL GLANDS***Giraud-Billoud M, Gamarra-Luques C, Castro-Vazquez A.**Laboratorio de Fisiología (IHEM-CONICET) y Departamento de Morfología y Fisiología (FCM-UNCuyo).**E-mail: mgiraudbilloud@gmail.com*

The proximal and distal glands are serial sets of mucosal plies, located on the dorsal aspect of the penial sheath. The proximal gland is located deep in the sheath's groove, while the distal one is located near the sheath's tip. Secretory ducts are absent in both glands. The surface mucosa is composed by a cylindrical epithelium crowned with branched microvilli, and with intermingled goblet mucous cells. Abundant mitochondria occur in the two basal thirds of cylindrical cells, whereas their apical region is full of vesicles. Cells with nuclei showing heavy chromatin clumps attached to the nuclear membrane, and cytoplasmic extensions (devoid of organelles), are placed underneath the surface epithelium and demarcate a labyrinth of extracellular spaces. Even deeper, epithelial "columns" are found, perpendicularly oriented to the surface mucosa. Their epithelial cells bear large granules which appear to contain lipids and proteins. Hollow spaces are found within these columns, reminding the pseudofollicles of the vertebrate adenohypophysis. The complex structures of these glands defy our comprehension of their proposed paracrine (or endocrine?) role on trophism of the male system.

145.**THE INFLUENCE OF TILLING AND NITROGEN DOSES IN THE NUMBER OF WHEAT STEMS**

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Wheat yield is related to its tillering capacity and to the fertility level of the productive lot. The objective of this work is to assess the increasing number of wheat stems cultivated with different tilling systems and nitrogen levels. The experiment was carried out in the Villarino Experiment Station in Zavalla town, province of Santa Fe (60° 53' West longitude, 33° 01' South latitude). An Vertic Argiudol soil in a soybean-wheat/late soybean – corn rotating sequence was used. A long cycle cultivar was sown on May 23th, 2005 using vertical tilling (VT) and no-tillage (NT) systems. Randomized completed block design with four VT and NT replications were arranged. The three-level nitrogen fertilization treatments evaluated were: N₀; control, N₆₀; 60 kg N ha⁻¹ and N₁₂₀; 120 kg N ha⁻¹. The 0,50 m² samplings were extracted at 1,3, 2,4, 2,5, 3,2, 3,6 and 6,9 stages according Zadok's scale (1974) from every experimental unit. No tilling doses interaction was observed. Significant treatment differences ($p<0,01$) were found beyond 2,5 stage with both tilling systems, having N₁₂₀ the highest values. Fertilization increased the stem production per m⁻² regardless the tilling system used.

146.**VITAMIN E PREVENTS THE DAMAGE INDUCED BY THE HERBICIDE DICAMBA AND ITS COMMERCIAL FORMULATION BANVEL®**

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The herbicide dicamba and its commercial formulation banvel® have genotoxic potential on different cellular types, among them, the Chinese hamster ovary cell line (CHO-K1). With the objective of determining the possible mechanism of action by which these compounds exert their cyto- and genotoxic effect, CHO cell were treated with 0, 50 or 500 µg/ml of either pesticide, in presence or absence of α-tocopherol (vitamin E, 50 µg/ml). Induced damage quantification was measured by the analysis of sister chromatid exchanges (SCEs) and cell-cycle progression assays. Statistical analysis was performed by the χ^2 and Student's *t* tests. Results showed that both test compounds induced a significant SCEs increase ($P<0,01$) and a delay in cell-cycle progression at 500 µg/ml dose ($P<0,01$). Nevertheless, vitamin E presence inhibited the cyto- and genotoxic effect of dicamba and banvel® ($P<0,001$). These findings would suggest that given the protection exerted by vitamin E on the deleterious effect produced by these herbicides, the damage would be induced by reactive oxygen species liberation.

147.**DISTRIBUTION OF FIBER TRANSVERSAL SECTIONS IN SEMITENDINOSUS MUSCLE SUBVOLUMENS OF THE PIG**

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The references about the existence of neuromuscular compartments permits to infer the presence of labor partitioning inside the whole muscle. Fiber populations with homogeneous physiological characteristics as types, area, and metabolism were found in these muscle subvolumens or compartments. Our aim has been to determine the variations of fiber areas inside the compartments of the semitendinosus muscle previously described. From 48 slaughtered pigs reared in an outdoor system, were identified by histochemistry I, IIA and IIX/B fiber types in each subvolumen of the semitendinosus muscle (R1, R2, R3, R4). There were not differences in area among fiber types in the four regions. The area arrangement for all fiber types was R1 > R2 = R3 = R4 ($p<0,05$). The increase of fiber area is an unfavorable characteristic in pork selection toward lean carcass of fast growth. We conclude that R1 subvolumen has undesirable characteristics to produce high quality meat assignable to the greater significant section, related to their smaller oxidative capacity determined in a prior research. This experiment could serve to future investigations in meat technology and indicates the importance to consider previously the compartments of the muscle.

148.**FIBER POPULATIONS IN THE SEMITENDINOSUS MUSCLE OF THE PIG**

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Inside a muscle there are subvolumens integrated by fibers with capacity for shortening or isometric contraction. Fiber type does more efficient subvolumen function, as maximum shortening velocity and metabolic capacity. The aim of this investigation was to know the fiber populations of the compartments (R1, R2, R3 and R4) previously determined, for applying in husbandry studies. Fibers types were identified by the adenosine triphosphatase myofibrillar enzyme activity in samples obtained from each subvolumen. I, IIA and IIX/B fiber types distribution was R4=R1>R2=R3; R4>R1=R2=R3; R3>R1=R4 and R3=R2 ($\chi^2 \alpha =0,05$). These results indicate a labor partitioning among medial R1 and R4 subvolumens with regard to R2 and R3 subvolumens and their aptitude to turn into high quality meat. This transformation is obtained in muscles which are composed mainly by fast oxidative fibers. The variation among the subvolumens indicates the importance to carry out a prior structural study for determining meat quality parameters.

149.**INTERSPECIFIC DIFFERENCES BETWEEN *Odontophrynus cordobae* AND *O. americanus* (ANURA: LEPTODACTYLIDAE)**
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The occurrence of natural polyploid bisexual species in amphibians provides evidences that the polyploidy is an important evolution mechanism in several families of anurans. *O. cordobae* (diploid) and *O. americanus* (tetraploid) individuals of nine localities of Córdoba province in central area of Argentina were analyzed. Two complexes of independent characters, ethological (bioacoustics) and cellular (erythrometry and cytogenetic) were used to evaluate the interspecific differences. Ploidy of the individuals was analyzed by counting the chromosomes, and the karyotype corresponding to each locality was performed. Secondary constrictions were observed in the pair 4 in diploid individuals and in the quartet 11 in tetraploids. The means of erythrocyte areas obtained for *O. cordobae* were between 167.67 and 172.23 μm^2 and for *O. americanus* were between 222.23-226.63 μm^2 . The acoustic analysis showed significant differences among species with an evident segregation of the syntopic diploid/tetraploid populations. The use of a group of independent characters is useful for the study of related species populations. Until the present, natural hybrids have not been found. The differences suggest that the call components represent an important prezygotic isolation barrier.

151.**DETERMINATION OF SIALIC ACID (AcS) IN INDIVIDUALS WITH HIGH ARSENIC LEVELS (As) IN THEIR DRINKING WATER CONSUMPTION**

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Tumoural cells show changes in the glycosylation process which is manifested in an increase of sialoproteins on the surface of tumoural cells. Different studies have associated a greater incidence of different forms of cancer in individuals exposed to drinking water containing high As levels. The aim of this study was to determine the levels of serum AcS in individuals who drink As contaminated water (levels higher than 0,05 mg/L). 97 individuals divided into two groups were studied : Group A: 68 people in a control group, and Group B: 29 people who drank drinking water containing As levels higher than 0,05mg/L. All of them were punctured on their vein and the serum was separated by spontaneous clotting and frozen up to - 20° until it was processed. The serum AcS dosage was done using the modified Warren colourmetric method. The average concentration \pm standard deviation was of 680 ± 148 mg/L for group A, and 795 ± 196 mg/L for group B, a statistically significant difference. These preliminary data show that people with high As level intake would have an increased sialoprotein replacement, which could be an early marker of the alterations caused by this carcinogen.

150.**INITIAL STEPS FOR A MICROBIOLOGIC STUDY OF RUMEN IN SMALL RUMINANT**

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We began a microbiologic study about microbiotic ruminal in small ruminant of Mendoza's zoo. We used adult Somalia's sheep and goats. The animals were previously vaccinated against Mancha, Gangrene and Tetanus and were treated with antibiotics and antihelmintics. The animals were uninformed 24 hours of food and 12 hours of liquid. Then, we made the surgery in Veterinary School of Maza University. This procedure is a rumenotomy, with application of intraruminal cannula. This dispository was made with PVC (polivinylchloride), sealed in the soft parts with rubber rings, that prevents the ebb tide. The animals were placed in a closed and clean place, for special cares, mainly for disinfection of the injury until healing and final adaptation of the cannula. A chronic no inflammatory fistula, with an animal of normal behavior, allows us to follow the next step: the application of an own design dispository to take aseptic samples, based in hydraulics forces. We compared thus results obtained with other procedures. The first isolation was made in a Tarozzi agar and 4% agar with an anaerobic atmosphere. Colonies will be picked in a slob tube with 2% agar. Then, we differentiated aerobic from anaerobic bacterium and made biochemical tests and the quantification of organic acid fermentation products, established by gaseous chromatography. We also prepared RCGA (40% ruminal fluid, glucose, cellobiose, agar) medium, in which the ruminal fluids were included in a 1:3 proportion in medium, to replace the nutrients that originated the own ruminal bacterium. We will also use Med 10 medium, prepared with a mixture of volatile fatty acids, without ruminal fluid. Ketorolac (0.3mg/kg/day) was used after surgery to handle the pain for a week, with the best recovery.

152.**SINGLE AND MIXED INOCULATION OF ALFALFA WITH PLANT GROWTH PROMOTING RHIZOBACTERIA (PGPR)**

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The combined inoculation with rhizobia and PGPR produces a positive effect in the growth of legumes due to an increase in the absorption of water and nutrients by the roots, a higher nitrogenase activity and an improved general sanity of the plant. The objective of this work was to evaluate the effect of both single and mixed inoculation with rhizobacteria on the growth of alfalfa (*Medicago sativa*). Phosphate and iron solubilizing rhizobacteria, which inhibit the fungus *Macrophomina phaseolina*, were isolated from alfalfa rhizospheric soil, characterized and selected in order to carry out single and mixed inoculation assays (with *Sinorhizobium meliloti* B399). The evaluated growth parameters were: shoot and root length, shoot, root and nodules dry biomass, number of nodules and effectiveness. The obtained results were statistically processed with ANOVA and both DGC and LSD tests ($p < 0,05$). The selected isolates evidenced a beneficent effect upon the growth of alfalfa, not only reflected in co-inoculation assays, but in the inoculation without the symbiont as well; the capacities of nutrients solubilization and biocontrol, both evidenced in *in vitro* assays, are characteristics that presumably facilitate such promoting effect, along with other potential activities that should not be discarded, such as phytohormones and antibiotics production.

153.**ALLOMETRIC STUDY OF BODY SIZE, FATTY ACID COMPOSITION AND SENSITIVITY TO LIPID PEROXIDATION IN BRAIN MITOCHONDRIA AND MICROSOMES FROM DIFFERENT BIRDS**

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The objective of this study was to examine the relation between the corporal size, fatty acid (FA) composition and sensitivity to lipid peroxidation (LP) of mitochondria (MIT) and microsomes (MIC) isolated from brain of birds ranging in size from 10g manon to 5Kg goose. FA was determined using gas chromatography and LP was assessed by chemiluminescence. In MIT, the percentage of MUFA, principally C18:1 was lower in the greater species and showed a significant allometric decline with body weight ($r=0, 96$). In MIC, a significant increase of C18:2 ($P<0, 01$) and PUFA ($P<0,004$) in greater bird's species was detected. Only MIT of all birds examined suffered LP showing a significant decrease of C20:4 n6 and C22:6 n3 in peroxidized compared to control samples. However, LP did not change when body weight increased. The QL was statistically significant in MIT but not in MIC. The allometric analysis of the QL of both organelles was not statistically significant. These results show that the brain of all birds studied has a high content of C22: 6 n3 being similar to the one of other vertebrates, indicating a high specificity by this tissue.

154.**LIPID PEROXIDATION OF LIVER, HEART AND KIDNEY FROM LONGEVOUS AND SHORT-LIVED RODENTS**

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Chinchillas (*Chinchilla lanigera*) are longevous animals which posses low metabolic rate, whereas rat (*Rattus norvergicus*) are short-lived animals and posses high metabolic rate. Aim of this study was to determine lipid peroxidation (LP) sensitivity of liver, heart and kidney homogenates isolated from both species. LP was evaluated using chemiluminescence (CL) and fatty acid (FA) profiles by means of gas chromatography. CL indicated that the LP process was higher in homogenates from rat than those from chinchilla. The FA composition of total lipids isolated from liver and kidney homogenates from both species was substantially modified when subjected to LP, with a decrease of C20:4n6 and C22:6n3 acids. However, changes were not observed in the FA profiles of heart. The lack of a relationship between FA unsaturation and sensitivity to LP observed in heart, suggests that other factors may be involved in the protection to LP in this organ. Polyunsaturated / monounsaturated rate was higher in rat than chinchilla. This observation may explain the differences noted when CL of both species was compared. It can be concluded that the low composition of unsaturated FA observed in liver and kidney of chinchilla is responsible of the low sensibility to LP of this species.

155.**EFFECT IN LUNG OF *YERSINIA ENTEROCOLITICA* O:8 INFECTION IN WILD-TYPE AND IL-12P40 KNOCKOUT MICE**

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Yersinia enterocolitica O:8 is a pathogenic bacterium in human and lethal for mice. The objective of the work was to evaluate the effect in the lung of an oral infection with a sublethal dose of *Y. enterocolitica* O:8. Wild-type (WT) and IL-12p40 knockout (KO) mice were orally infected with 2×10^6 colony forming units (CFU) of *Y. enterocolitica* O:8. Three days after infection, the mice were sacrificed. The bacterial load was determined in the spleen (S), Peyer's patches (PP), mesenteric lymph nodes (MLN) and the lung (L). Cellular differentiation and viability were evaluated in bronqueoalveolar lavages (BAL). Nitric oxide (NO) in BAL was determined by the Griess reaction. The bacterial load were similar in B, MLN, PP and L of WT and KO mice. Cellular viability in the BAL was lower in infected WT and KO mice compared with uninfected mice. Macrophages were the predominant cells and an increase of lymphocytes was detected in the LBA of KO mice ($p<0.05$). Moreover, an increase of NO production was also detected in this group of mice ($p<0.05$). We concluded that a *Y. enterocolitica* O:8 could affect the immune response in the lung. IL-12p40 could play a protective role in the lung after *Yersinia* infection.

156.**ANATOMICAL STUDIES OF *Baccharis grisebachii***

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The objective of the present work is to accomplish an upgrade of *Baccharis grisebachii* Hierom, scientific names, synonyms, and common names, to provide a brief description of the plant and principally to undertake the study of the internal anatomy of the vegetative organs. This work will therefore provide micrographic reference standards, useful for quality controls of the vegetal. The anatomical study was performed on leaves, petioles, stems and roots, killed and fixed in F.A.A. solution. Samples for the histological studies were collected in San Juan, Argentine. The adaxial epidermis presents isodiametric cells. The abaxial epidermis presents anticline undulate and stomata are anomocytic. Stomata at the same level slightly rise over epidermis cells. Both epidermis present trichomes non glandular and glandular. Dorsiventral mesophile with 4-5 palisade parenchyma layers. Spongy parenchyma with 3-4 cells with intercellular spaces. The petiole presents a flat-convex contour. Adult stems evidence a secondary structure of conductive tissue, without peridermis. A di-arched root structure is initially present, show a superficial tuber when maturing. The natural drug, whether complete or fragmented, is identified by means of structural characters, in order to provide micrographic reference standards, useful for quality controls of the crude drug stage.

We are grateful to UNSJ, ANPCyT PICT Redes 260, for financial support.

157.**ENVIRONMENTAL ENRICHMENT PROTECTS DOPAMINERGIC NEURONS IN A MODEL OF PARKINSON DISEASE**

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It has already been shown that Enriched Environment (EE) is neuroprotective in different models of neurodegenerative diseases, as well as being capable of inducing changes in endogenous Trophic Factors levels (TFs). On the other hand, exogenous administration of Trophic Factors has neuroprotective effect. We hypothesize that EE protects dopaminergic nigrostriatal system from 6-OHDA toxin and that TFs play a role in this effect. For this purpose, rats were housed in standard conditions or in EE and given a unilateral infusion of 6-OHDA. EE induced a significant protection of dopaminergic system 21 days after the injury: immunohistochemical analysis showed protection of DA somas in the SN, reduced amphetamine-induced behavior, and maintenance of the nigrostriatal connection assessed by FluoroGold. No changes in endogenous TFs after EE were observed by Western Blot neither in the Substantia Nigra nor in the Striatum in unlesioned animals.

Taken together, these results show that EE significantly protects the Nigrostriatal system. Even though TFs were unchanged in unlesioned animals after EE, these proteins may be involved in the protection mechanism once the toxin is injected.

159.**LEUPROLIDE ACETATE (GNRH AGONIST) AFFECTS THE ANGIOGENESIS IN GONADOTROPIN-TREATED RATS**

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Introduction: the GnRH agonists inhibit folliculogenesis through an apoptosis increase acting like direct factors of the ovary. We have observed that Leuprolide Acetate (LA) treatment, a GnRH agonist, decreases VEGF-A and ANPT-1 expression levels and increases caspase-3 protein content.

Objectives: To evaluate the local effect of a GnRH agonist (Leuprolide Acetate, LA) on VEGF (Flk-1) and ANPT1 (Tie-2) receptors, vascular density and caspase-3 enzymatic activity determining the cleavage of PARP, an endogenous substrate of caspase-3. **Methodology:** Prepuberal rats were injected subcutaneously with eCG (25 IU/rat, Control group) and then injected with LA (LA group) and/or Ant (Antidote, GnRH antagonist) (Ant group and LA+Ant group respectively) at time 0 and at 12-h intervals for 48 h. The animals were sacrificed after 3 h the last LA and/or Ant injection.

Antral follicles were microdissected for western blot analysis and ovaries were fixed for immunohistochemistry analysis. **Results:** The LA treatment decreased the expression levels of Flk-1 receptor when compared to the control group ($p<0.01$), while the co-injection with Ant interfered with this effect ($p<0.01$). No changes were observed in the levels of Tie-2 after treatment with LA. Moreover, LA produced a significant enhancement in the cleaved PARP fragment as compared to the control group ($p<0.05$). Conversely, Ant alone significantly decreased PARP cleavage as compared to the control group and, once again, the co-injection of Ant reversed the apoptotic effect of LA ($p<0.05$). In the LA treatment, the vascular density is reduced compared to the control group. Again, the co-treatment with Antidote restored the values to the control levels. **Discussion:** These results suggest that the ovarian inhibitory effect exerted by the GnRH agonist, LA, could be mediated through changes in angiogenic factors and their receptors expression levels, leading these follicles to the atresia.

158.**SITUATION OF THE INFERIOR ORIFICE OF THE LATER PALATAL CONDUIT IN TOOTHLESS SKULLS HUMAN ADULTS**

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The later palatal conduit communicates the region to ptérigomaxilar with the ceiling of the buccal cavity. Its inferior orifice is white of anesthesia of the infiltrative type. In toothless patients the dental references do not exist, therefore it is necessary to locate this orifice by other methods and to avoid this way, ectópicas infiltrations.

- To determine the location of the orifice in human skulls toothless adults.

- To establish anatomical parameters of reference.

- To verify if significant differences in the side exist.

The measurement was made in toothless adult skulls of the osteoteca of the FOUNLP. The sample was of 100 (one hundred) UE, considering to each hemicráneo like a unit. The data turned upside down in tables of registry for their later analysis.

Anatomical parameters settled down. These partial results will be used for their later one I collate with dentated skulls.

160.**ASSOCIATION BETWEEN POLYMORPHISM OF THE BoLA DRB3.2* AND NEUTRALIZING ANTIBODY TITERS AGAINST TWO BOVINE VIRUSES**

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Introduction: bovine viral diarrhea virus (BVDV) and bovine herpesvirus type 1 (BHV-1) produce important economic losses. Although controversial results, vaccination with inactivated vaccines is a common practice. High neutralizing titers against BVDV denote resistance to the infection, and high neutralizing titers against BHV-1 could be associated to resistance to primary infection.

Objective: to study association between the *BoLA DRB3.2** gene and titers of neutralizing antibodies. **Material and methods:** cattle from 7 dairy herds from different regions of Argentina were analysed. All the cattle belonged to the Holstein breed, and were more than three years old. Neutralizing antibody titers were determined by neutralizing assays and alleles of the *BoLA DRB3.2** gene were determined by PCR-SSOP. Animals with neutralizing titers >1:128 were considered resistant. Association between antibody titers and alleles was estimated by relative risk (RR). **Results:** *DRB3.2* 1701* allele is associated with high titers of antibodies against BVDV and BHV-1. *DRB3.2*1401 or 02* could only be associated with high titers of antibodies against BVDV. **Discussion:** alleles *1701 and *1401 or 02 are associated to resistance. The design of control programs could be aid by genetic control.

161.**EVIDENCE FOR THE VERTICAL TRANSMISSION OF AN ENDOCYTOBIOTIC CYANOBACTERIUM IN *Pomacea canaliculata* (AMPULLARIIDAE)**

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P. canaliculata hosts a cyanobacterium within the midgut gland's cells. This is the only known obligate endocytobiosis between an animal and a cyanobacterium, and one of the few known in eukaryotes. In a previous study, all embryos that were aseptically obtained from eggs and that were cultured in a sterile medium over 7 days showed the endocyanobionts in their midgut glands at the end of this period, suggesting a vertical mother-offspring transmission of the symbiont. Since no cyanobacterial cells were morphologically identified within the eggs or embryos, we explored here the existence of sequences of the gene encoding for 16S rRNA, in a DNA extract from newly deposited eggs, using PCR and the generalized probes CYA 106F and CYA 781R. A band of the expected size (~670 bp) was obtained and sequenced, and was found identical to the corresponding sequence of cyanobionts obtained from adult snails. An attempt was made to identify the same sequence in DNA extracted from aseptically obtained juveniles. Also, a band of ~670 bp was obtained after preamplification with the generalized bacterial probes EUB 27F and EUB 1525R, and amplification with the cyanobacterial generalized probes.

162.**DENTAL HEALTH HABITS IN ROSARIO**Kohli A¹, Pezzotto SM², Poletto L².¹Cát Histología, Fac. Odontología; ²Inst. Inmunología, Consejo Investigaciones. UNR.

With the objective to know the adherence to oral health cares recommended for dental health, a young adults sample from public and private services concurrence was investigated applying a standardized anamnesis: age, sex, time elapsed between consultations, restorations, dental daily brushing and sweets consumption were recorded. A total of 517 patients were incorporated, 65% females and 35% males. At private attention centers concurred (G1, 19%), and at the publics (81%): one of the down town area (G2), and a marginal the other (G3). Last visit time elapsed was minor in G1 ($p=0.04$), which patients in greater proportion did endodontics, restorations and prostheses applied to extend tooth useful life ($p<0.001$). Daily brushing habit was 84% in G1, 67% in G2 and 69% in G3 ($p=0.04$). With regard to the brushing habit before going to bed it was not fulfilled by 49%; and 15% of those who did it had some food at bed. Had supper without brushing their teeth 50% of G1, 68% of G2 and 64% of G3 ($p=0.013$). When comparing G1 with G2 and G3 with regard to the total absence of food before going to bed the statistical difference was ($p=0.008$). In conclusion: G1 had shorter periods of time elapsed between consultations and more treatments done. The habit of daily correct brushing schedules was similar in the three services. Sweets consumption is extended to all the social groups, but there were differences in the food type.

163.**THE CANDID: FACTOR OF RISK IN SMOKERS**

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Candida is an opportunist fungi of the mouth and could be a cofactor for carcinogenic risk. It can be identified by means of exfoliative cytology in the oral mucosa. Our aims were to compare in the cytology of smoking patients and not smoker the presence of *Candida*. A standardized anamnesis was applied: age, sex, habit of smoking, quantity of cigarettes, time smoking, bacteria and inflammatory cells. The samples were obtained by scraped of mucosa and tongue of 112 patients. The cells were fixed and stained with hematoxilin-eosin and PAS were observed to 450 increases. The groups of age considered were - less than 25 years (22%), -25-40 (14%), -41-60 (40%) and - major of 60 (23%). 53% of the patients had the habit of smoking, but it was major in males (63%) with regard to the women (47%). Those of 41-60 years were those who more smoked (44%). Every day, 48% smoked 11-20 cigarettes and 22% of 6-10. Those who smoked for less than 10 years were 19%, of 10-19 30%, of 20-29 16%, of 30-39 25% and for more than 40 years 10%. Smoker's condition was significant for *Candida*'s presence ($p=0.04$); of bacteria ($p=0.001$) and of inflammatory cells ($p=0.0001$). In our city this habit was extended to both sexes with predominance in males. In major of 60 years the increase of the number was observed of not smokers. The habit of smoking, harmful to the normality of the mucous oral one, it increases the probability of falling ill of *Candida* and increases the factor of risk of enduring mouth cancer.

164.**VIRUCIDAL ACTIVITY OF *Larrea divaricata* (Zygophylaceae) EXTRACTS**Konigheim B¹, Aguilar J¹, Núñez Montoya S², Contigiani M¹.¹Inst. Virología, Fac. Cs. Médicas and ²Dpto. Farmacia, Fac. Cs. Qcas. (IMBIV-CONICET) - UNC. Córdoba, Argentina. E-mail: virolog@cmefcm.uncor.edu

In the search of new natural agents with antiviral activity, we analyzed the inhibitory effect of different extracts from *L. divaricata* against 4 different viral models: Junin virus (JV), herpes simplex type I (HSV-I), Saint Louis encephalitis (SLE) and Venezuelan equine encephalitis (VEE).

Aerial parts (50 g) were extracted with EtOH-H₂O (1:1). EtOH was eliminated in vacuo and aqueous liquid obtained was extracted with solvents of increasing polarity, providing 5 extracts: hexane, benzene, ethyl ether, ethyl acetate and aqueous remaining. The cytotoxicity was established by neutral red assay in Vero cells. The virucidal activity was determined at CC₅₀ by a plaque reduction assay under agarose (n=3).

All the extracts were active against JV (50-100% inhibition, I), being inactive for HSV-1 and SLE. With exception of the aqueous extract (75% I), the others one presented low inhibitory percentages (10-30%) against VEE. A main compound was isolated and purified from the aqueous extract by chromatographic column in polyamide. Chromatographic and UV-V spectrophotometric properties of this compound were coincident with those of the nordihydroguaiaretic acid (NDGA). These results suggest that further chemical study of other extracts should be performed.

165.**STUDY OF ARTICULAR ADHERENTS CELLS OF TNFRp55^{-/-} MICE WITH REACTIVE ARTHRITIS***Lacoste MG, Guzmán AMS, Di Genaro MS.**Immunology; Chemistry, Biochemistry and Pharmacy Faculty. National University of San Luis. E-mail: gabnela@unsl.edu.ar*

Reactive arthritis (ReA) is an aseptic arthritis, which is induced by an extra-articular infection. The pathogenetic mechanism and the role of the different cellular types are unknown. The aim of the present work was to analyze the adherent cell populations derived from joints of TNFRp55^{-/-} mice with *Yersinia enterocolitica* O:3 triggered-ReA. The cells were obtained from explants of inflamed joints of TNFRp55^{-/-} mice, and cultured in DMEM at 37°C with 5% CO₂. Cells were passaged when they reached confluence. The cells were observed with phase contrast and Giemsa. After 48 h, we observed round, stellate and spindle-shaped cells with prominent nucleus and nucleoli. Two weeks later, we observed multinucleated cells. The cellular morphology was consistent with macrophages and fibroblasts. After the passages, the rounded and multinucleated cells were reduced and, in a third passage, the cultures consisted almost entirely of spindle-shaped cells. We concluded that macrophages and fibroblasts are the predominant adherent cell populations in cultures from joints of mice with ReA. This methodology could be used to obtain fibroblast cultures to study the role of these cells in ReA.

167.**HISTOLOGICAL AND BIOCHEMICAL CHANGES IN LIVER AND PLASMA RATS EXPOSED TO CADMIUM***Larregle E, Gil E*, Giménez MS.**Cátedra de Bioquímica Molecular. Fac. de Qca, Bioqca y Fcia.***UNSL. San Luis. Argentina. E-mail: mgimenez@unsl.edu.ar*

Cadmium (Cd) is an important environmental pollutant. The chronic intoxication with Cd modified the lipid metabolism of rat liver. Previously, we showed that the exposure to Cd increase the triglyceride (TG) concentration, the activity of Fatty Acid Synthetase (FAS) and the expression of GPAT in liver of Cd rats respect to the controls, and increase the serum TG of the Cd rats starved 12 h, without alter the VLDL secretion of liver. For determinate if the oral chronic administration of Cd affect the TG accumulation in liver and the activity of extrahepatic lipoproteinlipase (LPL), adult male Wistar rats (180-200 g) were exposed to cadmium in the drinking water (15 ppm of Cd as CdCl₂) for 8 weeks. Fasted rats were injected i.v. with heparin (0.1 unit/g), and blood was collected after 10 min. Postheparin plasma (PPH) was added to the substrate mixture (triolein, BSA, Triton X-100, and heat inactivated human serum in 0.1 M Tris-HCl, pH 8.6) and incubated for 60 min at 37°C. The amount of glycerol released was determinated (Van Handel and Zilversmit, 1957). For the histochemical study of lipids, the livers were included formaldehyde-calcium with and without post-chroming and 10% formalin, embedded in paraffin or gelatine and coloured with acetylated Sudan black B and mounted in neutral glycerine jelly. The LPL activity in PPH decreased in Cd rats with respect to the controls (7.971 ± 0.039 vs 21.468 ± 0.012 ig glycerol / ml PHP/ h). The Cd liver showed numerous sudanophilic lipid droplets. Conversely, the control samples showed scarce sudanophilic lipid droplets. The hypertriglyceridemia is consequence of decrease activity of LPL. For other hand, Cd induced accumulation of lipid droplets, this fact may be related with the beginning of fatty liver.

166.**EXPRESSION OF EPITHELIAL CADHERIN (Ecad) IN HUMAN BREAST CANCER CELL LINES IBH-4 AND IBH-6***Lapyckyj L, Matos ML, Castillo L, Lentz EM, Gabrielli N, Lüthy I, Vazquez-Levin MH.**Instituto de Biología y Medicina Experimental (CONICET-UBA). Buenos Aires, Argentina. E-mail: lapyckyj@dna.uba.ar*

Introduction: Ecad is a 120 kDa transmembrane protein involved in cell-cell adhesion. Human breast cancer cell lines IBH-4 and IBH-6 are highly invasive and have a distinct C-term truncated 89 KDa Ecad form (Ecad89). **Objective:** To further characterize Ecad expression in IBH-4/IBH-6 cell lines. **Methods:** Protein analysis involved immunocytochemistry and Western immunoblotting with antibodies to different Ecad domains. Protein levels were estimated by densitometry using rec-Ecad as mass standard. mRNA analysis was done by RT-PCR. MCF-7 cells served as control. **Results:** In IBH-6 and IBH-4 cells: 1) Ecad protein levels decreased to 30% and 10% compared to MCF-7 (0.6 ng/μg cell protein) and 2) Ecad localization was weak and patchy along the cell. Ecad protein changes did not result from culture stress (similar results in cultures with 10/20% FCS). No accumulation of 35KDa Ecad was detected, and Ecad89 did not co-migrate with the Ecad ectodomain. Ecad mRNA levels were low in IBH-4 and IBH-6, and mapping studies revealed major changes in its 3'end. **Conclusion:** significant changes in Ecad expression (mRNA and protein) were found in IBH-4 and IBH-6 cells.

168.**MICROBIOLOGICAL QUALITY OF GOAT'S CHEESE***Lazarte Otero V, Lucero Estrada C, Velázquez L, Escudero ME, Guzmán AMS.**General Microbiology, National University of San Luis, 5700 San Luis. E-mail: vale_sab@hotmail.com*

The increase of consumption of goat's cheese highlights the importance of microbiological quality of this product. The natural microflora and presence of pathogenic microorganisms were investigated in goat's cheese from San Luis. Samples of 10 g were homogenized into 90 ml peptone water 0.1% and serially diluted to perform mesophilic aerobic counts (MAC) on trypticase soy agar and determination of most probable number of total coliforms and *Escherichia coli* using the multiple fermenting tube method. Pathogens were investigated as described: cultures in peptone buffered saline pH 7 followed by subcultures in tetrathionate and modified Rappaport broths and isolation on bismuth sulfite agar for *Salmonella* spp; enrichment in phosphate buffered saline pH 7,6 added with 1% sorbitol and 0,15% bile salts and isolation on Mac Conkey agar for *Yersinia enterocolitica*; enrichment in asparagine broth for *Pseudomonas aeruginosa*, and trypticase soy broth followed by Baird Parker agar for *Staphylococcus aureus*. Values of $8.93 \log \text{cfu.g}^{-1}$ for MAC and $>1100/100 \text{ ml.}$ for total coliforms and *E. coli* were observed. Two non-fermenting sorbitol *E. coli* strains which were challenged against O157 antiserum produced negative results. Other pathogens were not detected. Inadequate pasteurization, the addition of raw goat's milk or contamination during storage would be causes of low microbiological quality of goat's cheese.

169.**TROPHIC STATUS AND SPATIAL VARIABILITY OF RÍO TERCERO RESERVOIR, CÓRDOBA**

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Ecological systems can be classified as oligotrophic, mesotrophic and eutrophic. This study was performed in the Río Tercero reservoir, in the province of Córdoba, Argentina. The aims were to determine the eutrophication degree and the concentration of chlorophyll-a in non sampled places and to predict alga flowering to prevent health risks. Samplings were done during spring-time in the years 2005 and 2006 coinciding with the pass of LANDSAT 5TM satellite over the study area. Temperature, chlorophyll-a, Secchi disk, total nitrogen and total phosphorous were determined both, *in situ* and in the laboratory. An analysis of simple lineal regression between the bands 2 and 4 and chlorophyll-a, was performed. The responses obtained were those estimated for the proposed models and the spatial distribution maps for that variable. The values predicted for that variable in both spring times were validated and compared. The trophic status corresponded to mesotrophic. The study demonstrated that the data provided by the LANDSAT 5TM images could be used to survey water quality parameters and to determine the spatial variability of chlorophyll-a.

170.**PROSTATIC GLAND RESPONSE IS MODULATED BY TESTOSTERONE IN RAT BACTERIAL PROSTATITIS**

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In the male genitourinary tract, bacterial prostatitis is one of the main causes of prostatic inflammation. Recently, we have described a differential response of stroma and epithelium induced by *E. coli*. The aim of this study was to analyze if testosterone modulates this response. We used a bacterial prostatitis model in adult Wistar rats: **IN-T group** received 2.5mg/rat/day of testosterone and then was inoculated with *E. coli* suspension; **IN group** only received bacterial suspension; both groups were sacrificed 24, 48 hours and 5 days after inoculation; finally, **T group** only received testosterone by 10 days. Rat ventral prostate was processed for morphological analysis ICQ for α -smooth actin and TGF β ; tissue homogenate was used for Western Blot of TGF β . Several aspects of bacterial injury response were modified by testosterone. At 24 h post-infection, in both groups the periacinar muscle was thickened; but in **IN-T** smooth muscle cells were more hypertrophied and exhibited secretory phenotype and less α -actin content. Moreover, testosterone prevented neutrophils entrance to the acine and TGF β upregulation by epithelial cells, as occurred in **IN**. At 48 h, periacinar muscle layer was interrupted in **IN**; by contrast, muscle cells maintained stimulated and thickened in **IN-T**, with low neutrophil infiltration; fibroblasts expressed TGF β . Testosterone avoided epithelial cells apoptosis that occurred in **IN**, and therefore epithelium did not atrophy. At 5 days, fibroblasts in **IN-T** exhibited marked stimulation, synthesized great quantities of collagen and assembled in a net. In summary, the results suggest that infection-testosterone combination is a highly stimulatory condition for both, stromal and epithelial cells, especially for fibroblasts that probably experimented transdifferentiation to myofibroblast and established a "reactive stroma".

171.**EFFECT OF EARLY STIMULATIONS ON HYPOTHALAMIC-PITUITARY-ADRENAL AXIS AND SOME IMMUNE PARAMETERS IN PRENATAL STRESSED RATS**

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Prenatal stress (PS) produces an alteration of the hypothalamic-pituitary-adrenal (HPA) axis that would induce a long-term alteration of the immune function in the offspring. Early postnatal stimulations (M) produce beneficial effects on the long-term emotional reactivity and HPA axis activity that could revert the effect of PS. The objective of this work was to investigate the effect of M in PS animals on HPA axis, the distribution of the subpopulations of leucocytes and the T lymphocytes proliferation *in vitro*, in response to acute stress. Males of three months of age were used, PS for immobilization (IMO) and controls (PC) during pregnancy. Half of the PS animals were M during the first week of life. Animals of both groups were IMO stressed (20 minutes). Blood was extracted at 0, 20, 60, 90, 120, 150 and 330 minutes, white blood cells were counted, and the subpopulations of leucocytes and the levels of corticosterone (COR) determined. The spleen of animals was removed for T cells culture. The response of white blood cells and lymphocytes is depressed. The COR levels, neutrophils and T-cells proliferation is increased in M animals. In conclusion, M reverts the effects of PS with re-exposure to the same postnatal stressor on: a) The activity of HHA axis, b) The dynamic of leucocytes distribution, which could improve the immune surveillance for pathogens, c) The functional response of T lymphocytes.

172.***P. putida* ENZYMATIC ACTIVITIES IMPLICATED IN THE RESISTANCE AND DEGRADATION OF TETRADECYLTRIMETHYLMONIUM**

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Pseudomonas putida responds to tetradecyltrimethylammonium (TDTMA) through quantitative changes in membrane phospholipids (PL). The specific variations observed were increase of phosphatidic acid (PA) and phosphatidylglycerol (PG) and decrease of cardiolipin (CL). The turnover of CL might be an efficient method to replenish PA and PG pools, through the hydrolysis of CL catalyzed by a phospholipase D activity (PLD). The first step of TDTMA degradation by *P. putida* is catalyzed by a monooxygenase activity (Mo), producing trimethylamine (TMA) and tetradecanal. PLD activity was assayed by a transphosphatidylation assay using [32 P]-Pi and ethanol. When cells were exposed 5 min to 50 mg l⁻¹ of TDTMA, a 72% increase of PLD activity relative to untreated cells, was observed. After 15 min of TDTMA-contact no significant changes were detected. The Mo activity was measured in cell-free extracts obtained of *P. putida* grown with TDTMA as C and N source. The enzyme was localized in periplasm and it was shown to be NAP(P)H-dependent and inhibited by TMA, effect reverted by the presence of Al³⁺. IEF analysis showed one band of pI 7.3 with Mo activity, which was further resolved by PAGE-SDS in 3 bands. One of these bands was identified as a 60 kDa GroEL chaperonin by MALDI-TOF and PMF. We propose that this chaperonin would be involved in the transport and folding of periplasmic Mo activity.

173.

ABSCISIC ACID LEVELS IN SALINIZED SEEDLINGS OF *PROSOPIS STROMBULIFERA*

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In glycophytes, stress tolerance is increased with high concentrations of ABA, whereas in halophytes the ABA role is not yet known. Thus, endogenous levels of ABA in leaves and roots of salinized plants of the halophyte *P. strombulifera* were determined. Seedlings were grown hydroponically in Hoagland solutions with addition of 50 mmol/l NaCl for NaCl treatment, 38 mmol/l Na₂SO₄ for Na₂SO₄ treatment and their isoosmotic mixture for bisaline treatment, every 48 h until final $\Psi_0 = -1, -1,88$, and $-2,6$ MPa were reached. Control plants were grown in Hoagland 25%. ABA levels were determined by HPLC-GC-MS at 6, 12 and 24 hours after the salt pulse. ABA levels differed with the type of salt, the concentration, the analyzed organ and the plant age. A remarkably higher ABA content was found in leaves in comparison with roots, maybe due to its protective role, and a rapid biosynthesis and distribution from roots, where higher levels were detected at the beginning of treatments. Sulfate treated leaves showed the highest ABA levels, in coincidence with toxicity symptoms showing up. Dynamics of ABA levels from 6 to 24 h in the different treatments would indicate that ABA would act like a triggering signal for adaptive biochemical and molecular mechanisms for the plant survival to salinity.

175.

EFFECT OF SALINITY ON GERMINATION OF *Caesalpinia paraguariensis* (D:Parodi) Burkart

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Adaptation of plants to salinity during germination and early stages of seedling is crucial for their establishment. With the aim of determining the effect of salt over germination of *Caesalpinia paraguariensis* in chaquenian woods, this species was exposed to different CINA and polietilenglicol concentrations. The treatments were: 0, 0.2, 0.25, and 0.3 molal of CINA and its equivalent value in osmotic pressure of polietilenglicol. Four repetitions of 25 seeds each one, were put in incubator at 25°C with a 12 hours photoperiod. The Germination speed index was determined (IVG) and also the medium time of germination (TMG). The treatments were compared using analysis of variance. There were significant differences in IVG and in TMG between the tester and all of the treatments. The IVG diminished more than 50% in treatments with CINA (5,87%) according to tester (18,41%). The negative effect caused by PEG increased when concentrations were superior. TMG doubled with CINA (12 days) treatment and with the two smaller concentrations of PEG. With 0.3 molal of PEG there was no germination. This data confirm that salinity produces primary injury and secondary stress damage on germination of these species.

174.

FIELD EVALUATION OF COMPONENT TRAITS OF BIOMASS PRODUCTION FOR THE IDENTIFICATION OF SOMACLONAL VARIANTS OF BUFFEL GRASS

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Fresh weight (PF) and dry weight (PS) are main traits of biomass production, of agronomic interest to evaluate of new genotypes. In order to determinate the utility of these morphological traits to discriminate somaclonal variants of Buffel grass, PF, PS and PF-(PS/PF) were compared to the control. The plant material used was offsprings of plants obtained from embryogenic calli (CE) exposed to different concentrations of NaCl (S) and ethylmethanesulphonate (M). After two consecutive years in the field, S progenies showed PS and PF higher than the control lines. M progenies had lower PF, although, this difference was not significant respect to the control. M progenies showed significant differences in relationship PF-(PS/PF) respect to control lines, whereas not statistically significant difference were observed in S progenies. Since, PF, PS and its relationship not allowed to discriminate between M, S offsprings and control lines, they do not constitute, by themselves, good discriminant markers. Summing up, PF, PS and PF-(PS/PF) should be used all together to evaluate somaclonal variants in Buffel grass.

176.

SELECTION AND IDENTIFICATION OF ESTERASE-PRODUCING MICROORGANISMS

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One of the most efficient and successful means of finding new microbial enzymes is to screen a large number of microorganisms, because of their diversity and versatility. The capability of *Bacillus* strains to produce and to secrete enzymes has placed them among the most important industrial enzyme producers. Esterases, one of them, have important biotechnology applications, such as drug synthesis. The aim of the present study was to select and to characterize good esterase producing bacteria. Esterase-producing colonies were selected by their ability to hydrolyze α -naphthylacetate on LB plates. The selected strains were characterized with molecular and biochemical methods. Five of fifty isolated were selected because of their high levels of extracellular and cell-bound esterases production. On the basis of their biochemical and molecular properties the strains A14 y A55 appear to belong to *B. pumilus*, A60 y A62 to *B. subtilis* and M2 to *B. cereus*. The nucleotide sequences of the 16S rDNA are available in GenBank under the following accession numbers A14 EF462914, A55 638794, A60 EF513611, A62 EF513612 y M2 EF 513610. This characterization will provide tools for designing new enzymatic processes.

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177.**HETEROLOGOUS PROTEIN EXPRESSION IN THE METHYLOTROPIC YEAST *Pichia methanolica***

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Recombinant proteins have become of wide use in laboratory as well as clinical applications. Different expression systems have been used for this purpose, becoming a common and frequent tool in Molecular Laboratories. In the past years, the methylotrophic yeast *Pichia methanolica* has developed into a highly successful system for the production of a wide variety of heterologous proteins. This eukaryotic organism has several characteristics that suggest it is well-suited for expression of recombinant proteins. The conditions for protein expression system in the methylotrophic yeast *Pichia methanolica*, were set up for a fragment of Angiotensin II AT₂ receptor. Starting from the fragment (585 bp) subcloned in pGEMTeasy vector, the C-terminal fragment was subcloned at the three open reading frames of pMETα vector. We achieved the expression, secretion and identification of the AT₂ fragment into the growth media of our recombinant protein. Yeast combines the ease of genetic manipulation and fermentation of a microbial organism with the capability to secrete and to modify proteins according to a general eukaryotic scheme.

178.**ANTIOXIDANT ACTIVITY OF METHANOLIC EXTRACTS FROM AMARANTH SEEDS**

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INTRODUCTION: antioxidants are diet components that reduce the oxidative stress caused for free radicals. Amaranths are recognized for their nutritional quality and the presence of bioactive components that contribute to optimize human health. Phenols, flavonoids and anthocyanins have been detected in amaranths in previous studies in our group.

OBJECTIVE : to evaluate the antioxidant activity of methanolic extracts from amaranth seeds.

METHODS: Extraction of total phenols (Singleton *et al.*, 1965) DPPH assay (Brand – Williams *et al.*, 1995), Scavenging activity against nitric oxide (NO test), (Marcocci *et al.*, 1994). Samples: *Amaranthus cruentus candil* (Acc), *Amaranthus cruentus mexicano* (Acm), *Amaranthus mantegazzianus* (Am), *Amaranthus hypochondriacus* (Ah).

RESULTS: Inhibition NO %: 45,1 (Acc); 42,5 (Acm); 29,3 (Am) and 35,2 (Ah). Inhibition DPPH %: 86,54 (Acc); 86,16 (Acm); 86,54 (Am) and 86,93 (Ah).

DISCUSSION: Our results are coincident with findings from other authors and are similar to effects detected in other plant species considered to have significant antioxidant activity. However due to the complexity of oxidation and antioxidation processes, it will be necessary to perform additional studies in order to improve the simulation of *in vivo* conditions.

179.**ICHTHYOFAUNA COMPOSITION OF LA VIÑA RESERVOIR (CÓRDOBA, ARGENTINA)**

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The biologic integrity of a fish community is a sensitive indicator of the “health” of an aquatic ecosystem and reflects environmental conditions of the water body. The goal of this study was to evaluate the specific richness and diversity of the ichthyofauna in La Viña reservoir (31°47'S, 65°01'W, 10.5 km², 846 m asl). Possible relationship between diversity indices values and sample size was assessed by means of Spearman's correlation. Fifteen samplings were performed in 1999-2002 using trawl nets and floating gill nets. 3242 specimens were captured of seven species distributed in five orders and five families. The pejerrey *Odontesthes bonariensis* was the species with greatest biomass (63.8%) and is the main target of fishing activities in the reservoir. The tetra *Astyanax eigenmanniorum* was the most frequent species (52.9%). Considering the total sampling, the diversity was $H'_r = 1.63$ (IC95% 1.49-1.77). The Shannon Wiener (H) and Simpson (λ) indices varied between 0.47 to 1.95 and 0.12 to 0.72 respectively. There was no significant correlation between these indices and the sample size (H : $r_s = 0.093$, $P = 0.74$; λ : $r_s = 0.25$, $P = 0.36$), which indicates that the diversity values are not an artifact in the number of captured fishes. Comparing with seven other reservoirs from central and northern Argentina, the specific richness of La Viña reservoir is considered moderate.

180.**USE OF DIFFERENT CONDITION INDICES IN PEJERREY *ODONTESTHES BONARIENSIS* (VALENCIENNES, 1835)**

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The biological parameters of the ichthyofauna vary in a temporal and spatial manner. The most common parameters of use are the weight-length relationships and the condition factor. In central Argentina, the pejerrey *Odontesthes bonariensis* (zooplanktivorous fish) is the main species for commercial and recreational fisheries. Precisely, diagnosis about *O. bonariensis* populations are based on the use of these indices and reflect indirectly the availability of food. The relationships between seven indices of *O. bonariensis* with the zooplankton abundance in a reservoir in Cordoba province were evaluated. Fifteen sampling procedures were performed and 952 fishes were captured. Also the organisms of the zooplankton were quantified (org/m³) in five points of the lake. Wet weight (W), cephalic length (CL) and standard length (StL) were registered for each captured fish. Fulton's condition factor (K : $r_s = 0.74$, $P < 0.01$), relative condition factor (ICr: $r_s = 0.72$, $P < 0.01$), weight at 200 mm of StL (W_{200} : $r_s = 0.66$, $P < 0.01$) and relative weight (Wr: $r_s = 0.63$, $P < 0.01$), showed the best correlation with zooplankton amount. The last three indices have the advantage of not being biased by the fish length. When grouping the indices by season of the year, significant differences ($P < 0.01$) were observed amongst them.

181.**GENOTOXICITY OF GLYPHOSATE AND ITS ENVIRONMENTAL METABOLITE AMPA BY MICRONUCLEUS TEST IN MICE**

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Glyphosate is the active principle of herbicides used in Argentina since 1997, and AMPA is its major environmental metabolite. Genotoxicity was quantified in Balb C mice (n: 4-5/group) treated with 100, 200 and 400 mg/Kg of glyphosate or 200 and 400 mg/Kg of AMPA in bone marrow through the micronucleus test. The mean of micronucleated cells/ 1000 erythrocytes (MNE) for the upper dose of glyphosate; AMPA 200; AMPA 400 mg/Kg and the positive control (Cyclophosphamide 20 mg/Kg) were 13,0±3,1; 10,0±1,0; 10,4±3,3; and 19,2±3,9 MNE; statistically different against the negative control (3,0 ± 0,8 MNE) in all cases (p< 0,01 Tukey post test). The genotoxicity observed at 200 mg/Kg of AMPA was absent at the same concentration of glyphosate with 4,2±0,5 EMN. In the comet assay in mice we have also detected DNA damage by glyphosate at 400 mg/Kg, not observed at 100 and 200 mg/Kg. EPA determined that AMPA has not to be included in risk evaluations studies. However, little is known about its biological activity and, taking into attention that glyphosate and AMPA residues are found in the environment, the valoration of possible health risk by both chemicals need to continue.

182.**MALE REPRODUCTIVE APPARATUS OF *Nantis indefessus* (TELEOSTEI, CHARACIFORMES)**

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Fund. Miguel Lillo. Tucumán.

Introduction: The presence of insemination (internal fertilization) in some species of "clade A" presents interesting questions respecting the phylogenetic pattern of insemination in Characidae. The genus *Nantis*, that would be included in this clade, presents external morphological features agreeing with insemination. This possibility, and its unknown phylogenetic position make interesting the study of its reproductive biology.

Objectives: To determine reproductive structures in males and to establish the possibility of insemination.

Methodology: Sexually mature males of *Nantis indefessus* from "La Caldera" river, province of Salta, were decalcified and processed to optical histology, making sagital and frontal cuts. Then, it were stained with Hematoxilina-eosina and Gallego's Trichromic.

Results: The male has testes divided in two portions well defined: a spermatogenic proximally and an aspermatogenic distally. A sperm storage area was found behind the aspermatogenic portion. It presents a paired stucture and has a bilateral conection with the efferent duct. The interior contains aquasperms (spherical structure sperms). **Discussion:** This species shares with Glandulocaudinae and Stevardiinae, both inseminating, the presence of a paired storage area in testes. Sperms morphology, however, is similar to no inseminating species.

183.**IDENTIFICATION OF THE INTERSTITIAL CELLS OF CAJAL IN THE ABOMASUM OF BOVINES**

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The interstitial cells of Cajal (ICC) participate in the control of the gastrointestinal motility. The ICC were described in different mammals including our findings in the digestive organs of bovines, pigs, horses, rabbits and cats. Here we describe the distribution of c-kit positive cells using the CD117 monoclonal antibody. Samples were collected from the pyloric region of abomasums of six healthy cows after slaughter. Tissue samples were fixed and embedded in paraffin. Immunohistochemistry was performed using LSAB® (DAKO) as a detection method. Ten images per slide were captured for morphometric purposes using an image analyser (ImagePro Plus 5.1 - Media Cybernetics). c-kit positive cells were found between muscle layers of the organ either surrounding ganglion cells of myoenteric plexus or isolated or forming clusters of cells. Morphometric parameters of c-kit positive cells reveal an area of $30.95 \pm 6.21 \mu\text{m}^2$; a major axis of $10.55 \pm 1.72 \mu\text{m}$ and a minor Axis of $3.87 \pm 0.56 \mu\text{m}$. Their location in the muscular layers, their relations with ganglion cells of the myoenteric plexus and the immunohistochemical and morphometric characterization led us to identify them as ICC.

184.**EFFICACY OF SUCROSE SATURATED SOLUTION IN DOMESTIC CAT RINGWORM**

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Microsporum canis invades layers of keratinized skin, nails and hair of *Felix catu*. In kittens, after 1 month of incubation, it produces the typical ringworm lesion: round bald patches with scales, crusts, erythema and peripheral hair of weak implantation. This mycosis can be transmitted to dogs and humans. The aim of this work was to determine the clinical efficacy of sucrose saturated solution with eugenol and polyethylenglycol 400 (peg) in *M. canis* lesions in young cats. A solution of 250 gr. sucrose dissolved in 100 ml water at 80°C, to which eugenol (0,4% v/v) and PEG 400 (0,4% v/v) were added, was applied topically every three days, in 19 cats of up to one year of age, over lesions positive to Wood's lamp test and to those cats in which *M canis* was detected though microscopy. The treatment was performed until clinical healing in all animals (mean= 21 days), follow-up extended for a month after healing without relapse. Response to treatment can be explained through water activity (a_w) decrease caused by the solution's nature and concentration and eugenol antisepsis; stabilizing PEG would additionally decrease a_w ; some cats could spontaneously involute (rare, because of age). The use of a saturated sucrose solution + eugenol + PEG is suggested as a therapeutic alternative against ringworm in cats.

185.**EARLY PREGNANCY FACTOR PLACENTAL IMMUNOLOCALIZATION AT MID-GESTATION AND AT TERM**

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It has been postulated that Early Pregnancy Factor (EPF) contributes to the differentiation and growth of embryos. It has a two-phase production, with a second tip of production around the day 40 and it staying until the end of pregnancy. Therefore it is interesting to investigate the presence of this important factor at reproductive tissues since EPF would be a good indicator of embryonic viability. The objective of this work was to evaluate the immunolocalization of EPF in pig placentas of 40, 50 gestation days and at term. Placenta sows from 40, 50 gestation day and at term were used. Regional distribution of EPF was determined by immunohistochemistry (Biotin-Streptavidin-Peroxidase) using polyclonal anti-swine EPF antibody developed in rabbit immunized with synthetic EPF (University of Humbolt - Berlin). A high intensity EPF expression was present at 40, 50 gestation days and less evident in at term placentas. To the 50 days only the fetal tissues were positive to EPF. We concluded that the EPF production decays as the pregnancy progress, coincident with the role as embryonic viability marker and growth factor.

186.**DISTRIBUTION OF LYMNAEID SNAILS IN MALARGUE, MENDOZA PROVINCE**

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Lymnaeid mollusks are the intermediate hosts of fascioliasis, a zoonotic parasitic disease that causes heavy losses in livestock. Even though ranchers and veterinarians in Malargue department of Mendoza province are familiar with the disease and human cases have been reported, there is very scarce information in relation to the distribution of the intermediate hosts, the lymnaeid snails. Our objective was to describe where these vectors are found in Malargue department, Mendoza. During spring and summer of 2005/2006, water bodies with ecological conditions appropriate for the development of lymnaeid snails were surveyed. In each site, altitude, latitude and longitude were registered by means of the GPS. The snails collected were transported live to the laboratory and identified according to the keys of Castellanos & Landoni. Lymnaeid snails were found in 9 sites at an altitude ranging from 1463 to 1894 meters above sea level. They were found in tributaries of the Malargue River and also in tributaries of the Rio Grande river. This ample distribution indicates its adaptability to the adverse climate in the region and the high risk of transmission to both cattle and humans.

187.**COPROPARASITOLOGICAL STUDY OF GOATS FROM MALARGUE, MENDOZA**

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Goat production is an important activity in Mendoza province. Most of the herds are located in Malargue department. Parasitic diseases are an important obstacle for attaining high productive levels and some of these are important zoonotic diseases. Our objective was to describe by means of coproparasitological studies the parasites in two goat ranches in Malargue department. Samples of feces were obtained from the rectum in 219 animals. In the laboratory the samples were thoroughly mixed and filtered and three different concentration techniques were used in each sample: simple flotation, formo-ether sedimentation and Lumbrales technique of rapid sedimentation. *Eimeria spp.* oocysts were found in 208 samples (95%), *Fasciola hepatica* eggs in 83 samples (37,9%), *Nematodirus spp.* eggs in 68 samples (31,5%), *Trichuris spp.* in 6 samples (2,7%), Trichostrongyles in 3 samples (1,4%) and 3 samples were negative (1,4%). These results indicate that parasites are affecting the health of these herds and could be producing important economic losses. The high prevalence of fascioliasis is of utmost importance since it is a zoonotic disease. Control measures should be developed and put into effect.

188.**PHOSPHOLIPID METABOLISM: EFFECT OF SALINE STRESS IN BARLEY SEED (*Hordeum vulgare*)**

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The plants developed a variety of physiological and biochemical responses to adapt to adverse environmental conditions. Saline stress is certainly one of the most serious abiotic factors that limit the normal development of the plant and it displays diverse signaling processes. Therefore, our aim was to study physiological and biochemical changes to levels of signaling lipids in aleurone of barley and during the seed early germination under saline stress conditions. Decreases of growth in seedlings (4 days), in sterile and dark conditions with different NaCl concentrations were observed. Phospholipids (PLs) identified were PIP₂, PIP, DGPP, LPA and PA when analyzed the lipid kinase activities through the incorporation of phosphate from [γ -³²P]ATP to their endogenous substrates. In epicotyls and roots without stimulus, PIP predominated whereas in aleurone, DGPP and PA were the majority phospholipids. These observations suggest that lipid metabolism is modified during the transition of the state of dormancy to germination and it affects in opposite manner phospholipid levels. Saline stress produced an increase in level of PA in root; however in epicotyls no change was observed. Although, plants respond physiologically to saline stress as a whole, the metabolic responses differ from an organ to another.

189.**HUMAN PAPILLOMA VIRUS (HPV) IN ODONTOGENIC PATHOLOGIES**

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The ameloblastoma is a local aggressive tumor that appears with a relative frequency. The OMS in 2005 consider the keratocyst as a tumor. It has been avire an association between HPV and keratocyst. The objective is identify HPV and associate it with ameloblastoma and queratoquiste using polymerase chain reaction (PCR). It has been analyzed eight ameloblastomas. The mean age was 43 years-old, 6 of the cases belong to females. All the tumors were mandibular. The keratocyst ($\eta=4$) with a mean age of 27.7 years-old present 3 cases on females and all were localized in mandible. The HPV was detected by PCR and LIS-SSCP (low extragelyc ionic solution). It has been used like primers internal and external My09/11 y GP05/06. A control sample was taken. One hundred percent of the serie was positive for HPV. The viral types found were: VPH 6(3)= 37,5%; 16 (3)= 37,5%; 11(1)= 12,5% ; 18 (1)= 12,5%. From the control samples ($\eta=10$), only two were positive. All the queratoquistes were negative for HPV. The association in the serie allow to infer a role of the virus in the etiology of the tumor. It can't be rule out a contamination from the mucosal surface. (Kahn M. and Sand *et al.*) The η should be bigger for valid the finding of ameloblastoma an keratocyst.

191.**ANATOMICAL REFERENCES OF THE FEMORAL VEIN IN THE INGUINO CRURAL REGION OF FETUS**

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The access to the main circulation can be made through the catheterism of the femoral vein, chosen mainly in newborns, and implies the recognition of the position of the vessel following some anatomical references of the inguino crural region, delimiting, therefore, the precise site for the puncture. Anatomical references: Distance pubis-femoral vein; Distance pubis-inguinal ligament and Distance inguinal ligament-femoral vein. It has been studied 20 inguino crural regions of fetus fixed in formalin 10% of both sexes without any specific pathology, between 23 and 33 weeks of gestational age. The cases were grouped in different models according to the obtained measures. It was concluded that in the obtained models, the variability found suggests to apply the following measures for the puncture: Distance pubis-femoral vein: between 11-12 mm; Distance pubis-inguinal ligament 13-10mm; Distance inguinal ligament-femoral vein: 3-5,5 mm. We consider of importance to respect these measures, constant in the first stage of life, to avoid injuring adjacent anatomical structures and repeated futile punctures that increase the risk of infection. These contributions will diminish the errors in the technique in the catheterism of newborns in intensive care.

190.**EMBRYOLETAL, PHARMACOKINETIC AND PHOTOTHERAPEUTIC STUDIES OF A PHTHALOCYANINE FOR PHOTODYNAMIC THERAPY (PDT) APPLICATION**

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PDT causes tumoral tissue damage although combination of a photosensitizer (PS) and visible light. The aim of this work was evaluate embryoletal, pharmacokinetic and phototherapeutic effects of a new PS, ZnPcCF₃ phthalocyanine, for its application in PDT. *Bufo arenarum* embryos were exposed to ZnPcCF₃ solutions (4,66 x 10⁻⁷ M and 2,80 x 10⁻⁷ M) with the objective of determine embryos mortality in darkness and illumination conditions. Nevertheless, the drug precipitated in the media where larvae were collocated. The examination of renal and hepatic toxicity and functionality (histological analysis and biochemistry parameters) in Balb/c mice, with the administration of 0,2 mg/kg ZnPcCF₃ indicated a low acute toxicity with revertible damages. Phototherapeutic effect was determined analyzing the percentage of tumoral death after PDT. It resulted in 89,40% at 4 day post- PDT. Biodistribution of ZnPcCF₃ demonstrated that it does not accumulate in brain, which should eliminate toxicity risks in this organ. The drug was not registered in skin, then it should not produce cutaneous photosensitivity. The results obtained indicate that the PS should be proposed for PDT.

192.**MELOLONTHIDAE (COLEOPTERA) CAPTURED WITH TRAP LIGHT “POZO DEL CARRIL” (AGRONOMY AND VETERINARY UNRC) CORDOBA, ARGENTINA**

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Melolonthidae, widely distributed in Argentina, live in different types of natural and modified vegetation. The larvae (white worms) and the adults can be agricultural plagues when consuming roots, leaves and/or fruits. The purpose of this research was to elaborate a taxonomic list and to analyze the phenology of the Subfamilies and Tribes of Melolonthidae registered in “Pozo del Carril” (experimental field FAyV-UNRC). Data were obtained daily from 01/XII/2005 to 30/XI/2006 and collected with light trap in maize culture with little forestation. From a total of 11,463 Scarabaeoidea individuals, there were determined 1,458 Melolonthidae, among them: Dynastinae (Cyclocephalini, Phyleurini), Melolontinae (Pahidemini, Diplotaxini, Melolonthini), Rutelinae (Anomalini, Rutelini). Diagnostic characters and behavior of the Subfamilies and Tribes of Melolonthidae were compared and a key was elaborated. Dynastinae, Melolontinae and Rutelinae presented high adult density between December and January, associated with the highest annual temperatures and precipitations.

193.**MULTIVARIED ANALYSIS OF SCARABAEINAE ASSOCIATION (COLEOPTERA) WITH ENVIRONMENTAL GRADIENTS IN THE PROVINCE OF CORDOBA***Monteresino EM, Corigliano M Del C.**Dpto. Cs. Naturales. FCEFQ y N. UNRC. Rio Cuarto. Córdoba.**E-mail: emonteresino@exa.unrc.edu.ar*

Scarabaeinae species (Scarabaeoidea-Col.) live in different type of biomas. There were examined samples taken from representative localities in order to know their distribution in Cordoba ecoregions and the species list was developed. Environmental variables of each locality were determined. The samples were collected with light and pitfall traps, during five year of studies. Gamma diversity was 75 species. Eco-ethological diagnoses were made of each species. From a presence - absence matrix a *Canonical Correspondence Analysis* (CCA) was performed. There were ordered species associated to each ecoregion and other species of wide distribution. The association *Gromphas lacordairei*, *Bolbites onitoides* and *Coprophanaeus milon* is indicative of the Eastern Chaqueño Forest and *Coprophanaeus bonariensis*, *Glyphoderus sterquilinus*, *Tetraechma tarsalis* of Western Chaqueño Forest. The species of greater vagility did not were ordered in relation to any environmental variable nor ecoregion. Therefore, it stand out from the studied assemblage, species of distribution restricted to determinate bioma, which could be considered vulnerable to changes on land use and its absence would be indicative of environmental impairment.

194.**POSSIBLE PARTICIPATION OF A NEUROTROPHIN RECEPTOR IN THE MECHANISM OF NEURONAL DEATH STATUS EPILEPTICUS-INDUCED***Montroull L, Unsain N, Mascó DH.**CeBiCeM. FCEFyN. UNCordoba.**E-mail: laura_montroull@hotmail.com*

The interaction of Brain-Derived Neurotrophic Factor (BDNF) with TrkB receptor has been implicated in neuronal survival, while the interaction of BDNF and/or its precursor (proBDNF) with p75ntr has been implicated in neuronal death (apoptosis). Severe and prolonged seizures (Status Epilepticus, SE) produce death in the hippocampus. SE can be studied in the lithium-pilocarpine model in rats. In this project we studied if SE induces changes in the protein levels of BDNF, proBDNF and their receptors. SE induced a remarkable decrease in TrkB levels determined by Western blot and this modification preceded the neuronal damage induced by SE. Interestingly, we found neither neuronal death nor TrkB decrease in animals that after being injected with pilocarpine showed mild, intermitent seizures but did not developed SE (NoSE). At the same time, the interaction of proBDNF with p75ntr, analized by co-immunoprecipitation, significantly increased after SE, but also occurred in the NoSE group. These results suggest that to produce neuronal injury, a decrease in TrkB receptor levels might be necessary, while proBDNF and p75ntr interaction would not be a determinant factor.

195.**NITRIC OXIDE INFLUENCES REACTIVE OXYGEN SPECIES PRODUCTION AND BOVINE OOCYTE IN VITRO MATURATION***Morado SA, Cetica PD, Beconi MT, Dalvit GC.**Biochemistry, School of Veterinary Science, University of Buenos Aires.*

Nitric oxide (NO) produces inhibition of cytochrome oxidase and stimulation of O_2^- production by inhibition of electron transfer at complex III. The aim of this work was to determine NO participation in reactive oxygen species (ROS) production and meiotic maturation. Cumulus-oocyte complexes (COCs) were recovered from bovine ovaries and cultured in medium 199 (control), 0,5 mM N-nitro-L-arginine methyl ester (L-NAME, endothelial NO synthase inhibitor) and 10 μ M sodium nitroprusside (SNP, NO donor) at 39°C, 5% CO_2 in humidified air for 22h. A cytochrome oxidase inhibitor, 1 mM potassium cyanide was also assessed. Meiotic maturation was determined by the presence of metaphase II. ROS production was determined in denuded oocytes at 0, 6, 12, 18 and 22h by the ratio between 2',7' dichlorodihydrofluorescein diacetate and fluorescein diacetate assays. Maturation percentages were 80, 81 and 58% for control, L-NAME and SNP, respectively. ROS levels fluctuated throughout the 22h of maturation, showing a similar pattern with the different treatments, but a decrease at 6h and an increase at 12h, were observed in COCs cultured in the presence of SNP compared to the control ($p<0,05$). Potassium cyanide increased ROS levels at 12h ($p<0,05$). These results suggest that exogenous NO modifies H_2O_2 production and reduces mitochondrial oxidative energy, impairing the maturation process.

196.**ABA INCREASE THE PHOTOSYNTHETIC EFFICIENCY IN *VITIS VINIFERA* CV. MALBEC***Moreno D, Berli F, Bressan-Smith R, Bottini R.**Facultad de Ciencias Agrarias. UNCuyo.**E-mail: dmoreno@fca.uncu.edu.ar*

ABA is a phytohormone that modulates the plant responses to different stresses (drought, temperature, salinity, UV-B). Tungstate (W) inhibits ABA biosynthesis by blocking the enzyme ABA-aldehyde oxidase. Malbec is the typical variety of red wines from Argentina, where the cultivated area is the highest in the world. The chlorophyll *a* fluorescence is used in order to know the photosynthetic efficiency. The objective of the work was to study the ABA effect on the photosynthesis in grapevine. Plants of *Vitis vinifera* cv. Malbec vineyard were sprayed every 7 days on to the leaves with ABA (250 ppm), W (165 ppm) and H₂O (control), from 15 days before flowering until harvest. The ABA-treated plants showed greater Fv/Fm and lower NPQ with respect to W, which means greater photochemical efficiency. In addition, the ABA-treated plants presented greater content of Chl *a* and photoprotective pigments, that might be important at noon when excessive intensities of PAR and UV-B are registered. In the W treated plants the berries were smaller. These results suggest that ABA protected the Chl *a* by increasing the photoprotective pigments, allowing a greater photochemical efficiency. This effect, added to a possible improvement in the carbohydrate partition observed in this and other species by our group, would be responsible for the greater berry size promoted by a greater production and transport of assimilates.

197.**VARIATION IN THE COMPOSITION OF ESSENTIAL OIL OF LIPPIA TURBINATA “POLEO” (GRISEB.) IN DIFFERENT CONDITIONS**

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The «Poleo» is a shrub needed in popular medicine and for the elaboration of liquors and compound herbs, is pressed by overexploitation. His spread has been studied and in this work there was studied the composition of the essential oil (AE) in plants that are cultivated in the highland locality of Juana Koslay, San Luis (33° 15' lat. Sur; 66° 12' long. Oeste). With plants obtained of seed (year 2003) culture began in two conditions: 1- dryness and 2- Irrigation + organic fertilization. In December, 2005 leaves and branches took, of the cultures 1, 2 and of plants that grow wild in the zone, for AE cuali-quantitative determination. The samples were revealed by steam dragging, with column and chamber of extraction and they analyzed for CG-FID-MS using an equipment Perkin Elmer model Clarus 500. The most representative compounds in the AE were: cis- α -bisaboleno, limoneno, β cariofileno, Germacreno D, in each of the cases, with the following percentages, respectively: 1- Dryness: 22,25%; 19,14%; 17,31%; 10,4%. 2- Irrigation: 17,98%; 11,96%; 10,82%; 16,17%. 3- Wild: 9,26%; 24,10%; 12,67%; 6,6%. The presence was not detected of lippiona not dihidrolippiona, considered scoreboards filécticos of poleo in the Argentine north. The environmental conditions affect AE's composition in poleo.

198.**HISTOPATHOLOGY OF ROOTS OF FOUR PEPPER CULTIVARS ATTACKED BY THE PHYTOPHAGOUS NEMATODE *NACOBBUS ABERRANS***

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The nematode *N. aberrans* damages pepper crops (*Capsicum annuum*). The reaction of the radical tissues of commercial pepper cultivars (AF 1434 and AF 2191) and experimental pepper cultivars (AF 6203 and AF 6185) to an attack of a parasite population from El Pucará del Aconquija, Catamarca, was evaluated. All the cultivars, except from AF 1434, carried genes resistant to *Meloidogyne* spp. Tomato plants (*Lycopersicon esculentum* cv. Platense) were used as control. Root pieces with and without galls were processed through traditional histological techniques until serial sections were obtained. The analyzed vegetables showed the formation of syncytia (nematodes' feeding sites) which developed in the tomato's cortical zone and the tomato's central cylinder but only in the pepper's central cylinder. The commercial cultivars showed thickened cell walls and bigger syncytia. The vascular tissues were notably reduced in AF 2191 and their cell walls were interrupted only in one restricted sector of the central cylinder. The nuclei reached a greater level of hypertrophy in AF 6185. No relevant differences between the alterations produced by the nematode in the analyzed cultivars were observed. In all the cases *N. aberrans* reached a complete life cycle, even in those hosts which carried genes resistant to *Meloidogyne* spp.

199.**USE OF MEDICINAL PLANTS IN URBAN AREAS OF SOUTHERN CÓRDOBA PROVINCE**

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Native medicinal plants have been gathered at Córdoba province (Argentina) without any regulations and its conservation status is now strongly affected. Primary use of medicinal plants is consumption by people and secondary, they are sold to shops, local market and traders from other provinces. Objective: to know the use of medicinal plants in urban areas of cities and towns at Córdoba province (Argentina). Methods: the ethnobotanical information has been surveyed by interviews. In 2006, the authors, together with specially trained research assistants, surveyed people in from 8 departments of Córdoba province. Interviews were open-ended in order to understand the general pattern of use. Results: 233 members of different age groups were interviewed. In total, 94 species from 30 vascular plant families were cited as medicinal plants. Most representatives families were *Lamiaceae* (14%), and *Asteraceae* (13%). Only 35 species from the total were natives. It has been mentioned 15 different types of uses: infusion (58.33%), local baths (7.64%), and tisane (7.64%). Species more frequently used were *Tilia cordata* (tilo), *Peumus boldus* (boldo), *Matricaria chamomilla* (manzanilla). Native plants are not the main used medicinal plants at urban areas, contrary the exotic are the preferred species. This reinforced the idea that market demands is the main cause of erosion of native medicinal germoplasm.

200.**INCORPORATION OF NATURAL ANTHRAQUINONES IN VERO CELLS**

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Since anthraquinones (AQs) isolated from *Heterophyllaea pustulata* (Rubiaceae) have demonstrated virucidal effects against herpes simplex virus type I (HSV-I) “*in vitro*”, we studied the ability of these compounds to enter into eukaryotic cells, by using a Vero cell line. This experiment would allow to estimate the potential antiviral effects of these natural AQs.

We started to test soranjidiol (AQ1) because is one of the most frequently found AQs in this vegetal specie. The cells were incubated with AQ1 during 30 minutes at 37°C, including controls (n=2). Then, samples were centrifuged and the pellet obtained was resuspended in CHCl₃ and it was frozen and defrozen abruptly in order to produce the disruption of cell membranes with the aim to release the intracellular AQ. It was centrifuged again and the AQ1 concentration was measured in the supernatant (CHCl₃) by UV-visible spectrophotometry at 268 nm. We determined that AQ1 entered into the cells in 29.1 ± 1.8%.

This result, along with the fact that the studied AQs have similar partition coefficients, would allow to presume that the other AQs isolated from *H. pustulata* could have similar behavior regarding their ability to enter into Vero cells; a necessary attribute to achieve antiviral action. This result encourages us to research the potential antiviral effects of these natural AQs.

201.**QUANTITY AND QUALITY OF SALIVA AND ITS RELATIONSHIP TO CARIES RISK**

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Saliva has a very important function related to caries activity a) formation of acquired salivary pellicle b) buffer capacity, that links to mouth PH and to the degree of acidity and alkalinity c) dilution of sugar in saliva, related to salivary flow d) capacity of remineralization according to the quantity of saliva in the mouth. The aims of this work were the following: to relate the function of saliva to caries activity; to determine the quantity of saliva present in the mouth and its relationship to caries formation; to describe the different ecological niches present in the mouth. Samples of saliva were collected from a population of 100 children aged 6 to 12 years old who were assisted at the School of Dentistry of La Plata. The Snyder method was used to determine the quality of the saliva (acid or alkaline) according to the speed in the shift from green to yellow colour. The quantitative method used was the recounting of colonies of Streptococcus mutans and of Lactobacillus. Results showed qualitative register: 70% susceptible; 19% moderate; 7% slight; 4% null. Quantitative register of Streptococcus mutans evidenced: 56% very susceptible patients; 20% susceptible, 17% moderate and 7% slight, register of Lactobacilli being 7% very susceptible, 73% susceptible, 20% moderate. The data obtained revealed the relationship between caries susceptibility as well as the quantity and quality of saliva.

202.**NORDIHIDROGUAIARETIC ACID ISOLATED FROM *Larrea divaricata*: BIOACTIVITY AGAINST *Candida spp.* FROM HUMAN CLINICAL SAMPLES**

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Plants constitute the principal source for the search of new drugs with pharmacological activity. Nordihidroguaialetic Acid (NDGA) of *Larrea divaricata* exhibits antioxidant, antibacterial and antiviral action. The aim of this work was to determine antifungal capacity against *Candida spp.* strains. This property was evaluated by microdilution technique, M27-A on 6 strains of *Candida spp.* isolated from clinical human cases of vaginal, respiratory and urinary candidiasis. Minimal Inhibitory Concentration (MIC), 50 % Inhibitory Concentration (IC50) (software Origin 7.5) and Minimal Fungicidal Concentration (MFC) were determined. All assays were carried out by duplicate. The ten concentrations (50-500 μ g/ml) that were analyzed inhibited all the strains in different grades. The IC50 had values between 217 and 405 μ g/ml and they revealed that strains number 4 (sputum) and 6 (vaginal exudate) were the most sensitive, and strain 2 (urine) exhibited the greatest resistance to the compound action. Values of MFC for any strain were not found. These results permit conclude that NDGA exerted fungistatic action for all strains studied.

203.**IN-VITRO EVALUATION OF A SELECTIVE CYCLO-OXYGENASE-2 (COX-2) INHIBITOR, CELECOXIB, AS A THERAPEUTIC ALTERNATIVE FOR ENDOMETRIOSIS (EDT)**

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Celecoxib is a specific inhibitor of COX-2. In cell lines from different types of cancer, as in animal models, it has been demonstrated that celecoxib inhibits cell proliferation, induces apoptosis and stimulates the expression of COX-2. The purpose of the present study was to investigate the effect of celecoxib on cell proliferation, apoptosis, expression of COX-2 and levels of VEGF and PGE₂ on endometrial epithelial cell cultures (EEC) from EDT patients EDT and controls. The EEC were performed from eutopic endometrium biopsies from 13 control women and 27 patients with untreated EDT, and were stimulated with different doses of celecoxib. Cell proliferation was evaluated by ³H-thymidine uptake and apoptosis, by the acridine orange-ethidium bromide technique. Expression of COX-2 was evaluated by Western blot and the levels of VEGF and PGE₂, in the culture supernatants, were measured by Elisa. Celecoxib 50, 75 and 100 μ M diminished significantly cell proliferation and enhanced apoptosis, in EEC from patients with EDT and controls. Likewise, 100 μ M of celecoxib significantly induced expression of COX-2 ($p<0.05$ vs. basal without celecoxib) in cultures from patients with EDT. Furthermore, concentrations of 25, 50 and 100 μ M of celecoxib inhibited secretion of VEGF ($p<0.001$, $p<0.05$ and $p<0.001$ vs. basal, respectively) and PGE₂ ($p<0.001$ vs. basal in all cases) in cell cultures from patients with EDT. Our results show that celecoxib is an effective inhibitor of cell proliferation, enhances apoptosis, induces COX-2 expression and inhibits the secretion of the inflammatory and pro-angiogenic molecules PGE₂ and VEGF.

204.**OLFACTORY AGNOSIA ASSOCIATED TO NEUROBIOLOGICAL PHENOTYPE IN FAMILIAL SCHIZOPHRENIA**

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INTRODUCTION. Olfactory identification deficits with intact acuity (olfactory agnosia) have been reported in schizophrenia. Functionally or structurally abnormal brain regions in schizophrenia overlap with major olfactory systems (e.g.: orbitofrontal and entorhinal cortex, dorsomedial nucleus of the thalamus, etc.). Olfactory identification (a measure of brain function) may provide important insights into the neurobiological phenotype of familial schizophrenia. **AIM.** To determine whether olfactory identification deficits are present in both psychotic and nonpsychotic members of multigenerational families with familial schizophrenia.

METHOD: The University of Pennsylvania Smell Identification Test (UPSIT) was administered birhinally to three groups of subjects (27-63 years): 11 psychotic (SP) and 21 nonpsychotic (SN) members of families with familial schizophrenia and 32 age- and sex-matched healthy volunteers (H). **RESULTS.** SN (nonpsychotic) individuals had significantly higher UPSIT scores than SP (psychotic) individuals but were impaired relative to healthy subjects. Group differences could not be accounted for by age, sex or smoking status. Based on UPSIT scores hiposmia was found in (% of subjects): 67% of SP, 48% of SN and 10% of H $\chi^2=6.8$, df=2, $p<0.03$. Olfactory performance was related to psychiatric diagnosis (psychotic vs nonpsychotic) $\chi^2=4.3$, df=1, $p<0.04$ and it was independent from family membership $\chi^2=0.96$, df=1, n.s. There was no interaction between family membership and psychiatric diagnosis $\chi^2<0.008$, df=1, n.s. **CONCLUSIONS.** Impaired olfactory deficits may add up in members of families with schizophrenia and they may indicate predisposition to psychosis in nonpsychotic familial schizophrenia-related individuals.

205.**OLFACTORY DEFICIT UNDER GENETIC INFLUENCE IN HUNTINGTON'S DISEASE?***Otero Losada M.**Neurociencias-CONICET. Hospital de Clínicas-Universidad de Buenos Aires. E-mail: mol@fmed.uba.ar*

INTRODUCTION. There is evidence supporting a strong relationship between neurodegenerative diseases and olfactory alterations. Hereditary factors related to neurodegenerative processes might increase the risk to suffer from these illnesses. However, whether olfactory deterioration associated to neurodegenerative decline may be linked to genetic factors as well is not clear. **AIM.** To evaluate olfaction in Huntington's offsprings. **METHODS.** The University of Pennsylvania Smell Identification Test (UPSIT) was administered to: eight subjects with Huntington's (**H+**), nine subjects without Huntington's (**H-**), ten hiposmic Huntington's offsprings (**h+**) and thirteen normosmic healthy subjects' offsprings (**h-**). **RESULTS.** Scores for the respective groups were (media \pm SD): 30 \pm 3 (**H+**), 37 \pm 2 (**H-**), 36 \pm 3 (**h+**), 38 \pm 1 (**h-**), [F_{group}(24.084;3;36), p<0.001]. **DISCUSSION.** Huntington's subjects showed poorer olfactory performance compared to neurologically healthy subjects. No differences in UPSIT scores were found between hiposmic Huntington's offsprings and normosmic healthy subjects. **CONCLUSION.** These data, obtained from a small number of cases, suggest that olfactory deterioration associated to neurodegeneration might not be under genetic influence in Huntington's disease.

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Nordin S, Paulsen JS, Murphy C: Sensory- and memory-mediated olfactory dysfunction in Huntington's disease. Journal of the International Neuropsychological Society 1995; 1:281-290.

206.**TERATOGENIC EFFECTS OF PHENOL IN EMBRYOS OF *BUFO ARENARUM****Paisio C, Agostini E, González P, Rivarola V, Bertuzzi M.**UNRC. Río Cuarto. Cba. E-mail: mbertuzzi@exa.unrc.edu.ar*

Phenol and its derivatives are thoroughly used in the industry and they have a high potential toxicity to animals and plants. In previous studies we demonstrated that phenol's solutions in concentrations from 100 to 250 mg/l produced mortality in metamorphic embryos of *Bufo arenarum*. The aim of the present work was to determine the teratogenic effects of phenol in premetamorphic embryos of this amphibians. The embryos were obtained by *in vitro* fecundation and they were exposed to solutions of phenol of 0 (control) 25, 50, 100, 150 and 200 mg/l for 4 days, to analyze malformations and behavior changes. The observed effects were: alterations in blastulae and gastrulae, irregular forms, reduction of the body, edema, absence of eyes, damages in the blastoporo, absence of the gills, deviations of the axis, necrosis, circulate swim, cutaneous alterations, and acephalism. In the concentrations of 25 and 50 mg/l prevailed the malformations in blastulae and gastrulae, deviations of the axis, damages of the blastoporo and acephalism while in 100 mg/l solutions, damages in the blastoporo, bend and reduction of the axis and acephalism were observed. In 150 mg/l, the most frequent alterations were the bend of the axis, widespread edema and irregular forms. The concentration of 200 mg/l produced 100% of mortality. The results showed that phenol produced teratogenic effects from the smallest concentration used and that the premetamorphic stages possess more sensibility than the metamorphic stages.

207.**INTRABURSAL ADMINISTRATION OF ANPT1 Ab INCREASES THE LEVELS OF CASPASE-3 AND AFFECTS THE STEROIDOGENESIS IN THE OVARY FROM HIPERSTIMULATED RATS***Parborell F, Abramovich D, Tesone M.**IByME-CONICET.*

In the adult, the angiogenesis is infrequent in physiological conditions except in the female reproductive tract. While VEGF is the principal factor in the angiogenesis; the maturation and differentiation of the vascular net require the coordinated action of the angiopoietins (ANPT1 and 2). In previous studies, we showed an increase in the number of atretic follicles and in the % of follicular apoptotic cells after the administration of the ANPT1 neutralizing Ab. The objective of this work was evaluate the role of ANPT1 on apoptosis, angiogenesis and ovarian steroidogenesis. We used prepuberal rats treated with eCG. ANPT1 Ab (10ng/ovary) was injected in one ovary and the contralateral was injected with IgG (C). The rats were sacrificed 48h post injection. The ovaries were extracted for IHQ for von Willebrand factor (endothelial cells marker) and for measurement sexual steroids. In addition, the antral follicles were isolated for western blot of caspase-3. The group treated with ANPT1 Ab showed increased levels of p17 caspase 3 active fragment (p<0.05). For RIA assay, it was observed that ANPT1 Ab produced an increase in androsterone (p<0.05) and a decrease in estradiol follicle levels (p<0.05). Besides, to evaluate the vascular development, it was performed IHQ for von Willebrand factor. The results were not showed significant differences in the % of endothelial cell area in both experimental groups. In conclusion, these results suggest that the apoptosis produced by ANPT1 inhibition is partially mediated by caspase-3 activation and by an increase in the ratio androgens/estrogens. These changes would be responsible by the increased number of atretic follicles observed in this model.

208.**STUDY OF ACUTE TOXICITY OF *Larrea divaricata* Cav METHANOLIC EXTRACT IN MICE***Pedernera AM¹, García Aseff S¹, Guardia T¹, Guardia Calderón CE², Pelzer LE¹.**¹Farmacología, ²Bromatología. Fac. Qca. Bqca. y Fcia. Univ. Nac. San Luis. San Luis 5700. E-mail: tguardia@unsl.edu.ar*

Larrea divaricata Cav (Zygophyllaceae), commonly known as "jarilla", is used in popular medicine for its anti-inflammatory and anti-rheumatic properties. In previous studies we showed anti-inflammatory and antiulcerous gastric activity of methanolic extract of *Larrea divaricata* leaves in rat. (J. of Ethnopharmacology Vol. 105:415-420 (2006)). The aim of this study was to assess the acute toxicity in mice of methanolic extract from leaves of *Larrea divaricata* (*LdMEL*). Mice were fasted for 4 h and water *ad libitum* and received oral increasing doses (100, 500, 1000 and 2000 mg/Kg) of *LdMEL*. Animals were assigned to five groups of 6 mice each, both sexes. Four groups were treated with the extract and one group (control) received vehicle (propylenglicol). Animals were observed for 14 consecutive days to record body weight (1st, 7th and 14th day), mortality or other toxic symptoms. After 14 days mice were sacrificed. Kidney, spleen and liver were observed macroscopically and the relative weights (organ/body) were determined. No toxic symptoms (restlessness, respiratory distress, diarrhea or convulsions) or death occurred. There were no significantly differences in body weight during experiment (ANOVA test). Relative wet weight of organs have not showed significantly differences from control. In conclusion, under the present experimental conditions leaves methanolic extract of *Larrea divaricata* showed no toxicity.

209.**ANTIMICROBIAL RESISTENCE OF *STAPHYLOCOCCUS* STRAINS ISOLATED FROM DAIRY HERDS IN THE CENTRAL REGION OF CORDOBA**

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Staphylococcus aureus is the most important pathogen of bovine mastitis, even though in the last years others strains were frequently isolated from intra-mammary infections. Antimicrobial agents represent one of the main therapeutic tools in veterinary medicine to control and eradicate staphylococcal infections. Nevertheless, there are greater pressures from the regulatory institutions to reduce the use of antibiotic in animals of human consumption. In the present work, 48 strains identified as *Staphylococcus* spp. isolated from dairy herds in the central region of Córdoba, were analyzed respect to the profile of resistance to 7 antibiotics of frequent use in bovine mastitis. Antimicrobial agents tested were: penicillin (10 µg), streptomycin (10 µg), ampicillin/sulbactam (10 µg/10 µg), gentamicin (10 µg), eritromycin (15 µg), rifampicin (5 µg), and oxacilin (1 µg). The disc diffusion (Kirby-Bauer) test was used. The highest levels of antimicrobial resistance were found to penicillin (20%) and streptomycin (14%). The lowest resistance was detected in 5 (10%) and 2 (4%) isolates for ampicillin/sulbactam and rifampicin, respectively. No resistance was detected for oxacilin and gentamicin. All the resistant strains were isolated from clinical and sub-clinical cases of mastitis.

210.**ANTIULCEROGENIC ACTIVITY OF *BACCHARIS POLIFOLIA* IN DIFFERENT EXPERIMENTAL MODELS**

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Baccharis polifolia Griseb., placed in the Cuyo region, have been used in folk medicine for gastrointestinal disorders. The aim of this study has been to determine the protective properties of *B. polifolia* against gastric lesions in rats. The antiulcerogenic activity of *B. polifolia* was assessed on 3 different experimentally induced gastric ulcer models. Wistar rats were fasted for 24 h. Gastric lesions were produced with 0.6 N HCl, 0.2 N NaOH and 25% NaCl as necrotizing agents. The ulcerogenic agents were orally administered to the animals which had been previously (1h) treated with *B. polifolia* ethanolic extract (250, 125 and 62.5 mg/kg). The results were expressed as Ulcer Index from 0 to 5 (maximal damage). *B. polifolia* prevents the formation of gastric lesions only at doses of 250 and 125 mg/kg: 0.6 N HCl ($p<0.001$ and $p<0.01$ vs. damage control, respectively); 0.2 N NaOH ($p<0.01$ and $p<0.05$ vs. injury control, respectively); 25% NaCl ($p<0.001$ and $p<0.01$ vs. damage control, respectively). The experimental results demonstrate that *B. polifolia* protects the gastric mucosa against the lesions induced by 0.6 N HCl, 0.2 N NaOH and 25% NaCl. These facts support the use in traditional medicine of *B. polifolia* to treat digestive disorders.

211.**BIOCHEMICAL AND MOLECULAR CHARACTERIZATION OF β -LACTAMASES EXPRESSED BY *Enterobacteriaceae* IN ROSARIO**

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Extended-spectrum β -lactamases (ESBL) are frequently expressed by species of *Enterobacteriaceae*. It is of interest to study biochemical and molecularly the types of ESBL produced by *Klebsiella pneumoniae* (Kp) and *Enterobacter cloacae* (Ec) to analyze regional variations in the spectrum of activity on β -lactams (BL). During the period 2005-2006, 170 enterobacterial isolates from inpatients and 29 enterobacterial isolates from outpatients resistant to oxyimino- β -lactams were detected in Hospital del Centenario (Rosario). The greater incidence was in Unit of Neonatology with 37%. The tests performed were: a) double disk synergy test, b) detection of the genes *bla_{TEM}*, *bla_{SHV}*, *bla_{CTX-M}*, and *bla_{PER}* by PCR (Polymerase Chain Reaction) and by direct sequencing, c) plasmid analysis and transference of genes, d) isoelectric focusing and enzymatic measurement. In all the Kp we found the genes *bla_{SHV}*, *bla_{CTX-M}* and in some *bla_{TEM}*, while in Ec we detected *bla_{CTX-M}* and *bla_{TEM}*. All the genes were located in a high number pair of bases plasmid transferable to *E. coli*. Isoelectric focusing defined bands of pI 5.4, 7.6 and 7.8. The identification of different types of BL hydrolyzing enzymes and the presence of transferable plasmids in multiresistant isolates suggest that BL of current use produce a selective pressure and aid the permanence of genes of resistance in the regional area of the Hospital.

212.**MODIFICATION OF LIPID COMPOSITION OF ERYTHROCYTES EQUINES AFTER INTENSE EXERCISE**

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The erythrocytes are cells exposed to high oxygen concentrations, rich in polyunsaturated lipids and iron. They are evolve in an atmosphere where constantly are exposed to sources intra and extracellular of free radicals. Fatty acid studies have been made in order to elucidate the mechanisms through which the free radicals cause membrane injuries to erythrocytes of sport equines. In this work were studied equines erythrocytes (7 animals in three treadmill testing turns, every thirty days) exposed an intense exercise. The blood samples were taken before (one hour), during and later (5 minutes) of the maximum exercise. Then were determinate the fatty acid composition, the relation between fatty acids saturated /no saturated and unsaturation index. As result the relation between fatty acids saturated /no saturated and the unsaturation index between the samples taken during the exercise and post-exercise were found significant differences, being these last ones more affected, which would indicate that the erythrocytaries injuries do not happen at the moment of greater exigency but later to itself.

221.

D-ALANYL-D-ALANINE CARBOXIPEPTIDASE IS ASSOCIATED TO SALT TOLERANCE IN *Ochrobactrum sp.* 11aPríncipe A, Jofré E, Mori G.

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Salinity is a severe stress condition in agriculture, which results in a decreased productivity of crop plants and has a detrimental effect on soil-microorganisms. Recently, some effective salt-tolerant PGPR belonging to *Ochrobactrum* genus were isolated. These strains are able to promote the maize growth. Since salt tolerance in bacteria is a complex phenotype, in which many regulatory mechanisms are involved it is necessary to characterize in detail genes conferring a salt tolerant phenotype in these PGPR. Random transposon Tn5-B21 mutagenesis was used to generate salt sensitive mutants of *Ochrobactrum* sp. 11a. The obtained transconjugants were tested for growth in minimal medium supplemented with NaCl 300 mM. Chromosomal and plasmid DNA isolation, Southern blotting and the construction of recombinant plasmids were carried out as described by Sambrook *et al.* 1989. Mutants which showed a salt sensitive phenotype were selected for further genotypic characterization. DNA sequence analysis of the locus tagged in the mutant called 1a showed significant similarity with the *dacB* gene of *Bacillus suis* 1330 encoding an D-alanyl-D-alanine carboxypeptidase. DacB is a periplasmic protein involved in cell wall biosynthesis. It can be assumed that the adaptation to high salt concentrations involves modifications of the external cell layers. These alterations in the cell wall probably establish an enhanced diffusion barrier to reduce the influx of inorganic ions into the periplasm.

223.

ETHER STRESS: STUDIES IN DIABETIC MURINE MODELSPuig N, Montenegro S, Gayol M, Martínez S, Elena G, Tarrés M.

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Stress could be defined as the adaptive response of the body to stimuli interfering with internal homeostasis. Although ether anesthesia is widely used in experimental procedures, ether itself might activate the hypothalamus-hypophysis-adrenal axis and the sympathetic adrenal axis.

The effect of ether stress in genetically diabetic rats, eSS and eSMT, and α eumetabolic controls, was studied. Glycaemia was assessed pre (G0, mg/dl) and 120 min after glucose overload (G120) in 6 and 12 month-old rats. Data were analyzed by ANOVA followed by multiple comparison tests, and expressed as mean \pm standard error mean for each group.

In 6 month old-animals, higher G0 y G120 values were found after ether treatment both in eSS and in eSMT (eSS: G0=98 \pm 26, n=120 vs 130 \pm 19, n=10 p<0.001; G120=194 \pm 51, n=120 vs 233 \pm 46, n=10, p<0.05; eSMT: G0=143 \pm 27, n=45 vs 170 \pm 17, n=9, p<0.01; G120=224 \pm 35, n=45 vs 317 \pm 78, n=9, p<0.001). The eumetabolic line α , showed no changes in G0 (69 \pm 22, n=27 vs 88 \pm 11, n=8, p>0.05) and enhanced values in G120 (106 \pm 20, n=27 vs 171 \pm 14, n=8, p<0.001). Twelve month-old eSMT and α rats followed the same tendency.

In basal conditions, only eumetabolic α rats managed to maintain glucose levels post ether treatment. Moreover, ether stress resulted in changes in the glycaemic homeostatic control after glucose overload both in diabetic and non-diabetic lines.

222.

ETHANOL INDUCED CHANGES ON MIGRATORY PARAMETERS OF *IN VITRO* NEURAL PROGENITORS OF THE OLFACTORY INTERNEURONSPueta M, Rovasio RA.

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In the mammal forebrain, subventricular zone contain progenitors of the olfactory interneurons, this cells migrate during fetal and postnatal period to the olfactory bulb through the rostral migratory stream (RMS). It has been shown that ethanol induces changes in the pattern of migration of some neural cells. The aim of the present work was to evaluate ethanol's effects upon morphometric and dynamic parameters of *in vitro* RMS neuroblasts. RMS explants from rat fetuses at gestacional day 20 were used. Coverslips carrying cultures were mounted in a Sykes-Moore chamber and perfused with ethanol 150mM, 38mM or culture medium (Control). Cells parameters were determined after 2,5 hs of time-lapse videorecording. We found that neuroblasts exposed to 150mM ethanol doses, exhibited a decrease in velocity and a reduction of distances traveled, as well as significant changes regarding cell shape when compared with Control cultures. 38mM ethanol dose failed to exert significant effect. The results indicated that ethanol has a deleterious effect upon migrating bulbar interneurons that could affect the development of olfactory bulb during fetal period. Taking into account that precursor cells for the olfactory interneurons migrate during early development and throughout postnatal life, present results suggest that the critical period for ethanol-induced damage may to involve pre- and post-natal stages in the main olfactory bulb. Therefore, odor encoding and discrimination may be affected by ethanol throughout lifespan.

224.

RETINOIC ACID AND FATTY ACIDS: EFFECTS ON HUMAN BREAST CANCER CELLSQuiroga PL, Eynard AR, Valentich MA, Soria EA.

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Introduction: The unsaturated fatty acids (UFA) and the all-trans retinoic acid (RA) could play a modulating role on the carcinogenic process, but their combined use has not been completely studied. **Objective:** To modulate tumor parameters in ZR-75-1 cells by RA and/or UFA (families: n-3, n-6 y n-9). **Methodology:** The cells were treated with RA 1 μ M and UFA 50 μ M: eicosapentaenoic (EPA, n-3), γ -linolenic (GLA, n-6), oleic (OA, n-9) or eicosatrienoic (ETA, n-9). It was assessed cellular proliferation (CP: MTT assay), eicosanoid production (HPLC), lipid profile (GC) and E-cadherin expression (Immunocytochemistry). **Statistics:** ANOVA + LSD Fisher test (p<0.05). **Results:** The membrane lipid profile showed the incorporation of the corresponding UFA. The CP was diminished by RA (-34.5%) and the UFA inclusion enhanced this antiproliferative effect. There was a 60% correlation between the 12-HETE formation (12-LOX pathway) and the CP. The E-cadherin expression was increased mainly by the n-9 serie. **Discussion:** The antitumoral effect observed with RA+UFA (decreasing CP) and the increasing differentiation (E-cadherin expression) were associated differentially to the specific UFA used into a synergic interaction.

225.**CURCUMIN CYTOTOXIC EFFECT ON THE HUMAN BREAST CANCER CELLS ZR-75-1**

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Introduction: The curcumin (dipheruloylmethane), a food additive, is a polyphenolic compound obtained from *Curcuma longa* rhizomes, which is used as an antitumour agent. The tumour development depends on the misbalance between cellular proliferation and death. **Objective:** To assess the cytotoxic effect of curcumin. **Methodology:** It was evaluated the curcumin effect (0, 5, 10, 20, 40 µM) with/out cupric sulphate (2.5, 10 µM) on: viability (MTT assay), LDH (lactate dehydrogenase) release and oxidant level (TMPD assay) of ZR-75-1 cell. Data were analysed by ANOVA followed by Tukey test ($p<0.05$). **Results:** The cellular viability was decreased by curcumin, without significant cupric effect. Also, it was inversely correlated with LDH release (Pearson Coeff.=-0.7). The cell cupric-induced oxidation was diminished significantly by curcumin. **Discussion:** Although curcumin acted as antioxidant against cupric sulphate, this plant polyphenol exerted a cytotoxic effect in dose-dependent way on the studied cell line with increasing cell death-associated LDH release.

226.**CHARACTERIZATION OF THE PROTEIN PROFILE OF STRAIN RC-98 OF BOVINE RESPIRATORY SYNCYTIAL VIRUS BY SDS-PAGE**

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Bovine Respiratory Syncytial Virus (BRSV) is an enveloped virus. Its genome is RNA and encodes for eleven proteins that migrate in SDS-PAGE with variations in their molecular weights (MW) according to the sub-group the strain belongs to. The objective was to identify the protein profile of strain RC98. The virus was purified in discontinuous sucrose gradients from which electroforetic runs were carried on. The gels were prepared according to the system of discontinuous buffer and gels in 12.5 and 15% polyacrilamide. The electrophoretic patterns obtained were characteristic of BRSV. In the 12.5% gel the following transmembrane proteins were identified: G-84kD, F0-77kD, and its cleaved polypeptides F1-46kD and F2-23kD. The ribonucleoproteic complex N-43kD, P-34kD and L-200kD were also observed. Matrix proteins M-29kD, M₂-22kD and M₂-11kD were visualized in the 15% gel. In addition another band of 18kD was identified that could be one of the nonstructural proteins NS1 or NS2, or the corresponding glycoprotein SH, which can appear glycosilated, or not with a MW that varies from 7.5 to 21kD. The main surface glycoproteins, such as G and F of the strain RC-98 were identified with neutralizing antibodies. On the basis of the pattern of MW of polypeptides obtained by protein F cleaving we suggest that the strain in study belongs to the intermediate sub-group (AB) of VSRB.

227.**IMMUNOCYTOCHEMICAL CHARACTERIZATION OF THE CYTOPATHIC EFFECT INDUCED BY THE BOVINE RESPIRATORY SYNCYTIAL VIRUS**

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Bovine Respiratory Syncytial Virus (BRSV) produces a cytopathic effect (CPE) characterized by intracytoplasmic inclusion bodies and syncytial cells which relates to its name. The aim of this work was to characterize the CPE of the BRSV strain RC-98 by immunocytochemistry in cultures of Hep-2 cells. Monolayers of Hep-2 cells were grown over coverslips, then infected with the BRSV strain RC-98. When the CPE affected extensively about 50% of the monolayers, the cells was fixed and immunocytochemistry was performed, using a polyclonal antibody anti-BRSV (VMRD) and commercial kit (Vectastain Elite ABC Vector). Viral specific signals were observed in the cytoplasm and nucleus of a high number of cells, and consisted in protein clusters of multiple irregular forms surrounded by a clear haul, corresponding to intracytoplasmic and intranuclear inclusion bodies. Another finding observed was the presence of syncytial cells which were very evident and showed specific signal in the cytoplasm of polykaryocytes. Intranuclear inclusion is very intriguing and appears to disagree with investigations on the replicative cycle of BRSV indicating that the events leading into viral replication occur in the cytoplasm, without involving the nucleus. Intranuclear inclusions can be a particular characteristic induced by the strain RC-98, nevertheless intranuclear inclusions have been also described for other viruses of the same family but not for the BRSV.

228.**REPRODUCTIVE CYCLE OF THE EUROPEAN HARE AND ITS RELATION WITH THE DATE OF COMMERCIAL HUNTING**

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The harvest of the *European hare* in the Pampean region of Argentina, is historically done during the months of May, June and July, which coincides with the date of hunting and the beginning of the reproductive period. The aim of this work was to analyze the reproductive cycle of the *European hare* through their gestational state and relate it to the present date of commercial hunting. In a slaughterhouse plant of Río Cuarto dedicated to the processing of hares for export, a study of the total number of hares was carried out during ten consecutive days (from the 15th to the 25th July, 2004). This observation was done by Veterinary. The total of examined hares 7315. 38% of the total (2780) were male; 61,7% adult females (4513) and 0.3% corresponded to young hare (22) that were not discriminated by sex. Of the total of adult females, 75% were pregnant (3385) with embryos in different periods of gestation. The percentage of pregnancy found in the month of July coincides with the findings of other authors, and with the great number of females during the period of study. The present date stipulated for the season of commercial hunting of the European hare in the Province of Córdoba would be affecting the reproductive cycle, it is proposed to begin a month before the habitual period (April) and to finish no further the last days of the month of June.

229.

TOWARD A OBESITY MODEL IN RATS. PRELIMINARY STUDIES*Razzeto GS, Escudero NL, Giménez MS.**Facultad de Química Bioquímica y Farmacia. UNSL. San Luis. E-mail: grazet@unsl.edu.ar*

It's known that hypercaloric diets provoke manifest obesity altering different metabolic parameters. The aim of this work is to induce an obesity model by the administration of hypercaloric diet AIN-93G modified (ED) (containing 34.14% sucrose, 42% of calories from fat, and 17% animal protein: casein). A lot of ten male Wistar rats (21 days old), animals were individually housed in stainless steel cages. They were initially fed with AIN-93G (CD) diet. After ten days of adaptation, they were separated in two groups: one was fed with CD and the other was fed with ED. The experience lasted 28 days. The animals were weighed and sacrificed and blood and liver were isolated. Total proteins (TP), (Biuret), Total lipids (TL), (Hara y Radin.), Total cholesterol (TC), free (FC) and esterified (EC), (Zak *et al.*), Triglycerides (TG), (Sardesai *et al.*) and Phospholipids (PL) (Rouser *et al.*) were determined. Increased parameters in ED group compared with CD group was observed: TL (mg/g) CD: 72.75 ± 10.48 ED: 95.28 ± 14.32, (P<0.05), TC (μg/g) CD: 623.11 ± 90.63 ED: 877.10 ± 51.43 (P<0.001) y FC (μg/g) CD: 47.54 ± 17.63 ED: 263.56 ± 34.54 (P<0.001), TG (μg/g) CD: 2021.56 ± 602.93 ED: 3190.66 ± 598.38 (P<0.05) y PL (μg/g) CD: 690.20 ± 129.77 ED: 962.024 ± 152.95. There were not change in serum lipid parameters and the body weight. These results indicate that feeding with hypercaloric diet produces changes on the liver lipid metabolism without phenotypic manifestation in the time of administration.

230.

PREVENTIVE ACTION OF NEONATAL GENE THERAPY WITH THE THYMIC HORMONE THYMULIN ON THE OVARIAN ATROPHY OF ADULT NUDE MICE*Reggiani PC², Flamini MA¹, Rodríguez SS³, Goy RG², Barbeito CG¹.**¹School Veterinary Medicine, ²INIBIOLP, Faculty of Medicine, National University of La Plata, Argentina.**E-mail: barbeito@fcv.unlp.edu.ar*

Congenitally athymic (nude) female mice show abnormal ovaries and a significantly reduced fertility. Recent evidence suggests that the absence of the thymic peptide thymulin in these mutants may be involved in the genesis of the reproductive derangements of these animals. Here we investigated the effect of neonatal thymulin gene therapy on the ovaries of adult nude mice. To this end we used an adenoviral vector harboring a synthetic gene for thymulin, or an irrelevant adenoviral vector used as control. Vectors were injected i.m. on postnatal day 1 or 2 and the animals were sacrificed at 70 days of age. Serum thymulin levels were measured by a bioassay. Nudes submitted to neonatal thymulin gene therapy did not show any deficiency in serum thymulin levels. Ovaries were processed for H & E staining. The gonads of the untreated nudes showed neither antral nor mature follicles and had scarce corpora lutea. In these ovaries, most of the follicles displayed different degrees of atresia. Untreated heterozygous (normal) females showed ovaries with normal follicles and corpora lutea. Nude mice submitted to thymulin gene therapy displayed gonads with intermediate characteristics, with some follicles displaying normal antra and corpora lutea. These normal follicles were fewer than in the normal mice. We conclude that neonatal thymulin gene therapy partially prevents the ovarian alterations of adult nudes.

231.

ENDOGENOUS JASMONATES IN SALINIZED SEEDLINGS OF PROSOPIS STROMBULIFERA*Reginato M, Abdala G, Miersch O, Luna V.**Fac Cs Exactas, UNRC, Río Cuarto Córdoba.**E-mail: vluna@exa.unrc.edu.ar*

Alterations in Jasmonic acid (JA) and its metabolism have been reported in plant tissues under salt stress; however, the response in halophytic plants is unknown. Endogenous levels of jasmonates and metabolites in the halophyte *Prosopis strombulifera* were analyzed in this work. Seedling were grown hydroponically in Hoagland solutions with addition of 50 mmol/l NaCl for NaCl treatment, 38 mmol/l Na₂SO₄ for Na₂SO₄ treatment and the isoosmotic mixture of salts for bisaline treatment every 48 h until final Ψ₀ = -1, -1,88, and -2,6 MPa were reached. Control plants were grown in Hoagland 25%. HPLC-GC-MS-SIM was used to separate and quantify the jasmonates. High content of the JA precursor, 12-oxophytodienoic acid (OPDA) and lower levels of JA were found in control roots. Under salinity, both compounds decreased with increasing salinity. In leaves, OPDA and JA levels were 4 fold lower than in roots, but the hydroxylated JA (11-OH-JA and 12-OH-JA) levels increased significantly in response to salinity, mainly 12-OH-JA. These results suggest that JA is not involved in salt tolerance, and the hydroxylation pathway would be an inactivation route of the JA signaling in this species, mainly in NaCl treated plants, confirming their increased tolerance to this salt in relation to Na₂SO₄.

232.

EPIDYDIMAL FLUID PROTEINS RELATED TO THE SPERM CELLS MATURATION*Rensetti DE, Monclus MA, Ortiz Maldonado V, Cabrillana ME, Vincenti AE, Fornés MW.**I.H.E.M. Facultad de Cs. Médicas, UNCuyo-CONICET. E-mail: mforne@fcm.uncu.edu.ar*

The epidydimal transit allows the maturation, protection and storage of the sperm cells until they are ejaculated. As part of these processes we verified the motility development and the sperm-association due to their interaction with the protein microenvironment that surrounds them, principally in the distal region (Tail) of the organ. By successive chromatography separation different protein fractions of caudal epidydimal fluid were isolated from adult Wistar rat. Then we incubated caudal sperm cells with the same protein fractions *in vitro*, to verify its capacity to maintain sperm motility and re associate epidydimal sperm cells forming rosettes. We detected that concanavalin A affinity fractions protein - with a MW between 80/45 KDa - (Fraction N° 2) is the most effective to re associate sperm cells into rosettes, while with minor to 40 KDa MW enriched proteins (Fraction N°4) participate to maintain sperm cells motility. Future efforts will conduct to isolated specific proteins form these fractions involved in sperm rosettes assemble and motility acquisition.

237.

EFFECT OF EXOGEN MELATONIN ON VISCACHA ADRENAL GLAND

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Arid and semiarid environments of subtropical climates are characterized by marked seasonal fluctuations of the natural photoperiod. Animals adjust their physiology and behavior to adapt to these variations along the year. The process of adaptation is mediated by the modulating action of the pineal gland through its main hormone, melatonin. In previous studies, we have shown that, during winter, the release of melatonin, corticosterone and adrenaline increases as a consequence of stress. The present study shows the results obtained from the administration of melatonin on cholesterol, and the release of glucocorticoids and adrenal catecholamines. A group of animals ($n=6$) was administered two daily doses of melatonin (Sigma-100 μ g/kg-subcutaneous administration) over nine weeks. The control group was only administered oleic vehicle. Experimental group: Tissular cholesterol (mg/100 mg) 16.51 ± 1.86 . Seric Corticosterone (ng/ml) 6.10 ± 0.57 . Tissular adrenaline (ng/g) 313 ± 53.1 . Control Group: Tissular cholesterol (mg/100 mg) 35.22 ± 2.25 . Seric corticosterone (ng/ml) 2.80 ± 0.38 . Tissular adrenaline (ng/g) 563 ± 35.7 . These results show that the administration of melatonin increases the secretion of corticosterone and adrenaline in a similar way to what was observed in habitat. In conclusion, the synchronizing action of the photoperiod, the modulating activity of melatonin, and the synthesis and release of corticosterone and adrenaline could be part of a complex system allowing wild animals to adapt to environmental changes, thus increasing the survival probabilities.

238.

DETERMINATION OF LYMPHOCITE PROLIFERATION AGAINST BOVINE AND EQUINE MILK PROTEINS IN ALLERGIC CHILDREN TO COW MILK, BEFORE AND AFTER A DIET WITH MARE MILK

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Many allergic reactions against cow milk (CM) are no mediated by IgE, so it is useful to evaluate cellular immunity. The lymphocyte proliferation allows detecting if there are sensitized T cells and allergens involved and it shows predictive effectiveness. The objective was to study in allergic to CM, indices of lymphocyte proliferation (ILP) to CM proteins and mares milk (MM) before and after a diet with MM. Twenty six allergic children of 6 months to 5 years old and 29 controls were studied. The lymphocyte proliferation by MTT assay with ConA, CM and MM was evaluated. The ILP obtained in cultures of lymphocytes before the diet were greater than that obtained ones after the diet ($p < 0.0001$), but without differences when the culture was added of MM. The results of cultures after diet with MM compared with those of controls showed significant differences against all used proteins, ($p < 0.0001$). The lymphocyte proliferation in allergic to CM could confirm hypersensitivity to the specific allergen and would be in agreement with the clinical symptoms. MM did not induce lymphocyte stimulation in allergic children to CM, which would be a formula suitable substitute, being able to be recommended to replace and to complete the infantile diet.

239.

NITRIC OXIDE-INDUCED CAPACITATION IS PARTIALLY MODULATED BY PEROXYNITRITE IN CRYOPRESERVED BOVINE SPERMATOZOA

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Sodium nitroprusside (nitric oxide donor) or SIN-1 (peroxynitrite donor) induce capacitation in cryopreserved bovine spermatozoa. Nitric oxide can activate guanylate cyclase or react with superoxide anion to produce peroxynitrite, leading to protein tyrosine phosphorylation or protein nitration, respectively. The aim of this work was determine the involvement of peroxynitrite in nitric oxide triggered pathways, during capacitation. Spermatozoa were incubated at 38°C in TALP medium with Ca²⁺ and BSA, during 45 minutes, with heparin (10 IU/ml), SNP (0,05 μ M) or SIN-1 (10 μ M) in the presence of different concentrations of uric acid (peroxynitrite scavenger). Capacitation percentages were determined by chlortetracycline fluorescence assay (CTC). Progressive motility and viability were determined by light microscopy and eosin/nigrosin technique, respectively. Uric acid (10 μ M) inhibited capacitation induced by SIN-1. Capacitation induced by heparin (37±4,9%) or SNP (31±2,74%) was significantly diminished by 10 μ M uric acid (15,4±2,7% and 15,4±3,58%; respectively). In the presence of uric acid, nitric oxide may still be inducing capacitation through guanylate cyclase pathway. Peroxynitrite is partially involved in mechanisms triggered by nitric oxide during capacitation induced by SNP or heparin.

240.

EXPRESSION OF VASCULAR ENDOTHELIAL GROWTH FACTOR IN PLACENTAE OF STRESSED RATS

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The vascular endothelial growth factor (VEGF) is implied in placental angiogenesis during normal development. The feto-placental unit is functionally similar to the hypothalamic-hypophysial system, supporting homeostasis during pregnancy. Any stressing situation affecting the mother could then induce alterations of VEGF expression in the different placental zones, with varied functional results. Our aim was to determine if immobilization chronical stress (IMO) results in alterations of VEGF expression in the rat placenta. Pregnant Wistar rats (200-300g) were used, divided into control (C) and stressed (S) groups, and the latter subjected to IMO for 45 min. The rats were sacrificed at 12, 17 and 21 days of pregnancy. The histological sections were treated with polyclonal anti-VEGF and revealed with DAB. More intense expression of VEGF with respect to control was found in giant throphoblast cells (GTC) of stressed rats at 12 and 17 days of pregnancy. No marks were found at 21 days in S or C groups. The observations that VEGF is highly expressed during pregnancy, especially in stressed rats GTC, could indicate the existence of a greater vascular development in these placentae, with effects in feto-maternal interchange.

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245.**ETHANOL DISTURBS THE ORIENTED MIGRATION OF NEURAL CREST CELLS***Sabatino ME, Jaurena MB, Battiato NL, Rovasio RA.**Centro de Biología Celular y Molecular, FCEFyN, UNCba. E-mail: jetama@hotmail.com*

The neural crest cells (NCC) of vertebrate embryos migrate and colonize specific sites by active and oriented migration, differentiating in many cell types. Besides the substrate composition, molecules released in their micro-environment contribute to modulate the speed and distance traveled (1), as well as directionality (2, 3) of this cell population. Data of our laboratory indicate that trophic factors *Stem Cell Factor* and *Neurotrophin-3*, as well as the chemokine *Stromal Cell Derived Factor-1* (SDF-1) induce a chemotactically oriented behavior of migrating NCC. It was also showed that morphogenesis of NCC are irreversibly modified by ethanol, supporting the essential participation of this cell population in the etiopathogeny of the *Fetal Alcohol Syndrome*. In the present work, mesencephalic NCC were obtained from stage 10-12HH chick embryos, then cultured during 24 hr, placed in chemotactic chambers with defined medium, exposed to SDF-1 gradient with 300ng/ml at de source, and registered by video-microscopy during 6 hs. The oriented migration towards SDF-1 was not observed when NCC were exposed to 150mM ethanol. These results indicate that ethanol disturbs chemotaxis-guided migration of cephalic NCC, without significant changes of cell morphology or dynamic parameters not involved in oriented cell migration, mechanism that could be relevant in the perturbed cell distribution of the *Fetal Alcohol Syndrome*.

(1) Alvarez et al., (2) Zanin et al., (3) Tolosa et al., in this meeting.

246.**MOLECULAR HLA-B27 DETECTION IN PATIENTS WITH SPONDYLOARTHROPATHIES***Salvarredi LA, Lacoste MG, Guzmán AMS, Di Genaro MS.**Immunology, FQBF, National University of San Luis. E-mail: sdigena@unsl.edu.ar*

The human leukocyte antigen (HLA) molecules are associated in many cases with autoimmune diseases. HLA-B27 is strongly associated with spondyloarthropathies (SpA) in which HLA-B27 is a diagnostic and prognostic marker. Therefore, HLA-B27 determination results of clinical interest in patients with rheumatologic diseases. The objective was to optimize an assay for HLA-B27 detection by molecular methods (PCR and real time- PCR) to be used in blood and synovial fluids (SF) of SpA patients. Two DNA salting out extraction methods were compared in blood and SF samples. A conventional salting out method and an optimized method (with NaClO₄) were used in 40 and 80 blood samples, respectively. The purity grade (DO 260/280) and the integrity were evaluated. The HLA-B27 genotypic determination was performed by PCR-SSP in clinical samples and in 100 aleatory selected blood samples from the Public San Luis Hospital. Co-amplification of HLA-B27 with β-globin gene as internal control was performed. DNA purity (DO 260/280) and integrity was higher in samples extracted with NaClO₄ method than those extracted with the conventional method. One of 100 blood samples (1%) resulted HLA-B27 positive. A SF sample from a patient resulted also HLA-B27 positive. This work contributed to optimize the molecular HLA-B27 detection for application in patients with arthropathies.

247.**FUMIGANT ACTIVITY OF ETHANOL AND HEXANE EXTRACTS FROM FRUITS OF *Schinus molle* var: *areira* (ANACARDIACEAE) AGAINST *Blattella germanica* (DICTYOPTERA, BLATTELLIDAE)***Sánchez Chopá C^{1,2}, Alzogaray RA³, Ferrero AA¹.**¹Dto. de Biología, Bioquímica y Farmacia, UNS. San Juan 670, (8000) Bahía Blanca. ²CIPEIN (CITEFA-CONICET), Villa Martelli, Buenos Aires. ³Becaria CONICET. E-mail: cschopá@uns.edu.ar*

The fumigant activity of ethanol and hexane extracts from fruits of *Schinus molle* var. *areira* were examined against adults' males of *Blattella germanica*. Filter paper strips (6x8cm) were impregnated with 1ml of different ethanol and hexane solution concentrations of the extracts or ethanol and n-hexane as control. After evaporating the solvent, the treated filter paper strip was placed at the bottom of 500ml glass vial. Six male adults of *B. germanica* were kept separately in small vials that had both open ends covered with a wire net. Each small vial was hung with thin metal wire at the geometrical centre of the glass vial and the glass vial tightly closed with the lid. The fumigant effect was evaluated after 72 h. The LC₅₀ of ethanol extracts from fruit was 93.31 mg/l of air whereas the LC₅₀ of hexane extracts was 300.05 mg/l of air.

248.**EFFECT OF DEHYDROLEUCODINE (DhL) AND 2H-DhL ON AMPHIBIAN OOCYTES MATURATION***Sánchez Toranzo G, López L, Bühlner MI.**Instituto de Biología UNT-INSIBIO.**E-mail: mbuhler@fbqf.unt.edu.ar*

In all animals so far studied, the activation of the maturation promoting factor (MPF) induces the nuclear maturation. In *Bufo arenarium*, this activation requires the dephosphorylation of Thr-14/Tyr-15 residues of the pre-MPF by the cdc 25 phosphatase. In this species, a small amount of cytoplasm containing active MPF is able to induce nuclear maturation in the immature oocyte by an auto-amplification loop of MPF. It has been demonstrated that the DhL block the progression of cellular cycle in G2. The objectives of this work were analyzed if the DhL and the 2H- DhL interfere with the activation of MPF and the nuclear maturation of *Bufo arenarium* oocytes and which signaling route would be affected. Nuclear maturation was induced in different experimental conditions: a) spontaneously, by removing follicle cells, b) progesterone treatment c) VG-content injection, d) mature cytoplasm injection. The results show that DhL and 2H-DhL induced inhibitions, in a dose-dependent manner, of spontaneous and progesterone-induced maturation. Nevertheless, DhL at the doses assayed had no effect on GVBD induced by mature cytoplasm injection but exerted an inhibitory effect on GVBD induced by VG-content. In addition, the treatment with H89 and OA suggest that the target of DhL could be some inhibitor of the pp2A phosphates, after the inhibition of PKA. In all cases 2H-DhL exhibited less activity and cytotoxic effect than DhL.

249.**LECTIN-BINDING PATTERN ON THE UTERUS OF SOWS WITH CYSTIC ENDOMETRIAL HIPERPLASIA AND ENDOMETRITIS**

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Surface glycoproteins that compose the glycocalyx of trophoblast and uterine epithelium are important to embryo implantation. Lectin histochemistry has been useful in the analysis of glycoconjugates of normal and pathological female genital tissues in domestic animals. In a previous paper we demonstrate changes in lectin binding patterns in the uterine tubes of sows with reproductive alterations. In the present work samples of uterus of 62 mature sows, with and without morphologic lesions were incubated with the biotinylated lectins: DBA, SBA, RCA-1, Con-A, UEA-1, PNA y WGA. Glandular and luminal uterine epithelium were observed. Cystic endometrial hyperplasia increased the RCA-I reaction only in the apical region of the glandular epithelium. There was higher intensity of marking of WGA in glandular epithelium in uteri with endometritis. In addition, increased Con A binding in the glandular epithelium and mild reduction of UEA-1 reactivity in the glycocalyx of the glandular epithelium were detected in endometritis. These changes were minor than those found in other species and might be involved in reproductive failure, once uterus may not be capable of providing the optimal microenvironment for an early embryo growth.

250.**TUBULIN-Na⁺,K⁺-ATPase INTERACTION IN HUMAN ERYTHROCYTES**

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Interaction between acetylated tubulin and plasma membrane Na⁺,K⁺-ATPase inhibits the enzymatic activity in different cells. In this work we determined the presence and nature of this association in membrane of human erythrocytes (MHE). Results indicated that MHE contain 4-fold less tubulin than brain plasma membrane and it is 50% less acetylated. The tubulin of MHE is a peripheral membrane protein, as was observed in other cells, probably by association with Na⁺,K⁺-ATPase since double immunofluorescence observed by confocal microscopy indicated that both proteins co-localize at the periphery of human erythrocytes. MHE were able to associate hydrophilic tubulin of brain rat and converted in hydrophobic component, and this association inhibit the Na⁺,K⁺-ATPase activity of the MHE. Biochemical experiments using isolated human erythrocytes demonstrated that L-glutamate and monensin, a Na⁺ ionophore, increase the Na⁺,K⁺-ATPase activity and decrease the amount of acetylated tubulin of the MHE. This effect is reverted by glucose addition. These results show that, in human erythrocytes, acetylated tubulin may be involved in the modulation of the control of Na⁺ and K⁺ transport by its interaction with Na⁺,K⁺-ATPase.

251.**RESPONSIVENESSES TO CAPACITATING CONDITIONS IN FRESH AND CRYOPRESERVED BOAR SPERM**

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Cryopreservation process results in increased sperm death due to plasma membrane damage and a decline in motility, which would seem to be an important reason for their relatively poor fertilizing potential. Oocytes can be fertilized only by capacitated sperm. Capacitation involves modifications in the plasma membrane and metabolic changes. Our aim was to evaluate the response of fresh and frozen-thawed sperm to *in vitro* capacitating conditions. Sperm samples were incubated with bicarbonate (40 mM) used as capacitation inducer. Aliquots were collected at 0, 15, 30, 45, 60, 90, 120 and 180 min. Pattern of sperm capacitation were determined by CTC staining. Sperm motility and viability were evaluated by optical microscopy and eosin-nigrosine technique, respectively. Motility, viability and intact sperm decreased significantly throughout the time, with significant differences between both treatments. Acrosome reacted sperm underwent a small but progressive increase in fresh semen, unlike the frozen-thawed one where there was a significant increase. While fresh semen showed a progressive increase in capacitated spermatozoa, ranging from low levels at the beginning of the incubation (3%) to 25% at 180 min; the cryopreserved sperm showed a fast increase at the beginning (12% to 28% at 30 min), but this proportion decrease (rapidly) to 9% at 180 min. Low fertility achieved with cryopreserved semen would be due to, not only, lower semen quality parameters, but also by premature induced capacitation.

252.**PURIFICATION OF A NOVEL SECRETORY PHOSPHOLIPASE A₂ ISOLATED FROM SOYBEAN SEEDS (*Glycine max*)**

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Phospholipase A₂ (PLA₂, EC 3.1.1.4) catalyzes the hydrolysis of phospholipids to obtain lysophospholipids. The purpose of this study was to purify an enzyme PLA₂ from soybean *Glycine max*. An homogenate of hydrated soybean seeds was treated at 95°C to inactivate proteases, delipidated with hexane, precipitated with cold acetone and centrifuged. To precipitate mostly high molecular weight proteins the supernatant was acidified at pH 4.5. It followed with an affinity chromatography using Eupergit-Cibacron Blue. The PLA₂ activity was determined by a turbidimetric method (ΔA_{340}) using soy lecithin as substrate and calcium as cofactor. The pool of active fractions was concentrated by lyophilization and then applied on Sephadex G-75 column. The active fractions were analyzed by SDS-PAGE and a principal band at 13 kDa was observed. The estimated pI was determined by isoelectric focusing giving an approximated value of 6.3. The molecular mass and the biochemical and catalytic properties (inactivation by Bromo Phenacyl Bromide, millimolar Ca²⁺ requirement, high stability) allow us to include this soybean enzyme in the secretory group of the PLA₂ superfamily.

253.

COMPARATIVE MORPHOLOGY OF THE REPRODUCTIVE FEMALE TRACT OF SUSCEPTIBLE AND RESISTANT***Fasciola hepatica* TO TRICLABENDAZOLE**Scarella S, Felipe A, Alzola R, Reeberg M, Solana H.

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Fascioliasis is a zoonosis of subtropical and tropical regions, with great incidence in suburban population and great economic impact in cattle production. For its treatment triclabendazole (TCBZ) is used. Its mechanism of action consists in the despolymerization of *F. hepatica* microtubules with the consequent loss of function, loosening and death. Today, the indiscriminated use of these drugs has generated the expression of the anthelmintic resistance phenomena (loss of ovicide effect, among others). In the present work the structure of certain components of the female reproductive apparatus of two strains of *F. hepatica* was analyzed (TCBZ susceptible, Cullompton strain, and TCBZ resistant flukes, Sligo strain). Conventional histology and immunohistochemistry with anti α -tubulin was done for both strains. No morphological changes were detected between both strains, so it is possible to speculate that the resistance phenomena expression in the Sligo strain could be produced by modifications at genetic or metabolic level without detectable changes in morphological aspects. These results contribute to the understanding of the pharmacological activity of this type of drugs contributing to the understanding of the mechanism of resistance to TCBZ by trematodes.

254.

HISTOPATHOLOGICAL EVALUATION AND OXIDATIVE STRESS IN HUMAN PLACENTA OBTAINED FROM SPONTANEOUS ABORTIONSSgro P, Mañas F, Gorla N, Monti S, Aimar N, Lavaselli E, Palacios F, Lafalce M, Vivas A.

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The mechanisms involved in human spontaneous abortion remain unclear. In the present work the histopathological appearance and the oxidative stress level as Thiobarbituric acid (TBARS) quantitation and Superoxide dismutase (SOD) activity in placentae at term (PT) and those from spontaneous abortions (PA) between 3-12 weeks (n=12) were evaluated. All PA showed severe histopathological damage: immature gestational age villi (100%), degenerated syncytiotrophoblast (100%), high levels of leucocytes (60%) and fibrin deposits (45%). TBARS (nmoles/g) levels were $20,10 \pm 5,23$ in PT and $12,28 \pm 5,14$ in PA ($p < 0,05$). SOD levels were $21,48 \pm 7,7$ and $4,51 \pm 2,32$ respectively ($p < 0,05$). By 13th week the fully established blood flow caused a PO₂ raising, thus explaining the higher levels of TBARS and SOD in PT. PA histologic tissue degradation associated to an abnormal blood flow increasing the PO₂ in the early placenta, could lend to a possible oxidative imbalance due to the decreased SOD activity in PA. This study suggests a relationship between histopathological lesions and the oxidative stress down regulation in spontaneous miscarriages placentae.

255.

IS HOMA-IR A TYPE 2 DIABETES PREDICTOR IN FIRST DEGREE RELATIVES?Siewert S, Gonzalez I, Codazzi L*, Filipuzzi S*, Ojeda MS.

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Introduction: Insulin resistance (IR) is a decreased biological function of insulin and is involved in the etiology of Type 2 Diabetes Mellitus (DM2), coronary diseases and arterial hypertension.

Aim: to determine the Insulin Resistance Index by using HOMA-IR in first degree relatives of patients with DM2, with or without metabolic syndrome (MS).

Materials and methods: 132 non diabetic patients from two different locations in San Luis were evaluated (age $36,8 \pm 9,5$ years old, sex F/M 79/53). BMI, waist circumference (wc), blood insulin and glucose, BP and lipids were determined. MS was defined according to the IDF criteria. HOMA-IR was calculated according to the Matthews equation..

Results: w/MS vs w/oMS: IMC $30,4 \pm 5,1$ vs $24,6 \pm 4,4$, wc $96,7 \pm 12,1$ vs $78,3 \pm 8,8$, fasting blood glucose $98,1 \pm 7,7$ vs $93 \pm 6,1$ mg/dl, blood Insulin $16,2 \pm 13,2$ vs $4,8 \pm 4,1$ μ UI/ml, HOMA-IR $3,21 \pm 2,2$ vs $0,88 \pm 0,7$, Quicci $0,33 \pm 0,03$ vs $0,42 \pm 0,06$, triglycerides $155,5 \pm 11,2$ vs $139,3 \pm 13,7$ mg/dl, total cholesterol $206,14 \pm 48,8$ vs $182,47 \pm 37,2$ mg/dl. Significant differences between w/MS and w/oMS were found in all the determinations.

Conclusions: there is a strong association between HOMA-IR and MS in the first degree relatives of Type 2 diabetic patients in the studied populations. This suggests that HOMA-IR could be a predictive factor in the development of this disease.

256.

NO SURGICAL TREATMENT FOR REFRACTORY PERI-APICAL PERIODONTITIS. CLINICAL ALTERNATIVESiragusa M.

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Bacterial biofilms have been found to develop on root surfaces outside the apical foramen and are associated with refractory peri-apical periodontitis and they do not heal despite the debridement of biofilm in the root canals and they need surgical treatment.

The aim of the present study was to evaluate clinical, and radiographic the effectiveness of interappointment medication as another therapy for chronic inflammatory periapical lesions. One hundred volunteer patients (n= 100), independent of age, race and sex, were enrolled in the study and their teeth were clinically judged as having refractory periapical periodontitis. Informed consent was obtained from all patients. All selected teeth, showed sinus tract and a periapical radiolucent area. All of them received endodontic treatment and were filled with iodine and calcium hydroxide as temporal medications. After 10, 15 or 30 days the canals were obturated with definitive material. The patients were recalled to be reevaluated every to 12, 24 and 36 months

The results registered had demonstrated that the 98% of the apical lesions were clinically and radiographically healthy .Recently, evidence has been found regarding the presence of bacteria within extraradicular areas and periapical lesions.

The excellent results regarding the effectiveness of iodine and calcium hydroxide interappointment dressing as an alternative endodontic therapy to the apical surgery.

257.**REACTIVE INTERMEDIATES AND ARSENITE CYTOTOXICITY ON CHO-K1 CELLS***Soria EA^{1,2}, Eynard AR^{1,3}, Bongiovanni GA^{1,3}.*¹*Instituto de Biología Celular, FCM-UNC. ²SECYT-UNC.*³*CONICET. E-mail: gbongiovanni@ceprocor.uncor.edu*

Oxidative stress has been increasingly recognized as a possible mechanism in the arsenic toxicity and carcinogenicity. Previous results showed a protective effect of flavonoids against *in vitro* arsenical cytotoxicity on CHO-K1 cells treated with 200 µM NaAsO₂ (As) and/or flavonoid (5 µM silymarin, S; 50 µM quercetin, Q). The objective was to evaluate stress markers induction and cell death by arsenite. It was studied: cellular viability (CV), hydroperoxides (HP, aqueous and lipidic), L-citrulline and JNK (c-Jun N-terminal kinase) activation. Statistical analyses were performed by the Info Stat 2007e.1 software ($p<0.05$): ANOVA followed by Tukey test and linear regression. As decreased CV 6.9% per hour ($R^2=0.89$, $p<0.05$) and increased HP after 2 hour exposition. Both flavonoids were able to counteract these effects. Preliminary results indicated early As-induced JNK activation (1-4 h). Taken together, results showed that, in the described conditions, the As cytotoxicity was time-dependent, promoting the cellular hydroperoxidation, without increasing the nitric oxide pathway. These effects were prevented by antioxidant flavonoids.

258.**PLASMATIC MMP-2 AND MMP-9 EXPRESSION IN PATIENTS WITH ENDEMIC REGIONAL CHRONIC HYDROARSENISM (HACRE)***Soria de Gonzalez AG, Arias NN, Tefaha L, Valdivia M, Guber RS, Bellomio C, Martinez M.**Facultad de Bioquímica, Qca. y Farmacia, Universidad Nacional de Tucumán, Tucumán, Argentina.**E-mail: gonzalez_stojan@uolsinectis.com.ar*

Metaloproteinases (MMP) are proteolitic enzymes involved in extracellular matrix components degradation (MEC), participating in processes where a lack of balance between synthesis and degradation of MEC occur. In this study, the expression of MMP-2 and MMP-9 through zimographic analysis was analysed in patients with HACRE. 25 healthy donors were analysed in this study, and also 8 patients with HACRE, 4 patients with chronic hepatitis C with increased MMP-2 and MMP-9 levels, which explains why the bands were used as a reference for their analysis. Euglobulins were prepared from plasma. Zimograms were done with Mini Protean III system (electrophoresis in 9% SDS-polyacrilamide gel with 0.2% gelatin). Gels were incubated for 20 hours in Tris-HCl and CaCl₂ buffer. They were marked with Coomassie Brilliant Blue R-250. The expression of proteolitic activity was determined by evaluation of non-marked bands. MMP-2 and MMP-9 activity in patients with HACRE was similar to that of normal controls. MMP determination would not be a useful marker for patients with skin clinical signs. Nevertheless, its usefulness should be studied in patients with preneoplastic lesions.

259.**BIOCHEMICAL EVENTS DURING EMBRYOGENESIS OF *DIPETALOGASTER MAXIMA* (HEMIPTERA: REDUVIIDAE)***Sosa L, Aguirre S, Canavoso L, Rubiolo E.**Dpto. Bioquímica Clínica, CIBICI-CONICET, Fac. Cs. Químicas, UNC, Córdoba. E-mail: laurasosa@mail.fcq.unc.edu.ar*

Like all oviparous metazoan, insect oocytes accumulate large amounts of nutrients in an organized manner inside the eggs. This stock-pile or "yolk" is critical since the egg can only survive if it contains enough nutrients to support embryonic growth, which occurs in isolation from the maternal body. In this work, we have investigated the biochemical events occurring during embryogenesis of *D. maxima*, a vector of Chagas' disease. The study included: [1] the changes in lipids, proteins and carbohydrates; [2] the regulatory mechanism involved in vitellin degradation, the main egg protein. Studies included chemical assays, western-blot analysis and the determination of acid hydrolase activities (acid phosphatase and cathepsins). Results showed that at the end of the embryonic development (day 28), there was a decrease in lipid and carbohydrate content (42% and 32%, respectively). It was also observed a minor content in proteins. Vitellin degradation during embryonic development was coincident with the increases of cathepsin and acid phosphatase activities. However, the increase in phosphatase was detected earlier. Finally, at the end of embryogenesis, vitellin represented about 30% of the total protein of embryo. The information derived from this study could be potentially applied in the future design of effective methods for the biological control of Chagas' disease vectors.

260.**EFFECT OF ASCORBIC AMPHIPHILIC DERIVATES UPON ERYTHROCYTE DEFORMABILITY***Spengler MI¹, Rasia M¹, Palma S², Allemandi D².*¹*Cátedra de Biofísica, Facultad de Ciencias Médicas, UNR.*²*Departamento de Farmacia. Facultad de Ciencias Químicas, UNC. E-mail: isabelspengler@hotmail.com*

Amphiphilic derivates from ascorbic acid (ASCn) show tensioactive behavior in aqueous solution, reason why they could be employed as solubilization agents of pharmacological drugs. The aim of the present paper has been to study the effect of ASCn ($n = 8, 10$ and 12) upon red blood cell (RBC) deformability. RBC extracted from healthy donors were suspended at 10% Ht in PBS (control) or PBS additioned with ASC₈, ASC₁₀ and ASC₁₂ at different concentrations, above and below the critical micellar concentration (CMC) [concentration at which the surfactant molecules begin to form aggregates or micelles], were incubated at 37 °C for 30 minutes. RBC deformability was estimated by its inverse function: the rigidity index (RI), which has been obtained from the flow time for 1 ml of RBC suspension passing through the filter 5µm pore size. The results showed a correlation between the RI and the employed concentration (to n=8: $r=0,77$, $p<0,001$; to n=10: $r=0,96$, $p<0,0005$; to n=12: $r=0,98$, $p<0,002$): Moreover, the RI values for concentrations < CMC did not significantly differ from the RI control value; although, for concentrations > CMC, RI values were significantly higher ($p<0,01$), to n=8, 10 and 12 than the control RIs. With such results as a ground, we suggest that ASCns in the micellar state interact with the constituents of the membrane, impairing erythrocyte deformability.

261.**REPELLENT ACTIVITY OF ESSENTIAL OIL FROM *Artemisia absinthium* (ASTERACEAE) AGAINST ADULTS OF *Tribolium castaneum* (INSECTA, COLEOPTERA: TENEBRIONIDAE)**

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The repellent activity of essential oil from *Artemisia absinthium* was evaluated against adults of *Tribolium castaneum*. The repellency test was conducted using an experimental arena consisting of five circular glass boxes, the central box was united to the other boxes with glass tubes, diagonally. The samples and the controls were distributed respectively in two opposed boxes containing 4 g of treated and untreated diet with solvent alone. The solvent used was hexane. In the central box forty adults were released. After 24 h the number of adults present at the boxes were counted. Each concentration was tested three times. Preference Index was calculated as PI= (% of insects at treated diet)-(% of insects at untreated diet) / (% of insects at treated diet)+(% of insects at untreated diet) where PI = -1 to -0,10, repellent plant; PI= -0,10 to +0,10, Neutral plant and PI= +0,10 to +1, attractant plant. At 0.04% PI was -0.42 and at 0.4 PI was -0.77.

263.**INFLUENCE OF TRANSPORT CONDITIONS OF OVARIES ON REDOX STATE AND *IN VITRO* MATURATION OF PORCINE OOCYTES**

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Reactive oxygen species (ROS) production have been associated with metabolic dysfunction and cellular damage. Transport conditions of porcine ovaries may affect oocyte redox state and maturation competence. The aim of this work was to evaluate oxidative activity and ROS production of immature oocytes according to different storage times and temperatures and its relationship with oocyte maturation. Ovaries collected from slaughtered gilts were transported at 2, 4, 6h and 15, 25, 35°C. Immature cumulus-oocyte complexes (COCs) were denuded and oxidative activity and ROS levels measured with 2,3,4,5,6-penta-fluorodihydrotetramethylrosamine and 2',7'-dichlorodihydro-fluorescein diacetate, respectively. COCs were matured in medium 199 for 48h and maturation evaluated by metaphase II. ROS levels increased when storage time or temperature increased ($p<0.05$). When storage time or temperature increased the oxidative activity diminished or increased, respectively ($p<0.05$). Maturation rates were higher with ovaries transported at short times and high temperatures ($p<0.05$). These transport conditions may induce high oxidative activity with moderate ROS levels in immature oocytes, contributing to maintain their maturation capacity.

262.**MOLECULAR STUDIES OF GLYCEROL-3-PHOSPHATE DEHYDROGENASE FROM *TRIATOMA INFESTANS***

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Triatoma infestans (*T. infestans*), Chagas' disease vector, acquires wings and the ability to fly after the last molt from fifth instar nymph to adult. The ability to fly is important for insect dispersion. In previous studies, in our laboratory, it was demonstrated that glycerol-3-phosphate dehydrogenase (GPDH), which is involved at glycerophosphate shuttle, increase its activity 30 fold in adults thoracic muscles. Adults muscles should have higher glycolitic and respiratory capacity to support fly activity. Electrophoretic studies from thoracic muscles showed two GPDH isoenzymes. Nymphs predominant isoform has less mobility. The aim of this work is to begin GPDH molecular studies. Using cDNA pools with degenerated primers, we amplified a 200 bp RT-PCR product from 1 to 3 days old *T. infestans* adults thoracic muscles. Upon sequencing and database alignments the fragment shows to be part of GPDH cDNA. Based on the sequence obtained, specific primers were designed for RACE experiment. We successfully amplified complete cDNA of two GPDH isoforms. Sequencing results allow identity, homology and conserved domains comparative studies with others species. GPDH isoform expression, studied by RT-PCR showed stage and tissue specific pattern.

264.**ORAL STRUCTURE AND ITS RELATION WITH *Heptapterus mustelinus* DIET**

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An organism feeding is related to its morphological oral structure. The main aim of this research is to analyse *Heptapterus mustelinus* oral morphology and to associate it with this organism diet. The examined sample was composed by 50 young and adult individuals as well as their oral pieces. The later were slides and coloured according to the Wassersung method (1976). Concerning the oral morphology; the mouse shape and position were analysed. Apart from this, the oral and pharynge's teeth like the gill's spine shapes, arrangement and size also evaluated. *H. mustelinus* adults as well as youngsters diet examination, show that this organism behaves as insectivore specie which mainly prefers Chironimidae like Trichoptera larvae. This correlates with this organism oral structure, which did not show any alterations during their development.

265.

Heptapterus mustelinus* DIET IN LERMA VALLEYS RIVERS AND STREAMS-SALTATerroba A, Vera-Mesones R, Romero M, Floriani G.**Universidad Nacional de Salta. Consejo de Investigación. Avda. Bolivia 5150, CP: 4400. Salta. E-mail: aterroba@unsa.edu.ar*

Heptapterus mustelinus, also known as "yusca" or "bagre anguila" is widely spread specie found in almost every river or stream in the province of Salta. The main aim of this research is to deeply analyse *H. mustelinus* diet. To fulfill this, 41 different samples were collected from Arenales river and some streams from Lerma valleys in different seasons. The alimentary canals were kept under formaldehyde to its 10%. All of the preys were taxonomically analysed in detail. The IRI (Importance Relative Index) was calculated to each alimentary item. The diet strictly examined was composed by: Diptera (Chironimidae 61,4% and Simuliidae 5,36%); Trichoptera (larvae 12,64% and adults 4,26%); Megaloptera 3,71%; Coleoptera (Lutrochidae 4,81%, as regards as adults, it is not determined yet); Plecoptera 1,37%; Odonata (Anisoptera nymph 5,36%). In the analysed Lerma Valleys rivers and streams, *Heptapterus mustelinus* behaves as an insectivore specie which mainly prefers Chironimidae as well as Trichoptera larvae.

266.

IN VIVO EXPRESSION OF ACTIVE MOLECULES IN CHEMOTACTICALLY GUIDED CELL MIGRATION*Tolosa EJ, Jaurena MB, Zanin JP, Battiatto NL, Rovasio RA.**Centro de Biología Celular y Molecular, FCEFyN, UNCba. E-mail: ezequieltolosa@yahoo.com.ar*

Chemotaxis modulates the accurate cell distribution in response to concentration gradient of molecules released by "target" regions. In our laboratory, we showed chemotactic response of *in vitro* neural crest cells (NCC) induced by Stromal cell-Derived Factor-1 (SDF-1) and Neurotrophin-3 (NT-3), as well as expression of some specific receptors on NCC. In this work, our main objective was to determine the *in vivo* expression site of these molecules. After training on *in situ* hybridization method with known embryonic models and probes, we studied the expression of SDF-1 and NT-3 mRNAs during early stages of chick embryos, as well as immunolocalization of the corresponding proteins.

Our results showed expression of SDF-1 and NT-3 in a cephalic region that is "target" of NCC population, which then differentiate in ciliary ganglion and other ocular and cephalic derivatives. The spatio-temporal expression of SDF-1 and NT-3 were consistent with our biological system, where NCC are oriented by gradients of these molecules.

The NCC ability to respond chemotactically to concentration gradients of molecules segregated by "target" regions extend our knowledge about the oriented migration of embryonic cells, supporting additional functions to chemokines and trophic factors.

267.

PREVAIL OF BACTERIAL PLAQUE IN PREGNANT IN THE FIRST THREE MONTH*Tosti S, Baudo J, Dominguez G, Di Salvi N, Allegretti P.**Physiology. F.O.U.N.L.P. 2006. E-mail: sobetis@yahoo.com.ar*

In gingival disease during pregnant there is an exaggerate inflammatory response of the gingivitis tissue because of the bacterial plaque. The response obeys to high seric levels of progesterone and estrogens which are current during pregnant period. The point in this research is to determine the prevalence of bacterial plaque in pregnant during the first three month. There were studied 60 women between 18 and 30 years old. Thirty were no pregnant (control group) and thirty were in the first three month of pregnant. The prevalence of bacterial plaque in no pregnant and in pregnant was, in both 100%. The grade of gingival inflammation were low and moderate. According to Silness and Loe low levels are between 0.1 and 1.0 and moderate are between 1.1 and 2.0. In the whole cases we found gingival inflammation. Nevertheless the analysis of prevalence according to severity levels (low, moderate and severe), show us that under same conditions of oral hygiene, low levels of gingival diseases prevail in no pregnant groups. The relationship between gingival alteration and increase of hormones results polemic. Although it is clear for Willerhausen. Samant and Cohen. O Neil put it in doubt.

268.

REPAIRING ACTIVITY OF THE BONE MARROW (BM) IN GASTRIC ULCER (GU) INDUCED BY ACETIC ACID (AA), IN RATS*Traverso M, Bedini O, Cesolari J, Naves A, Santos L, Zuivarra V, Laudanno O, San Miguel P, Palazzi J, Calvi B, Godoy A.**Experimental Gastroenterology, Medical School, UNR. E-mail: yo_mt23@hotmail.com*

Autologous BM has been studied in rats, injected via intra gastric mucosa to achieve repair of the GU. The aim was to study the action of BM repairing the antral GU induced by AA. In groups of Wistar rats, n=7, 200 g, deprived of food for 24 hours, except water ad libitum, anesthetized and with analgesic, a perforation was made in the tibia and extraction of BM was performed with a heparinized syringe, obtaining a sample for histopathologic and histochemical studies. After by laparotomy, BM was injected via intra antral mucosa. GU was induced with AA in gastric antral peritoneal subserosa. The laparotomy was closed, 3 days went by during which the rats received food and water ad libitum. After this they were sacrificed, the stomach was removed, the % of the area with lesions was established, obtaining slices for histologic and histopathologic studies. Statistical evaluation: "t" of Student and ANOVA. Results: Group A: physiological solution (1.0 +/- 0.1 mm), Group B: AA 25% (2.2 +/- 0.5 mm), Group C: AA 60% (4.2 +/- 1.5 mm) Group D: AA 100% (7.5 +/- 2.3 mm) Group E: BM (1.0 +/- 0.1 mm) Group F: BM + AA 25% (0.5 +/- 0.1 mm) Group G: BM + AA 60% (2.3 +/- 0.5 mm) Group H: BM + AA 100% (5.5 +/- 1.5 mm). It was concluded that autologous BM decreased the lesional area of the antral GU caused by AA in rats, helping the mechanisms of the tissue repair.

269.

TRYPANOSOMA CRUZI PRODUCE AN INCREASED DETACHMENT OF THE SYNCYTIOTROPHOBLAST IN VITRO, COMPENSATED BY TROPHOBLASTIC PROLIFERATION

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Trypanosoma cruzi can cause alterations in the placental tissue, and the congenital transmission might depend on the differential capacity of placentas to maintain the integrity of the syncytiotrophoblast (STB), the first placental barrier. Objectives: a) To analyze the effect of the infection on placental integrity produced by *T. cruzi* *in vitro*. b) To establish the capacity of the placenta to regenerate the STB. Material and method: co-cultures of human placental explants with trypomastigotes of *T. cruzi*, Tulahuen strain, and Lucky (congenital) for 24hs. Histological slices were stained: H/E, Hoesch and Immunofluorescence for cadherins P and E (cellular unions). HCG (RIA) which refers placental integrity was measured in culture media. Controls without infection. Significant detachment of the STB with proliferation of cytotrophoblast cadherins P and E positive and decreased values of HCG. Discussion: the infection by *T. cruzi* produces an increase in syncytiotrophoblast detachment corroborated by structural observation and decreased endocrine function (hCG), compensated by cytotrophoblast proliferation, that would be induced by the presence of the parasite and not by the invasion of the placental tissue. This phenomenon might participate in the pathogenesis of congenital Chagas disease. SECyT.UNC, SECyT UNLar.

270.

ANDROGENIC RECEPTORS IN COELIAC GANGLION

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Working with the coeliac ganglion-superior ovarian nerve-ovary (CG-SON-Ovary) system, we have shown, since a point of view physiologic, that androstenedione (A_2), at the end of pregnancy in the rat, would act through ganglionic androgen receptor. The objective of present work was to investigate the presence of such receptors in ganglion by means of immunohistochemistry. Female Holtzman rats at 21 days of pregnancy were used. The coeliac ganglion was removed and fixed in Bouin's fluid. The tissue was dehydrated in a graded series of ethanol, cleared in xylene and embedded in paraffin. Serial sections were cut at 5 μ m and mounted. Haematoxylin-eosin's staining and technique of immunohistochemistry were realized, antibody anti-androgen receptor N-20 sc 186 was used and the immunohistochemical visualization was carried out using the Super Sensitive Ready-to-Use Immunostaining Kit. The stained sections were examined under a light photomicroscope Olympus BX 40.

The results of haematoxylin-eosin's staining show that the coeliac ganglion presents a slim capsule of connective tissue. Numerous neuronal somas with basophilic cytoplasm and nucleus of dispersed chromatin with one or two evident nucleolus are observed. Respect of immunostaining, this is observed in scarce neuronal somas. The pattern cytoplasmatic of distribution is heterogeneous, presenting points marked intensely. We concluded that the immunohistochemistry study confirms the presence of receptors of androgens at cytoplasmatic level in neurons of coeliac ganglion.

271.

BIOLOGICAL RESPONSE OF BIOACTIVE GLASS-DERIVED GLASS-CERAMIC SCAFFOLDS IN CHICK EMBRYOS

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The use of chicken embryos for the study of the biocompatibility of materials is growing. The aim of the present study was to evaluate the biological performance of bioactive glass-ceramic scaffolds in chick embryos. Fertile eggs (*Gallus sp* Negra INTA) were used. The *ex ovo* embryo development was carried out as previously described by our laboratory (*Dev Biol* 2007; 306 Iss 1:407). Chick embryos were divided into two groups: control (C, n: 15), and experimental (E, n: 15). 45S5 Bioglass®-derived glass-ceramic scaffolds (5x5x2 mm) were placed on the chorioallantoic membrane (CAM) at 7 days of total incubation in embryos of group E. Embryos were monitored every 24hs and staged. The embryonic survival was estimated. All embryos were killed at 14 days of total incubation. The embryos were fixed, weighed, measured and processed for differential staining of cartilage and bone. No statistically significant differences in % of survival, stage of embryonic development or weight were observed between both groups. Body length was greater in group E embryos (43±2 mm) than in group C embryos (39±2 mm) ($p<0.05$). Histological analysis revealed that the epithelial tissue of the CAM developed a continuous interface with the biomaterial. No inflammatory response was observed. The control embryos exhibited a chondrogenic phenotype. Conversely, the experimental embryos exhibited ossification in the elements of the cranial and post-cranial skeleton.

272.

MURINE MODEL FOR THE GENETIC ANALYSIS OF THE HOST-PARASITE RELATIONSHIPS

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Murine models have demonstrated to be useful in biological studies. Four lines obtained by index selection from a synthetic mice population, CBi+, CBi-, CBi/L and CBi/C, and the control non-selected line, CBi, were evaluated in their behavior when challenged with different parasites to determine if these genotypes showed differences in their resistance. Natural (NE) and experimental parasitosis, using *Heligmosomoides polygyrus* (EE) and *Trypanosoma cruzi* (TE), were studied in adult mice of both sexes. The natural enteroparasites found in these lines were *Trichomonas muris* (*Tm*), *Spirotrichomonas muris* (*Sm*), *Syphacia obvelata* (*So*) and *Aspicularis tetraptera* (*At*). CBi/C was the genotype least resistant to *Tm*; CBi/C and CBi were the only ones to host this parasite. CBi- was the least resistant to *Sm*. *So* and *At* parasite load was different between sexes within genotype and among genotypes (CBi/L males were the least parasitized). CBi/L was also the most resistant genotype to both EE and TE. The aforementioned differences would be due to genetic differences among the lines, generated by selection and/or dispersive processes. It can be concluded that these lines constitute a useful experimental model to study the influence of the host genotype in the resistance/susceptibility to parasites.

273.

ANALYSIS OF *HELICOBACTER PYLORI* GENOTYPES AND THEIR ASSOCIATION WITH ANTIMICROBIAL SUSCEPTIBILITY AND CLINICAL OUTCOME

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The predominant *Helicobacter pylori* strains circulating among geographical locations differ in regard to genomic structure. The aim of this study was to analyze the antimicrobial susceptibilities of *H. pylori* and their association with virulence markers and clinical outcome. The isolates were characterized by antimicrobial resistance pattern, *cagA* status *vacA* s m alleles and *iceA* type. *H. pylori* resistant strains to antimicrobials were: 41.66% for Clarithromycin and 33.33% for Metronidazole. The virulence spectrum of the *H. pylori* strains showed 47.22% *cagA+*; 55.55% s1m1 genotype and 50.0% *iceA1* allele. Among clarithromycin resistant strains, the combination of *cagA+*, *vacA* s1m1, *iceA1* genotypes was significantly associated with patients with peptic ulcer disease (PUD) ($p < 0.05$). This study showed that isolates from patients with PUD were associated with specific virulent genes as well as clarithromycin resistance. This fact was of special importance because strains which were difficult to eradicate resulted more virulent. The risk predicted by such genotypes is based in the high toxicity demonstrated by *iceA1* and s1m1 isolates that contributes to the development of ulceration. There is a need for constant surveillance of both prevalence of the resistant strains and virulence markers among the *H. pylori* isolates that circulate in our community.

274.

A PHOTOSYNTHETIC MICROORGANISM IN DARKNESS

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This caenogastropod ampullariid snail lodges a cyanobacterium within its midgut gland's cells. Other ampullariid genera, including *Pila*, which is present in both Asia and Africa, also bear morphologically similar corpuscles. Though this cyanobacterial/ampullariid association may have occurred more than once in phylogeny, one would parsimoniously assume that a single episode of association has occurred, and that the symbiont has been living in darkness even before the fragmentation of Gondwana (>150 My). In free-living cyanobacteria, photosynthetic pigments capture light energy for the production of both sugars and oxygen, with the participation of the enzyme ribulose-bisphosphate carboxylase-oxygenase (RubisCO), so the question was raised to what extent these endocyanobionts may have preserved the photosynthetic hardware of their free-living ancestors. Pigments present in the endocyanobionts were identified as steryl-pheophorbides *a* and *b* (through HPLC, mass spectrometry and hydrogen nuclear magnetic resonance analysis). Also, by means of Western blot analysis with antibodies against RubisCO, two bands were detected in protein extracts. We suggest that this microorganism might fix CO_2 using RubisCO, although energy sources other than light should have to be used.

275.

DETERMINATION OF URIC ACID IN THE PREGNANCY AND ITS IMPORTANCE LIKE VALUE PREDICTS OF PREECLAMPSIA

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Preeclampsia is the main cause of maternal death. Hiperuricemia >55 mg/L differentiates this syndrome of all the other causes of HTA in the pregnancy. Increase of the uric acid is previous to measured elevations of creatinine or urea, and often before the development of HTA and proteinurea. Levels of uric acid usually diminish in the pregnancy due to increase in rate of glomerular filtration (GFR), for what you value superiors to normal stocking in embarrassed (38 mg/L), they indicate decrease of this, and possible hiperproduction on the part of the ischemic placenta. From January to May of 2007 they are study in the city of San Luis at 364 pregnant that converged to the laboratory, for the routine control, ages understood between 14 and 44 years. With posteriority to a fast of 12 hours, was carried out veined extraction. Limpid serum was obtained, free of hemólisis. Enzymatic method was used. Statistically: value superiors to the normal stocking (>38 mg/L), in 41/364 "11.3%" being 25/41 "61%" <30 years; and you overcomes the value I criticize (>55 mg/L) in 1/364 "0.3%" also <30 years, (whose case you correlates with creatinina >9 mg/L, and HTA through inter-later doctor consults). Uric acid it's an important marker and their precocious control in pregnant from early age it would contribute to a soon treatment, avoiding this way maternal and fetal complications.

276.

LIPIDIC PROFILE IN THE GESTATION FIRST MONTHS

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To ignore the increases in the lipidic profile (LP) it is the main one cause of risk of appearance of cardiovascular illness (ECV), especially in pregnant. In 2006 they are determined the lipidic profile at 249 embarrassed in their first trimester, ages understood between 15 and 42 years, and to who said exam was not requested in the order it prescribes. Veined extraction post fast of 12 hours; lipemics serums was not observed. Determination of total cholesterol, triglycerides, HDL-cholesterol and LDL-cholesterol for enzymatic methods according to Trinder, previous selective precipitation in two o'clock finishes. Statistic: mean, arithmetic mean, fashion and range. On the 249 samples, PL met with abnormal values (according to the III Panel of Experts) in: Triglycerides 10.4%, Total cholesterol 7.6%, HDL-cholesterol 14% and LDL-cholesterol 11.2%.

Determination	Mean g/L	Arithmetic Mean g/L	Fashion g/L	Range g/L
Cholesterol	1.62	1.6	1.5	1-2.6
HDL-Col	0.485	0.49	0.53	0.35-0.6
LDL-Col	0.98	0.93	0.8	0.49-1.99
Triglycerides	1.049	1	1	0.3-2.8

It would be important to include, in the pregnancy first trimester, the PL study like routine analysis, since their early knowledge would help to diminish the risk of ECV and this way to reduce the derived social and economic costs of its complications.

277.

SEROPREVALENCE OF CHAGAS DISEASE IN PREOCCUPATIONAL TESTS IN SAN LUIS CITY*Verdugo RA, Ampuero VE, Rodríguez GB, González LE.**Parasitología y Micología. Universidad Nacional de San Luis. San Luis, Argentina. E-mail: huergonz@unsl.edu.ar*

American trypanosomiasis or Chagas disease is usually asymptomatic, for this reason, its diagnosis is mainly based on laboratory tests. During the indeterminate and chronic clinical periods, detection of immunoglobulin (IgG) against *Trypanosome cruzi* by different serologic tests is the standard for diagnosis. In the present work we communicate the seroprevalence of *Trypanosome cruzi* antibodies in pre-occupational tests in workers of San Luis city. A total of 500 blood samples were analyzed. All samples were tested with indirect hemagglutination test and IFI assay. Samples reactive for two assays were considered positive. Serological evidence of human *Trypanosome cruzi* infection was demonstrated in 35 (7.0% / 93%) out of 500 individuals. In the 22-39 years age group the percentage of negativity was 94.5% / 5.5% and in the 40-71 years age group 90.5% / 9.5%.

- a) Prevalence in the 40-71 years age group was 73% higher in relation to that found in the 22-39 years age group ($p<0.05$).
- b) The prevalence shown no differences when compared male vs. female or place of birth.
- c) These findings show general infection prevalence in this community was similar to communicated in national average estimated rate (5.7%), (National Epidemiological Bulletins).

279.

YIELDS AND QUALITY OF FRUITS OF TOMATO CHERRY (*Lycopersicum esculentum* Mill. var. *cerasiforme* Alef.)*Villagra EL, Sluka E.**Dpto. Producción Vegetal. Fac. Agronomía y Zootecnia, Universidad Nacional de Tucumán. Tucumán. Argentina. E-mail: evillagra@faz.unt.edu.ar*

The tomatoes “mini” or “cherry” were positioned in the market and it interests penology, management and production of the crop. The objective was to compare and to evaluate yields and quality of fruits of cycle end, with two management systems in cultivation. One worked in El Corte -Tucumán. It was sowed in speedling with two systems: 1) greenhouse and 2) to field. In 1) transplant has more than enough borders covered with black plastic, nutrition with 18-46-0, I water for leak and it plants hung, $d=2,7$ plantas/m². In 2) transplant without conduction. In laboratory it was determined in mature fruits: soluble solids with manual refract meter (0-32°Brix), acidity in %acid citric fruit titrated with NaOH 0,1N and sugars reducers (Eynon Lane). It was evaluated: fruits/plant number; I weigh fresh of fruits (WFF) and dry weight of fruits (DWF); °Brix, pH, acidity and sugars reducers. He took samples at random 6 parcels, ANOVA was carried out and it arose that: among the two handling forms significant differences exist in fruits/plant number: being superior in plants taken place in greenhouse (29,8) regarding the handling to field (15,9). There is not significant difference in WFF neither in DWF. Concerning quality: the greenhouse fruits evidenced bigger content in soluble solids (6,8°Brix) compared with field fruits (5°Brix). The results are consistent with those obtained by Picha (1987), and they are directly associated with better flavour in fruits, parameters of quality and duration postharvest of the commercial product.

280.

GERMINATIVE RESPONSES OF *Atriplex crenatifolia*, (QUENOPODIAECEA) IN SALINITY CONDITIONS*Villarreal V, Rodríguez Rivera M, Sosa L, Moriconi J, Fernández E.**Proyecto P2-8305. Área Ecología. Universidad Nacional de San Luis. E-mail: mfrodr@unsl.edu.ar*

Atriplex crenatifolia is used as a native forage species in dry and saline environments of San Luis Province. The objective of this work is to study the germinative responses of his seeds to different salinity conditions with Na⁺ and K⁺. Harvested seeds in the Salinas del Bebedero, San Luis, which were threshed manually and scarified by 2'30" with H₂SO₄ previously to the test. The seeds were placed in Petri boxes and moistened with 5ml of NaCl; NaSO₄; NaCl+NaSO₄; KCl; KSO₄ and KCl+KSO₄ solution, in adequate concentrations to generate osmotic potentials of: 0 (control); -0.4; -0.8; -1.2; -1.5 and -1.9MPa. The test was made in stove of culture to 26±2°C. The germinated seed number was recorded and Percentage of germination (PG) and Germination Velocity Index (GVI) was calculated. The NaCl and NaSO₄ reduced the PG and the GVI significantly (36% y 55%; 8.22 and 8.80 respectively) from -0.4MPa; whereas with the combination of both, this parameters does not differ from the control (81%) but until -0.8MPa. The KCl and KSO₄ solutions affected the PG and GVI significantly from -0.8MPa (66% and 63%; 14.67 and 9.47 respectively) and the combination of these reduced the PG and IVG significantly from -1.2MPa (47% and 11.87.) The Na⁺ and K⁺ salts affect differently the germination of the seeds of *A. crenatifolia* being more negative the Sodium salt effect.

278.

CONDUCT OF *Cydia molesta* B. AND CARACTERISTICA OF THE DAMAGE PRODUCED IN SHOOT OF PEACH TREE*Viale S, Guevara E, Terenzzani P, Varsi E.**Dpto. Producción Vegetal, FAV. UNRC. Río Cuarto. Córdoba. E-mail: sviale@ayv.unrc.edu.ar*

Cydia molesta B. (Grafolita) is a pest of great importance in peach trees affecting shoot and then fruits. In shoot, al to be fed breaks apical dominance and affects the vigor. The objective of the work was to determine the population fluctuation of grafolita and to characterize the damage in shoot of peach tree located in different quadrants of the cup. The work was carried out in Río Cuarto, in 2005/06 season, in a 6 years old commercial orchard. 4 plants were selected at random in 4 varieties. In each plant two heights were delimited: upper and lower and two expositions: east, western. The pest was monitored with traps of sexual pheromone and the damage in shoot was determined monthly counting damaged shoots and totals by quadrant. Data was compared with ANOVA ($\alpha=0.05$). The grafolita captures occurred from September to end of March. 5 generations were determined in the season, occurring the overwintering at the end of September. The dates of occurrence of the 4 remaining generation peaks and the individuals captured in each peaks were: 21/11 (19), 23/12 (26), 16/01 (27) and 20/02 (16). The damage average found in shoot by plant was of 2,9% (14/12), 4,5% (10/1), 10,2% (14/2) and 14,3% (21/3). Statistically significant difference between heights was found but not among expositions. Damage is produced along the entire season, as well as there is new and scarcely hardened shoot.

281.**ABA SIGNAL: ROLE OF DGPP AND PHOSPHATIDATE PHOSPHOHYDROLASE**

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Abscisic acid (ABA) exerts an important role in seed germination since it inhibits the response to gibberellin (GA) in aleurone. Phosphatidic acid (PA) is an intermediary in ABA signal however diacylglycerol pyrophosphate (DGPP) role in germination processes is not clearly established. Here, we showed that PA produced by phospholipase D (PLD) during antagonist ABA effect in GA signal is rapidly phosphorylated by phosphatidate kinase (PAK) to DGPP. This fact is critical for aleurone since dioleoyl-DGPP exogenously added was able to inhibit the α -amylase secretion. Inhibition of PLD activity by 1-butanol during ABA treatment results in a normal secretory activity. This effect is overcome by addition of dioleoyl-DGPP. It also showed that ABA decreased the activity of Mg²⁺-independent and NEM-insensitive form of phosphatidate phosphohydrolase (PAP2). Using *Arabidopsis thaliana* Lpp protein sequences as queries it was identify two putative molecular homologues, which we term HvLpp1 and HvLpp2, encoding putative Lpps. The presence of well conserved structural Lpp domains in these sequences and the detection of protein by immunoblots suggest that both proteins are functional enzymes. Results showed are consistent with a role for DGPP a regulator of effect antagonist ABA in GA signal.

282.**CLINICAL AND SUBCLINICAL MASTITIS PREVALENCES AMONG DAIRIES OF VILLA MARÍA (CÓRDOBA) BASIN HERDS**

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Bovine mastitis represents an important loss to the dairy industry because of involuntary culling, treatment prescriptions, and milk production drop caused mainly by the subclinical form of the disease. In Argentina, no mastitis surveys is available under the current conditions of the dairy industry.

The aim of this study was to estimate the clinical and subclinical mastitis prevalence (P) among cows from Villa María dairy herds. To accomplish that, eight hundred fifty eight milk composite samples of cows from 20 radomly selected dairy farms were collected, between March and June of 2007. Subclinical mastitis (SM) was defined when pathogens were recovered from the milk sample and the somatic cells count was ≥ 250000 cel/ml. Subclinical and clinical (CM) mastitis gross prevalences were 46.8 and 2.5%, respectively. That represents a SM:CM ratio of 20 to 1. In addition, statistical differences on SM were observed between heifers and cows (49% vs. 26%). Also, a significant SM prevalence trend toward the end of lactation was observed. Regarding on CM prevalence, no trend by parity nor for lactation stage was found. These preliminary findings showed that mastitis remains challenging, specially due to the SM high prevalence.

283.***Melanoides tuberculatus* IN HIGH PARANÁ RIVER: INFERENCES OF ITS ORIGIN IN OLD WORLD FROM MITOCHONDRIAL RIBOSOMAL GENES**

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Introductions of invasive species have received particular attention due to the economic damage they cause, their impact on endemic species and their role in the transmission of parasitic diseases to humans. The parthenogenetic snail *Melanoides tuberculatus*, present in tropical waters of the Old World, has invaded several freshwater bodies in American Continent since 1950, from its two origins proposed in literature: Africa and Asia. The report of its presence in Paraná River in 1999, between Paraguay and Argentina, has extended its known southwest distribution area in the New World.

The phylogeography of this mollusk was examined to make a genetic characterization of the population recently discovered in Yacyretá Dam influence area. In this study, the population origin was evaluated trough the analysis of the sequence identity of two genes present in mitochondrial DNA. Partial sequence datasets from 12S RNAr and 16S RNAr genes were additionally verificated to test that variability at molecular level was adequate and useful for a reliable phylogenetic reconstruction by distance methods. The results from the sequence level in the studied population, permit us to establish a relation whose sequences identity, compared with samples over the world, reflects an Asiatic origin.

284.**IN VITRO EVALUATION OF *Larrea divaricata* AGAINST FUNGAL PHYTOPATHOGENS**

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In the biotechnological field, the sanity of plant crops of agro-alimentary impact is tried through organic practices that involve the use, among others, of plant extracts for plagues and diseases control without altering the environmental balance and human health. Objective: To determinate the *in vitro* antifungal activity of *Larrea divaricata* (Jarilla) Chloroform Extract (CE). Material and Methods: *CE Production:* The CE was obtained from plant shoot after both a hexan and a chloroform extraction, and it was dissolved in DMSO. *Antifungal activity:* It was evaluated on the growth diameter of *Fusarium solani*, *Fusarium graminearum*, *Fusarium verticillioides*, *Macrophomina phaseolina* and *Sclerotium rolfsii* until 144 h of incubation by dilution in potato dextrose agar supplemented with CE at a concentration of 1.000, 500, 300 and 100 μ g/ml. Result: All the CE concentrations inhibited the fungal growth, for *F. solani* in a range between 54-14%; *F. graminearum* 72-36%; *F. verticillioides* 80-25%, *S. rolfsii* 97-19% and 91-61% for *M. phaseolina*, this last one presented the highest inhibition in the lower evaluated concentration. Discussion: The CE of *L. divaricata* showed to be a potent antifungal agent against phytopathogenic fungi, which incentives further studies on *in vivo* systems.

285.**EFFECT OF TWO DIETS IN THE LENGTH OF LIFE AND TABLE OF LIFE IN *NEZARA VIRIDULA* VAR. *SMARAGDULA* (HEMIPTERA: PENTATOMIDAE)**

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The purpose of this study was to investigate the length of life cycle and horizontal table of life by age of the *Nezara viridula* var. *smaragdula* fed on two diets: A (fruits of *Phaseolus vulgaris*) and B (fruits of *Phaseolus vulgaris* + bud of *Glycine max* + cob of *Zea mais* + seeds of *Helianthus annuus*). One group of adults received diet A and other diet B under controlled temperature (28 ± 1°C), RH (60/70%), and photophase (14h). One hundred and ten eggs masses (10,200 eggs) were examined and its postembryonic development was observed. The length of life in days and mortality of the immature stages was recorded. Second instars from fifty five eggs masses were fed on diet A and an equal number were fed on diet B. Diet B significantly increased nymphal survival and reduced development time ($p<0.05$).

SECyT-UNS. PICTO-UNS N: 925.

286.**INTERACTIVE REGULATORY PATHWAYS CONTROL EXOPROTEASES PRODUCTION IN *Staphylococcus aureus***

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Staphylococcus aureus is a major pathogen of humans and animals that synthesizes a large number of extracellular proteins that play a role in bacterial virulence. Several global regulatory loci regulate the production of these virulence factors, among which the most relevant are the *agr*, *sar*, *sae* and *sigB* loci. Two-component signal transduction system, SaeRS, characterized in our laboratory, up regulates the synthesis of several exoproteins at the transcriptional level. *S. aureus* synthesizes several extracellular proteases. The synthesis of these proteases is positively regulated by *agr* and negatively regulated by *sae*, *sarA*, and *sigB*. In this study we examined how the interactions between these pathways affect proteases production. Total protease activity from concentrated supernatants of cultures in late stationary phase, measured using azocasein as a substrate, showed a 3- to 4-fold increase with the *sae* mutant as compared to the wild type strain. This activity increased about two and three fold with *sae agr* and *sae sigB* double mutants, respectively, than the *sae* mutant. These over-expression was visualized in sodium dodecyl sulfate polyacrylamide gels containing 0.1% gelatin. These observations indicate the existence of some type of additive and (or) synergistic effect between the two mutations that further increases the production of exoproteases.

287.**EXPRESSION OF SUBUNIT β 3 OF INTEGRINS DURING PORCINE PLACENTATION**

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The integrins are a superfamily of membrane glycoproteins, composed of α and β subunits, implicated in implantation of several species. The porcine placenta is epitheliochorial, non-invavise, folded and diffuse. To analyze the presence of placental integrins will allow us to understand the adhesion and implantation in this species. The aim of this study was to determine the presence of β 3 subunit in placental tissues of different gestational periods. Placental tissues from porcine females of 35, 60, 70 and 80 days of gestation, at term (114 days) and empty uterus were used. The presence of integrin β 3 subunit was analyzed by indirect immunocitochemistry. The fetal trophoblastic and maternal villi, uterine glands and blood vessels exhibited high intensity of the integrin β 3 subunit throughout the pregnancy. Fetal and maternal connective fibres showed moderate positivity. In conclusion, these results would suggest a probably role of integrin β 3 subunit in the events of adhesion of fetal trophoblast with endometrium during the gestation.

288.**CHARACTERIZATION OF LTCD8+ AND LEVELS OF IFNg IN ALERGIC CHILDREN WITH INFECTION OF VIRUS HERPES SIMPLEX TYPE I (HVS-I)**

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In children allergic manifestations like atopic dermatitis are associated frequently with viral affections, among them, those produced by Virus Herpes Simplex Type I (HVS-I). The objective was to investigate in allergic children with active infection by VHS-I, the cellular immunity, through the characterization and quantify of LTCD8+ and levels of IFNg. The patients were twenty two children of 5 to 12 years old, 14 allergic and infected with HVS-I and 8 without symptoms (controls). LTCD8+ were characterized and quantified with direct immunofluorescence from a culture of stimulated mononuclear cells with PHA. The levels of IFNg were quantified in the supernatants of lymphocytes stimulated with different antigens by the ELISA technique. The percentage average of LTCD8+ was 67.5% (61% - 74%) for in course infection (IC) and 44.5% (46% - 65%) for controls, ($p<0.0008$). The children with IC showed a higher levels respect to controls, ($p<0.03$). The active infection stimulates as the subpopulation of LTCD8+ than the production of IFNg which would demonstrate through its participation in the process of virus elimination.

289.**EMBRYONIC DEATH IN A MOUSE MODEL. OF BOVINE TRITRICO MONOSIS. RELATION WITH THE ENDOMETRIAL CHANGES**

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In a previous work we demonstrate that in a model in mouse BALB/C of bovine tritrichomonosis the embryonic death takes place during the second quarter of the gestation. In the present work the endometrial alterations are analyzed in mice with 5-10 days of pregnancy infected by *Tritrichomonas foetus*. The uterine horns were tried for its coloration with HE, Giemsa, Toluidina's Blue and for the histochemistry techniques of PAS and TUNNEL. Some animals already lost its pregnancy in the day 6. In most cases the animals of more than 8 days of pregnancy had lost the *conceptus*. Though in the endometrium some lesions were observed like exudate in the glandular light, expansion of this light and edema periglandular, the principal change is the great inflammatory infiltration that includes: neutrophils, eosinophils, lymphocytes, natural killer cells, plasmatic cells and macrophages. In the miometrium and perimetrium there were mast cells. These cells might be responsible for the embryonic death because alter the local immunological state necessary for the gestation. Also it was a notable find the great quantity of cells in apoptosis in the luminal and glandular epithelium and in the corion of the endometrium. The death of the luminal epithelium might alter the normal implantation and that of the glandular cells the production of histiotropho. Both processes also might participate in the embryonic early death.

290.**INVOLVEMENT OF p75 AND TrkB RECEPTORS IN NEURAL CREST CELL CHEMOTAXIS**

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Neural crest cells (NCC) migrate through defined pathways in the embryo and differentiate in multiple derivatives. Many factors are involved in cell migration, but the signals that guide the cells to their final destination remain unknown.

In our lab we study chemotaxis as a possible mechanism to colonize the target sites and we showed *in vitro* chemotactic response of mesencephalic NCC towards concentration gradients of *Neurotrophin 3* (NT-3).

In the present work, applying *in situ* hybridization, we have shown the expression of NT-3 mRNA in the optic vesicle and neural tube of chick embryo, that could be the source of a chemotactic gradient capable of induce NCC oriented migration through the optic vesicle, target region where NCC originate neurons and glia of the ciliar ganglion. Likewise, we have shown immunolabeling of p75 and TrkB receptors in NCC cultures and western blot analysis of NCC homogenate. On the other hand, we were able to block the chemotactic response of NCC towards NT-3 (40ng/ml) after treatment with 100 or 200 nM of Wortmanin, meaning that Protein 3 Kinase would participates in the signal pathway that modulate NCC oriented migration.

This data amplify previous results, and suggest that p75 and TrkB receptors (besides their canonical receptor TrkC) would participates in control of oriented response of NCC induced by NT-3.

291.**LECTINHISTOCHEMICAL STUDY OF RABBIT APPENDIX FOLLICLE ASSOCIATED EPITHELIUM DURING *Solanum glaucophyllum* INTOXICATION**

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The appendix is one of the rabbit immunocompetent organs. Luminal antigens pass over its follicle associated epithelium (FAE) to initiate local immune responses. *Solanum glaucophyllum* (Sg) is a calcinogenic plant that contains high levels of 1,25-dihydroxyvitamin D, a hormone that regulates cellular differentiation. We analyzed the effects of Sg on the lectin binding pattern of rabbit appendix's FAE. Three months-old New Zealand male rabbits received 125 mg/kg Sg leaves powder during 15 or 30 days. Samples were included in paraffin. Three μm of experimental and control sections were then incubated with the following biotinylated lectins: WGA, CON-A, DBA, SBA, PNA, RCA-1 y UEA-I. We only found differences in the binding pattern of DBA. It moderately bound to the FAE apical glycocalix and M cell intraepithelial pockets in the control group. However, there was a progressive reduction in the labelling of experimental groups' tissues with time. This might indicate a reduction in N-acetyl galactosamine expression, a glucidic residue implicated in microorganism adherence to FAE. Reduction on its expression could alter the adequate local immune response.

292.**GLYCOSYLATED RESIDUES OF PORCINE PLACENTA BY LECTINHISTOCHEMISTRY**

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The glycans are implied in cell-cell and cell-matrix adhesion and recognition. Its role in the maternal-fetal interphase of the pig placenta is of special interest. The objective of this work was to characterize the glycoconjugates present in the placenta. Histological cuts of ±4 μm from pig placentas of ± 30, 55, 70 and 114 days of gestation were used. The presence of the following glycosylated residues was determined: terminal and nonterminal α-D-manosil and α-D-glucosil, terminal and nonterminal N-acetyl galactosamine, L-fucosa and terminal β-D galactosil. Glycosylated residues were found to exist only in the uterine epithelium, trophoblast and glands of early gestational periods, such as α-D-manosil and α-D-glucosil. The residues terminal α-D-manosil and terminal α-D-glucosil, terminal and nonterminal N-acetyl galactosamine, terminal β-D-galactosil and L-fucosa are found in uterine epithelium and trophoblast in all the evaluated periods. In maternal connective tissues the residues α-D-manosil and α-D-glucosil were negative, being the rest of the evaluated residues positive in almost all the gestational periods. In fetal connective tissue a differential label of L-fucosa residues as observed between the chorion and villi. The glycoconjugated residues studied present labelling patterns in the tissues that constitute the pig placenta in the different selected gestational periods.

293.

HUMAN OVIDUCTAL SECRETION MODULATES THE SPERM CAPACITATION PROCESSZumoffen C¹, Caille A¹, Munuce M¹, Cabada M², Ghersevich S¹¹Área de Bioquímica Clínica. ²Área de Biología, IBR (CONICET), FCBYF, UNR, Rosario, Argentina.

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In contact with the female genital tract secretions, sperm (SP) undergo the capacitation process, acquiring the ability to fertilize the oocyte. The objective was to evaluate the effect of the conditioned medium (CM) obtained from human oviductal tissue culture (HOTC) on sperm capacitation. The CM obtained from HOTC (DMEM/Ham'F12 medium) of premenopausal patients, were collected, dialyzed, and their protein concentrations were estimated. The motile SP (swim up) from normozoospermic donors were incubated under capacitating conditions (6 and 22 h) in the presence or absence of CM ([proteins] 0.2, 0.8 and 1.6 mg/ml). After incubations the sperm viability (V) and the tyrosine phosphorylation pattern of sperm proteins (TPP) by Western blot were determined. The sperm ability to undergo the acrosome reaction (AR) induced by human follicular fluid was assessed by *Pisum sativum*-FITC, considering as inducible population (PI) = % induced AR - % spontaneous AR. The ANOVA and the test of Tukey-Kramer were used for the statistical analysis. The CM did not affect sperm V. A significant dose-dependent decrease was observed in the TPP (6 h, p <0.05) and in PI (6 h and 22 h, p <0.01) in the presence of increasing concentrations of MC proteins. These results would support a role of the human oviductal secretion as modulator of the reproductive process, contributing to limit the number of SP with ability to interact with and to fertilize the oocyte.

294.

ROLE OF THE VENTRAL HIPPOCAMPUS ON THE EXPLORATORY PREFERENTIAL DECISIONS IN RATSAbrego AV, Alvarez EO.

Área de Farmacología, Facultad de Ciencias Médicas, Universidad Nacional de Cuyo, Mendoza.

When an animal is exposed to a new environment, exploration can be present in two opposed ways. Exploration by one of these ways is called "preferential decision". Exploration of spatially neutral environments should be expected to be at random. However, previous work in rats in our laboratory has shown that, animals tend to explore by the left from left/right alternatives. Since this type of decision involves spatial preference, the objective of the present work was to evaluate if the ventral hippocampus participates showing laterality in these spontaneous behaviors. Groups of rats were bilaterally implanted with guide cannulae into the right ventral hippocampus (HPCr) and left ventral hippocampus (HPCl). 5 min before all animals were tested by 3 differential spatial models; different groups of rats were microinjected into the HPCr or HPCl with lidocaine (2 µg/µl). Saline microinjection was considered control. Preliminary results in the Lateral Double Holeboard Maze show that control animals (n=12) presented left rearing preference (32 ± 6.1 Counts/5min Vs 16 ± 4.8 Counts/5min, p<0.01). Lidocaine treatment in the HPCr (n=13, 33 ± 4.6 Counts/5min Vs 21 ± 2.7 Counts/5min, p<0.01) or in the HPCl (n=5, 47 ± 1 Counts/5min Vs 19.5 ± 7.5 Counts/5min, p<0.01) did not modify this preference. It is concluded that apparently the HPC is not involved in modulation of these preference choice behaviors.

295.

***Brucella abortus* RB51 BEHAVIOR IN THE ENDOCYTIC PATHWAY**

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Brucellosis is a zoonosis of high impact in our country. *Brucella* is a facultative intracellular parasite. In Argentina, the S19 strain of *B. abortus* is used for cattle immunization. However, this strain is not innocuous for human beings, since it is a smooth variant of attenuated virulence. In the rest of the world the rough strain RB51 is used for cattle immunization. The traffic of these two *B. abortus* strains across compartments of the J-774 macrophage endocytic pathway was tracked, labeling lysosomes with 60-nm gold particles (GP) through 3-h incubation at 37°C. Macrophages were then exposed to bacteria and incubated at 37°C for 2 to 18 h. Early endosomes were marked with 20-nm GP by a 15-min exposure before fixing the cells. The phagosomes containing lysosomal marker (60-nm GP), endosomal marker (20-nm GP) or no markers were counted with transmission electron microscopy. The rough strain always occupied large phagosomes and showed early signs of marked digestion, but the fusion between phagosomes and lysosomes is similar to that observed for the S19 strain. After long incubation periods, macrophages destroyed the rough strain more efficiently than the attenuated virulence strain. Considering the cases of brucellosis caused by the S19 strain, a comparison of its intracellular traffic with that of RB51 may help to take preventive actions against a highly prevalent disease in our country.

296.

PSYCHONEUROENDOCRIN BOARDING OF AGGRESSIVE BEHAVIOR IN A SEMIMILITARIZED INSTITUTION

Bianchi RA, Nanfaro F, Leytes E, Diaz M, Abud M, Ponce C, De

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In 1964 Sólon indicates the relation between emotions, immunity and neuroendocrin. The aim of the following work is related to: a) Study of the psychological diagnosis of aggressive and violent behavior (Test of Rorschach and Bender). b) Evaluation of the aggressive commitment through the neurological examination. c) Relation between aggressive conduct and hormonal rhythmic profile. d) Relation between aggressive conduct and immunological profile. There were studied 37 male agents of the Department of Justice and Security, aged between 22 to 43 years old and 30 controls with similar ages. In both groups there were realized the test of Rorschach and Bender and there quantified the levels of thyroid hormones, adrenocorticotropic hormone (ACTH), Prolactin (PRL), cortisol (CRT), dehydroepiandrosterone (DHEAs) by means of RIA and immunoglobulin IgG, IgM and IgA through radial diffusion. All subjects presented indicators of impulsiveness in the psychodramas: reflection response. 85% of the studied individuals showed low cortisol (14,60 /-2,56 experimental Vs 25,00 /-3,20 controls). The levels of IgA and IgM were observed diminished in 75% of the cases. (IgA mg/dl 120,00 /-3,00 experimental Vs 220,00 /-3,80 controls). In addition, Rhythmic differences were observed in the levels of ACTH, PRL and CRT. Relations are established between aggressive behavior; and psychological and neuroendocrin events.

301.**XANTHATIN INHIBITS CALCIUM IONOPHORE-INDUCED MAST CELL DEGRANULATION**

Vera M, Mariani ML, Favier S, Tonn C, Piezzi RS, Penissi AB.

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In previous work we have demonstrated that xanthatin (Xt), a novel xanthanolide sesquiterpene with antiulcer properties isolated from *Xanthium cavanillesii* Schouw, inhibits mast cell exocytosis induced by the G protein stimulant compound 48/80. The present work examines the effect of Xt on mast cell degranulation induced by the calcium ionophore A23187, to determine whether Xt acts upstream or downstream of cytosolic calcium increase. Rat peritoneal mast cells were purified in Percoll and incubated with: 1) Tyrode solution or 2) A23187 or 3) Xt+A23187. Serotonin release studies by high performance liquid chromatography (HPLC), evaluation of mast cell morphology by light microscopy, dose-response and time-response studies, cell viability evaluation by the tripan blue dye exclusion, comparative studies with ketotifen (Ket), and drug stability evaluation by thin layer chromatography (TLC) were carried out. Calcium ionophore increased serotonin release from mast cells and elicited evident morphological changes. These effects were inhibited by Xt in a dose- and time-dependent manner. The inhibitory effect exhibited by Xt was stronger than that of ketotifen, a classical mast cell stabilizer. In conclusion, the present study demonstrates that Xt inhibits A23187-induced mast cell activation, acting downstream of cytosolic calcium increase.

303.**GLUTAMIC ACID: LOCATION AND FUNCTIONAL STUDIES IN SPERM, SPERMATOGENESIS AND FALLOPIAN TUBE IN MOUSE**

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IHEM - Facultad de Ciencias Médicas - UNCuyo. Mendoza. The Glutamic Acid (Glu) is well studied at the central nervous system (CNS), although Glu receptors (GluRc) and Glu functions have not been fully described in sperms or fallopian tubes (FT). We reported the presence of GluRc in sperm cell and Glu as a triggering molecule of acrosomal reaction (AR). Now GluRc location was also extending to FT and mouse's testis. The GluRc was detected during the spermatogenesis in specific stadium related to acrosomal development. On the other hand, the same antibodies imuno stained the epithelium of FT according to sexual cycle. Again superfusion assays (SA) to establish the release-uptake of Glu was definitive demonstrated at the CNS, but not at the FT. In FT is important because is the physiological place for induction of AR. Recently, SA in the FT was positive. Moreover, these interchange had also a relationship with the stadium of the female sexual cycle. The uptake and release was different within the female cycle showing high uptake during diestrus and releasing at the estrus. Results indicate that sperm possesses GluRc –progressively expressed during spermatogenesis–, Glu promote AR and Glu is secreted and capture by FT –following the sexual cycle– to promote high level of Glu at the fertilization place and moment.

302.**OPPOSITE EFFECTS OF CHRONIC VERSUS ACUTE HYPOXIA ON CARDIOMYOCYTE SODIUM CURRENT**

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Chronic hypoxia prompts many physiological adaptations. The effect of chronic vs acute hypoxia on tetrodotoxin-sensitive sodium current (INaT) from control and chronically hypoxic rats. Cells were obtained using standard techniques and studied with whole-cell patch-clamp. INaT was measured as the difference in current magnitude before adding tetrodotoxin (6 μ M) and after it. Data were obtained with pclamp6 Clampex digitizer and analyzed with Clampfit (Axon Instruments). Results reported are mean \pm SEM. Seal resistance 2.60 \pm 0.20 G Ω ; resting membrane potential – 69.6 \pm 1.2 mV; cell surface area (by capacitive method) 9 000 \pm 80 μ m 2 . Acute hypoxia induced by switching the bubbling gas from O₂ to N₂ caused a dramatic fall of INaT measured after 5 min (n = 4; p < 0.0003). The effect of chronic hypoxia was assessed by comparison of INaT recorded in cardiac cells obtained from control rats (n = 6) and from rats submitted to a pressure of 0.5 atm for 8 h/day during 10 days. When studied at ambient pressure and clamping potentials from – 80 to + 60 mV, the INaT of cardiomyocytes from hypoxic rats was roughly twice that of cells from control rats (p < 0.001). The increase in INaT found in cardiac cells from chronically hypoxic rats may represent an adaptative response to counteract the acute depressing effect of hypoxia on cardiac sodium current. Our next step is to compare the response to acute hypoxia of cardiomyocytes obtained from control rats vs those obtained from chronically hypoxic rats.

304.**LICOPENE EFFECTS ON PROLIFERATION, MIGRATION AND ADHESION OF PROSTATE CANCER CELLS**

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Based on the evidence from epidemiologic, animal, and in vitro data and human clinical trials, it is evident that lycopene, a non-provitamin A carotenoid, is a promising agent for prostate cancer chemoprevention, because of the inverse association between diary ingestion of tomatoes and derived products and cancer. In order to study the effects of lycopene on proliferation, migration and adhesion, a human prostate cancer cell line androgen independent DU145 was used. Cells were grown in DMEM 10% fetal bovine serum, 37°C in 5% CO₂. Lycopene concentrations were: 0.3 μ m, 3 μ m and 6 μ m. Inhibitory concentration of lycopene on proliferation was 6 μ m. The possible molecules participating in migration (N-cadherine) and adhesion (E-cadherine), as well as the relation of E-cadherine/ β -catenine are evaluated.

The results suggest that lycopene inhibits the growing of prostate cancer androgen independent cells in a dose dependent way. People consuming high amounts of tomato based food may be protected against prostate cancer.

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