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Abstracts

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C1.**DECIPHERING THE ORIGIN, EVOLUTION AND FUNCTION OF EPITHALAMIC ASYMMETRIES***Concha ML**Laboratory of Experimental Ontogeny (LEO), ICBM, Faculty of Medicine, Universidad de Chile.*

Animals show behavioural, cognitive and neuroanatomical asymmetries but how brain lateralisation is established in the vertebrate brain is not clearly understood. One region of the brain showing widespread structural asymmetries is the epithalamus, which is formed by the habenular nuclei and pineal complex. Recent studies in the teleost zebrafish (*Danio rerio*) provide a conceptual framework on which to start building our understanding of the developmental control and function of lateralised circuits in the vertebrate brain. *In vivo* analysis of transgenic zebrafish has revealed that epithalamic asymmetry is initiated by positioning of the parapineal nucleus on the left side, an event that shows a rigorous genetic and morphogenetic control, and serves to amplify existing habenular asymmetries. Habenular asymmetries are characterised by a differential development of distinct types of projection neurons in the left and right habenula, and the spatial segregation of their connectivity within the main midline midbrain targets: the interpeduncular nucleus and raphe. The physiology of the habenula is still poorly understood although recent studies indicate a sensorial modulation by light. Comparative approaches also indicate that structural asymmetries of the habenula are conserved among teleosts, and recent findings also extend these observations to humans.

C2.**BEHAVIOURAL ADAPTATIONS IN AVIAN BROOD PARASITES***Reboreda JC**Departamento de Ecología, Genética y Evolución, Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires.*

Interspecific avian brood parasites lay eggs in nests of other species, the hosts, which provide parental care for their progeny. Unlike other birds, female brood parasites must locate and recognize host nests and decide whether to parasitize them or not. In addition, they may enhance the hatchability of their eggs by synchronizing parasitism with host laying and the survival of their chicks by reducing the competition for food their young face in the brood. Within the parasite cowbirds (family Icteridae) the shiny cowbird (*Molothrus bonariensis*) is a wide generalist that parasitizes more than 250 hosts. In this talk I will describe behavioural adaptations to brood parasitism in shiny cowbirds. Although this species uses many hosts at population level, individual females do not lay their eggs randomly, but preferentially parasitize particular hosts, leading to host-specific lineages within a parasite's population. Female shiny cowbirds find host nests individually, probably because of higher spatial memory capabilities, or by following other females (social learning). They parasitize host nests at dawn and before laying they peck and puncture host eggs. Females that parasitize large hosts lay larger eggs and peck and puncture more host eggs than females that parasitize small hosts. Our results indicate that cowbird females using different hosts may have had divergent evolutionary histories that resulted in the adjustment of their parasitic behaviour to the characteristics of hosts.

C3.**AMPHIBIAN POPULATION DECLINES AND EXTINCTIONS IN THE NEOTROPICS***Lavilla EO**Fundación Miguel Lillo – CONICET. Miguel Lillo 251 (4000) San Miguel de Tucumán. E-mail: eolavilla@gmail.com*

In the late 1980s, herpetologists perceive that many amphibian populations showed clear signs of decline, while others were extinct. This situation, initially seen as local phenomena, proved to have global scale and, to complicate matters further, in some cases we could not find a clear explanation. The traditional causes, such as the irreversible transformation of habitats by population growth, the expansion of agricultural frontier, mining and hydrocarbons exploitation, the introduction or translocation of species and climate change alone were insufficient to understand some extinctions. A decade later we were able to identify some emerging diseases caused by viruses or fungi associated with populations at risk, but each answer generated new and more complex questions. Confronted with this reality conservationists around the world began a race against time trying to reverse, or at least to stop, these processes that are leading our society towards a world without frogs. A quarter century later, and after hundreds of meetings, reams of documents, thousands of hours in the field and millions spent, we must realize that the problem exceeds our professional field. Biologists are essential but not sufficient, since the detection of the problem and the proposed actions will become useless in the absence of interest and support of the society as a whole, especially of those responsible for establishing conservation policies, enact the laws that support them and ensure its compliance. This presentation focuses on the status of amphibians in South America, the region of greatest species richness, and aims to draw attention on the problem and is an invitation to reflect on an issue that concerns us all.

C4.
PANCREATIC BETA CELLS: CHANGES IN TYPE 2 DIABETES

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Diabetes, especially type 2 diabetes (T2DM), is a disease whose prevalence increases constantly in the whole world. In Argentina it has passed from an 8.4% in 2005 to an 9.6% in 2010; when it is not correctly treated it generates chronic complications that augment the attention costs and decrease life quality of patients. A decreased response of tissues to insulin and a progressive decrease of islet beta cell mass and function are associated to its pathogeny. A complementary disfunction of alpha cells is observed, which secrete inadequate glucagon levels. This disfunction plays an important role in the development of the metabolic alterations characteristic of diabetes. Two mechanisms become associated to produce the mentioned alterations: glycol-oxidative stress and endoplasmic reticulum stress. The former obeys to an increase in the production of free radicals in the mitochondria and to the action of NADPH oxidase. The latter is a consequence of the increase of the secretory activity of beta cells that try to compensate insulinresistance. The concentration of antioxidant enzymes in the islet turns it very sensitive to oxidative stress. The decrease in beta cell mass and function are evident in previous phases to overt diabetes, i.e. in the altered glucose tolerance phase. Furthermore, in T2DM, the only identified cause capable of decreasing beta cell mass is the increase in the apoptosis rate. Although to date there are no drugs available capable of regenerating the lost beta cells, some drugs have shown some efficacy in diminishing apoptosis, which permits to hope that applied early-on they may be able to prolong beta cell life. This implies that to date prevention is the best therapeutic strategy to maintain the effective beta cell mass in order to obtain an adequate diabetes control.

C5.
GENETICS AND EPIGENETICS OF THE METABOLIC SYNDROME

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Although the concept of metabolic syndrome (MS) is under debate, five screening variables used to identify individuals with MS are waist circumference, circulating levels of triacylglycerols and high density lipoprotein cholesterol (HDL), fasting glucose and blood pressure. MS is recognized as a major prevalent cardiovascular and type 2 diabetes risk factor by WHO, NCEP-ATPIII and IDF, including a constellation of complex diseases such as type 2 diabetes (T2D), dislipidemias, central obesity, hypertension, a proinflammatory and prothrombotic state, ovarian polycystosis and hepatic steatosis. The genetics of these diseases is complex and vary in a spectrum from monogenic and syndromic forms, usually rare, to the most common polygenic and multifactorial form. Indeed few patients with certain rare single gene disorders, such as mutation of *MC4R* and *PPARG*, express the clusters of abnormalities seen in MS. Using the candidate gene or GWA approaches, studies indicate that common genetic variants of genes such as *TNF α* , *ADRB3*, *SLC6A4*, *INSIG2*, *GAD2*, *CLOCK*, are associated with the development of MS. In addition, animal models through comparative genomics can give a map for candidate loci in human. Taken together several thousands of putative genes emerged from these studies. One promising approach is to narrow the list by using bioinformatic tools for prioritization. Epigenetic factors such as DNA methylation, and mitochondrial DNA copy number may also play a role. As with most complex diseases, it is premature to propose molecular genetic testing for diagnosis (at least for the polygenic and multifactorial forms) and treatment. However, the association of MS with genes such as *SLC6A4*, *PPAR α* and *PPAR γ* , already targets for approved drugs may suggest new avenues for MS pharmacological treatment.

ANIMAL REPRODUCTION, CLONING AND TRANSGENESIS**S1-1.****EMBRYO QUALITY IN EMBRYOS GENERATED BY FIV CLONING AND *IN VIVO****Mutto A**Grupo de Biotecnología de la Reproducción, Instituto de Investigaciones Biotecnológicas Instituto Tecnológico de Chascomús (IIB-INTECH, UNSAM-CONICET, Buenos Aires.*

Reproductive biotechnology includes techniques that increase the reproductive efficiency of animals for meat, milk, wool and hair production, etc. However, with the advent of *in vitro* fertilization, somatic cell nuclear transfer (cloning) and transgenesis, has shown that embryo competence may be severely compromised, without necessarily a correlation with obvious changes in morphology. These changes in embryo quality are commonly manifested in the ability to establish pregnancies and that these, come to the end of gestation resulting in a healthy offspring. The evaluation and prediction of the quality of embryos produced *in vitro* has acquired a key role for both the commercial sector and for scientific research. It is known that *in vitro* production techniques printed alterations in gene expression in embryos and oocytes, compromising their quality. These changes may affect different physiological functions (metabolic, respiratory, immune, etc.) and are considered a type of stress response caused by embryo culture conditions and protocols used in assisted reproduction techniques.

S1-2.**BIOTECHNOLOGY TECHNIQUES APPLIED IN SOUTH AMERICAN CAMELIDS***Miragaya M**Cátedra de Teriogenología INITRA, Facultad de Ciencias Veterinarias, UBA, CABA.*

Reproductive biotechnology techniques, which include artificial insemination with cooled or frozen-thawed semen, embryo transfer (ET), *in vitro* embryo production (*in vitro* fertilization (IVF), intracytoplasmic sperm injection (ICSI) and cloning) are widely used in domestic species such as bovines and equines. The industry that surrounds these species promotes the research and development of these biotechnologies, but in South American Camelids (SAC) research in this area has been minimal. Complex reproductive characteristics of these species contribute to this lack of research. The induced nature of ovulation, the short life-span of the corpus luteum, the difference in luteolytic activity between the two uterine horns, the short period for maternal recognition of pregnancy (MRP), the highly viscous semen with low sperm concentration, constitute serious challenges for scientists. Nevertheless, some assisted reproductive techniques (such as synchronization of ovarian follicular development, ovarian superstimulation and ET) demonstrate a greater increase in knowledge while others show less advances (artificial insemination, IVF and ICSI) and certain basic reproductive physiology remains unclear, such as MRP signaling. Ovarian superstimulation (with FSH or eCG) to induce follicle growth for *in vivo* embryo production or *in vivo* follicle aspiration entails prior administration of hormones that inhibit follicular growth (progesterone, progestagens and estrogens). The cumulus-oocyte complexes that are obtained must be matured *in vivo* (buserelin administration) or *in vitro* before being subjected to techniques such as IVF or ICSI. All these techniques also require morphologically normal, motile spermatozoa to be used to achieve fertilization. The methods used to decrease semen viscosity (incubation with 0.1% collagenase) and to select the best spermatozoa (Percoll®; Androcoll-E™) will be described. Embryos that are obtained by any of the three techniques: *in vivo*, IVF or ICSI, need to be transferred to synchronized recipient females. The best results obtained are after transfer to the left uterine horn with an ipsilateral ovulation. Applying reproductive biotechnology techniques such as those described, will permit an increase in the number of offspring from genetically selected animals and from the wild camelid species, vicunas and guanacos, whose embryos could then be transferred to the uterus of a domestic species.

S1-3.**AGEING AND REPRODUCTION IN DOMESTIC ANIMALS***Losinno L.**Laboratory of Equine Production, Faculty of Agronomy and Veterinary Medicine, Universidad Nacional de Río Cuarto.*

The aging of an organism results from a series or sequence of processes that produce morphological and physiological changes consistent with the gradual loss of homeostasis and increased susceptibility to certain specific diseases. The effects of aging could be a combination of programmed processes and genetic alterations induced by exogenous and endogenous factors. Ageing is therefore an ongoing process, widespread and irreversible that affects all systems in varying degrees, which produces structural and / or functional DNA alterations via oxidative processes (free radical) and / or environmental contaminants. One of the most important effects is the changes in the neuroendocrine and metabolic regulation (thermoregulation, biological rhythms, hormonal feed-backs), with the consequent decrease in homeostatic capacity and its profound effects on the hypothalamic-pituitary-gonadal axis. Reproductive senescence, is a general phenomenon related to aging of the organism and is somewhat related to this chronology, ie the gradual deterioration of cellular processes extends equally to different organs and systems. Although in general the concepts of chronological age and biological age are managed (and often manifest themselves) independently. In females, the decline in age-related fertility has at least two components measured: 1) the decrease in rates of conception / pregnancy and 2) the increase in the rate of early losses and / or late pregnancy. In fact, one of the first signs of reproductive senescence is a decrease in fertility rates in each individual predictor. Currently, there is a large consensus that reproductive aging is associated with age in female mammals is also related, among many other factors to a loss of viability of oocytes and embryos and possible alterations in gene expression. Among the most obvious signs of this loss of reproductive potential are increasing the number of oocytes / embryos for numerical and structural chromosomal abnormalities, cytoplasmic fragmentation associated with apoptosis, polyspermy, abnormal development of embryos and consequent increase in embryonic or fetal mortality.

NEUROSCIENCE

S2-1.

HIGH NOON: MOVING THE HANDS OF THE CLOCK FROM WORMS TO MICE

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C. elegans is a nematode widely used as a model system in genetics and developmental biology, although it has not been completely characterized in terms of its chronobiology. In our lab we have described the locomotor activity rhythm of this model, which includes all of the features of bona fide circadian rhythms: 1) they have a period close to 24 h, 2) they can be entrained by environmental stimuli (such as light or temperature) and 3) they have temperature-compensation mechanisms (with a Q10 close to 1). Although the genes responsible for such rhythmicity are not known, we have identified a few candidates participating in the origin or entrainment of circadian cycles. In addition, we have characterized other circadian variables in this model, including metabolic phenomena (feeding, defecation, pharyngeal pumping, oxygen consumption) and susceptibility to biotic and abiotic stressors. The photosensitive mechanisms of *C. elegans* are not completely understood. According to our data, there is a positive phototactic index towards green light, and there have been reports of negative phototaxis towards blue/ultraviolet light. The *lite-1* pathway (with members of a novel family of photoreceptors sensitive to blue/UV light in this species) does appear to be involved in circadian entrainment. However, mutants of the *tax-2* pathway (another member of the family) show alterations in synchronization. Finally, we have found diurnal/circadian rhythms in the levels of melatonin and the activity of its synthesizing enzyme, *aaNAT*. The nocturnal peak of *aaNAT* was inhibited by white or blue light, indicating that this variable constitutes an additional photosensitivity assay for *C. elegans*. It is interesting to consider the adaptive value of circadian rhythms and entrainment in this subterranean species. A cyclic environment (in terms of potentially toxic stressful challenges) might lead to the selection of circadian features, which obviously require to be entrained even by the subtle environmental cues of this nematodes, which include light exposure, thermal and chemical changes.

S2-2.

ADULT-SPECIFIC ELECTRICAL SILENCING OF PACEMAKER NEURONS DOES NOT ALTER THE PACE OF THE MOLECULAR CLOCK IN *DROSOPHILA*

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Circadian rhythms regulate physiology and behavior through the action of self-sustained transcriptional feedback loops of clock genes. In *Drosophila*, over 150 neurons in the fly brain are implicated in the circadian regulation of rest-activity cycles, but the small ventral lateral neurons (sLN_vs) are clearly crucial. The preservation of molecular oscillations within the sLN_vs is key to command rhythmic behavior under free running conditions. The sLN_vs transmit this time information releasing a neuropeptide known as pigment dispersing factor (PDF), and likely changing synaptic partners by remodeling their axonal terminals in a circadian fashion. Electrical activity of PDF neurons is also required for rhythmicity. Silencing PDF neurons by expressing a K⁺ channel (KIR) throughout lifetime leads to behavioral arrhythmicity and blocks molecular oscillations in the sLN_vs (Nitabach *et al.* Cell 2002). To gain insight into the relationship between ion conductances through the cellular membrane and molecular oscillations taking place within the nucleus/cytoplasm we developed a new tool for temporal control of gene expression in PDF neurons. Silencing the PDF circuit only during the adult stage led to behavioral arrhythmicity as previously described. Surprisingly, once *kir* expression was shut down, flies recovered rhythmicity in a phase reminiscent to that of the initial training. PERIOD oscillations in the sLN_vs showed that the molecular clock remained intact through the silenced phase, supporting that arrhythmicity is a consequence of the inability of these neurons to transmit information rather than an effect on the clock, as previously proposed. Accordingly, both the complexity of the axonal terminals as well as PDF accumulation were severely affected during the silenced phase. Interestingly, long-term silencing of PDF neurons (i.e. throughout developmental and in the adult) indeed altered molecular oscillations suggesting that long-term effects on membrane potential might trigger undesired effects on cell physiology or viability, instead of specifically drive molecular oscillations.

S2-3.

TRANSCRIPTIONAL REGULATORY MECHANISMS INVOLVED IN THE ESTABLISHMENT AND MAINTENANCE OF CIRCADIAN RHYTHMS

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The self-sustained circadian clocks, including those located in the mammalian hypothalamic suprachiasmatic nuclei, run based on interlocked transcriptional and post-transcriptional feedback loops. This organization is highly conserved among species. Twenty-four-hour rhythms in physiology and behavior result from these widespread oscillators. Though our understanding of the molecular basis of circadian rhythmicity has advanced considerably, unresolved questions remain. For example, when and how are specific circadian rhythms first established? How are they ontogenetically regulated? What mechanisms determine tissue-specific circadian responses? Since transcriptional events have been thought to be crucial in determining circadian rhythmicity, several cis- and trans-acting elements have emerged as components of the circadian clock core. In mammalian promoters, for example, it is known that perfect E-boxes (CACGTG) are responsible for recruiting the complex BMAL/CLOCK to regulate expression of clock and clock-controlled genes. However, neither this 6-bp DNA element nor these two members of the bHLH transcriptional factor family are sufficient to drive specific circadian rhythmic patterns exhibited by a continuously growing number of genes. E-box-like sequences and bHLH factors are considered promiscuous; they have been involved in controlling diverse mechanisms beyond circadian transcription, including proliferation, differentiation, tissue-specific responses and cell death. Recent advances have uncovered molecular connectors between the circadian clock and the cell cycle clock. These results should encourage us to continue to search for other potential molecular bridges among the E-box/bHLH-dependent mechanisms. It is expected that the study of transcriptional factors that are known to play a tissue-specific differentiation role during ontogeny will bring new insights into the establishment and maintenance of a defined circadian rhythm.

EFFECT OF NATURAL PRODUCTS ON BIOLOGICAL SYSTEMS**S3-1.****BIOLOGICAL PROPERTIES OF MEDICINAL PLANTS FROM HIGH ALTITUDE ECOSYSTEM AND POTENTIALS USES IN THE PHARMACEUTICAL INDUSTRY**

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Plants from arid and semiarid regions are subjected to intense environmental stress and to adapt to these conditions of life, they have developed valuable defense strategies such as the chemical compound production. The finding of new molecules with potential uses in the pharmaceutical industry in the treatment and prevention of some pathologies is very interesting. The aim of this study was to evaluate the biological properties (antioxidant, antimicrobial, anti-inflammatory and genotoxicity activity) of medicinal plants from high altitude ecosystem and the isolation of the bioactive compounds. In this work were selected plant species used traditionally as medicinal in the Argentine Puna, including: *Baccharis incarum*, *Baccharis boliviensis*, *Chuquiraga atacamensis*, *Parastrephia lucida* (Asteraceae family) and *Fabiana punensis*, *F. bryoides*, *F. densa* and *F. patagonica* (Solanaceae family). The plants extracts showed antibacterial activity against antibiotic resistant bacteria, also showed antioxidant activity, by scavenging activity of free radical and reactive oxygen species, and showed anti-inflammatory activity by the inhibition of enzymes involved in the inflammatory process. None of the plant extracts were genotoxic. From the most promissory plants extracts, were obtained some bioactive compounds through assay-guided isolation and were formulate phytotherapeutic products with antibiotic and antioxidant properties. The results of this study justify the traditional medicinal uses of plants from high altitude ecosystem.

S3-2.**EFFECT OF A SECONDARY METABOLITE OF ARTEMISIA DOUGLASIANA BESSER, THE DEHIDROLEUCODINA, THE MEIOTIC CELL CYCLE**

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Sesquiterpenic lactones (STL) are a group of secondary metabolites. The natural diversity of this group of natural compounds is related to a very wide range of biological activities, which have an important defensive role in the plants that synthesize them. The majority of STL have been isolated from the Asteraceae family, which belong to one of the biggest and most diverse group of plants in our planet. Dehydroleucodine (DhL) is a sesquiterpenic lactone, isolated from the leaves of *Artemisia douglasiana* B, popularly known as "matico". Nowadays, there is a lot of interest in finding vegetal compounds that act on the cellular cycle. In this regard, active vegetal compounds have demonstrated potent control of cell proliferation. The utilization of ovocyte meiosis as a study model of the effect of lactones on cellular cycle has many advantages. In amphibians, meiosis stops at two points: Prophase I and metaphase II. Its progression is controlled mainly by the activity of a cytoplasmic compound, maturation promoter factor (MPF). Our group has demonstrated that DhL and its hydrogenated derivative, in which the alpha-methylenelactone (2H-DhL) function has been inactivated, inhibit the reinitiation of meiosis in amphibian ovocytes, keeping cells in prophase I. The analyses of the effect of DhL on transduction signals, which lead to the activation of MPF, suggest that the Myt1 kinase is the target of the lactones, either directly or through the inhibition of phosphatase PP2A. During fecundation, the sperm induces ovocyte activation by the inactivation of MPF, that permits meiosis end and the formation of pronuclei. Our laboratory has demonstrated that DhL induces the activation of mature ovocytes of *Rhinella arenarum* by interfering with the activity of MPF. DhL could be useful in techniques of reproductive biotechnology, where the low rate of viable offsprings is attributed to failures in the process of ovocyte activation.

S3-3.**CHEMICALS PRODUCTS FROM VEGETABLE ORIGIN USED ON WEEDS CONTROL**

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Since the birth of agriculture, one of the major concerns was the weeds control. On the other hand, species of agricultural interest tend to lose their defenses during the domestication process, making them more susceptible to pests and diseases than wild species. The plants synthesize and accumulate in their bodies a variety of secondary metabolites. One of the theories proposes that plants use these compounds for their defense against weeds, pathogens and superior opponents. Substances with various chemical structures and functions, such as organic acids, naphthoquinones, coumarins, flavonoids, tannins, sesquiterpene lactones, alkaloids and others, were found having biological activities. Numerous studies indicate that they may inhibit seed germination, seedling development and persuade natural detractors, fungi or bacteria. Our team has focused its research on some highly invasive plant species as *Centaurea diffusa* Lam. (native from Minor Asia) and the Argentine species *C. tweediei* Hook & Arn, both from the Asteraceae family, looking for the metabolites they produce. The purpose of isolating them is to study its effect on other weeds and pathogens, so as to serve as tools for integrated pest management. We evaluated the extracts, subextracts and pure compounds isolated of those species, on *Triticum aestivum* (wheat), *Lycopersicon esculentum*, var. San Pedro (tomato), *Lactuca sativa* (lettuce) and the weed *Leonurus sibiricus* (black weed). The tests were performed *in vivo* in organic substrate, and *in vitro* in Petri dishes containing soft agar as a support base to contain the seeds. We evaluated the percentage of germination, seedling development, weight and activity of dehydrogenases. The results obtained were varied, allowing evidence of different effects as inhibitors of germination, seedling growth retardant and antibacterial action.

PRESERVATION AND CONSERVATION OF SPECIES DIVERSITY

S4-1.

ASSISTED REPRODUCTIVE TECHNOLOGIES IN SHEEP AND GOATS

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The possibility of preserving sheep and goat semen has been a priority topic of research, since artificial insemination (AI) was considered a rapid and appropriate reproductive biotechnology for disseminating characteristics of high genetic value rams and bucks. The AI has facilitated the diffusion of genes, avoiding transport, stress and sanitary risk of males. The high demand from Argentine breeders to improve their production has determined the need to implement genetic breeding and evaluation programs based on the use of AI. In this context, we have accomplished, with special consideration to our production system, a series of applied research on assisted reproductive biotechnologies that allow the dissemination and preservation of genetic material from sheep and goats. In order to have biotechnology tools to improve animal production and to preserve genetic material by cryogenic conservation, we have developed and evaluated several methods of estrus synchronization, AI techniques and seminal or embryo conservation methods. Certainly, the natural reproductive potential is a limiting factor for genetic progress. Indeed, another assisted reproductive technology, in which we are carrying out research, is multiple ovulation and embryo transfer (MOET). This is a tool for rapid improvement in the genetic level of different breeds or cross-breeds. The AI and MOET, together, constitute excellent biotechnologies for genetic improvement and species preservation. In turn, the genetic material could be used to incorporate genes of interest in a breed, to develop a new genetic group, support *in vivo* conservation programs and provide material for molecular studies aiming to identify genes of economic or biological interest. The sheep and goat production system will have its future in the continuous challenge of increasing investment in genetics, with special attention to quantitative and qualitative features that can add value to its production and taking in consideration the biodiversity and sustainable development of their production system. More information: <http://www.inta.gov.ar/bariloche/info/indices/animal/reproduc.htm>

S4-2.

EFFECTS OF INDIRECT IMPACTS (TOURISM AND ROADS) ON BIODIVERSITY IN ISCHIGUALASTO PROVINCIAL PARK: A CASE STUDY

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Tourist activities and associated infrastructure (e.g., roads, trails) are anthropogenic activities causing disturbances in ecosystems; however, at the same time they promote social and economic development. Ischigualasto provincial park (San Juan, Argentina) is a protected area that aims at protecting an arid ecosystem with important paleontological remains and an interesting extant fauna, since it harbors endemic species and species of conservation concern. Tourist activity is very important and gradually increasing. The Central Bioceanic Corridor is under construction; this road will connect ports in the Atlantic and the Pacific Oceans, and will traverse the park in San Juan. The tourist activities and the corridor will produce direct and indirect effects on wildlife: modifications in time allocated to behaviors related to biological fitness (e.g., foraging), modifications in the use of space, abandonment of high-quality habitats, increased vehicular traffic, increased probability of collisions, among others. We studied the effect of tourist activity in the park and areas near the road and trail (circuit) within the park on fauna species of high biological interest (*Lama guanicoe*, *Dolichotis patagonum*) relative to abiotic variables (e.g., bare soil, rocks) and biotic variables (e.g., cover of different vegetation strata). The results indicate that the presence of tourists and short distance to the road and trails negatively affect the use of space by *D. patagonum* and time allocation in *L. guanicoe*. Sites of high probability of road cross were detected and the construction of fauna crossings as a mitigation measure is proposed. Studies of the disturbances produced by ecotourism and associated infrastructure on individuals, populations and ecosystem processes are still scarce worldwide. However, the scientific community recognizes the need to study the impact of ecotourism on biological diversity, considering a balance between human enjoyment and use of nature, to contribute to appraisal of biological diversity and therefore to its conservation.

S4-3.

ANDEAN CONDOR CONSERVATION PROGRAM: JOINING SCIENCE WITH ANCESTRAL TRADITIONS

Jácome NL.

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During thousands of years, the Andean Condor (*Vultur gryphus*), the biggest flying bird of the world, has been honored by the South American origin communities, considering it as a sacred link between people and God. Abundant, in other times, this emblematic bird today is a conservation challenge. For this reason, in August 1991, The Andean Condor Conservation Project (ACCP) began within Chilean and Argentine Program. The main goal is the conservation of these legendary birds and their majestic environment, all around the South American mountain ridge, in order to ensure the survival of which is considered The *Living Spirit of the Andes*. Through an artificial hatching and isolated breeding program, as well as the creation of a rescue and rehabilitation center, the project could release 101 specimens in entire South America. The ACCP, in 1997, was pioneer in the use of satellite technology to study the flight of the condor, in their diary movements, in a place as difficult as the Andes Cordillera. Over 42 transmitters have been used to study their movements in Venezuela, Bolivia, Chile and Argentina. The use of radio telemetry and satellite transmitting, in association with hard in situ work, has permitted the creation of GIS (geographical information system). Besides it was created a special software, DECOSAT, as a flight simulator program as a helpful way to understand their movements. Thanks this modern technology it is possible discover the environment use, sleep sites, flight capacity, preferred habitats and other biological characteristics. The origin communities, who knew how to live together and respect this specie, before each release, do ancestral ceremonies. They are the ones in charge of sending their prayers as a message of hope, following the millenary traditions of their cultures.

ECOLOGY AND ENVIRONMENT

S5-1.

STRATEGIES OF *P. PUTIDA* TO COUNTERACT THE EFFECT OF TOXIC IONS AND UNDESIRABLE COMPOUNDS FROM CATIONIC DETERGENTS DEGRADATION*Lucchesi GI.**Dpto. Biología Molecular. FCEFQyN. UNRC. (5800) Río Cuarto. Córdoba. Argentina.*

The Br-tetradecyltrimethylammonium (TTAB) is a cationic surfactant widely included in antiseptic solutions and fabric softeners. Released into the environment, its biodegradability may be limited by their antimicrobial activity. The TTAB degradation by *P. putida* ATCC 12633 is initiated by N-dealkylation catalyzed by a TTAB-monooxygenase activity resulting in the formation of tetradecylalkanol and trimethylamine (TMA). The TMA is metabolized to NH_3 through oxidation and demethylation, and also accumulated inside the cell, affecting the bacterial growth, an effect counteracted by the addition of AlCl_3 . Al^{3+} acted as a Lewis' acid, playing a role in the control of TMA intracellular levels by the formation of Al^{3+} :TMA complex, and the TTAB is fully consumed without accumulation of undesirable compounds. Thus, the use of Lewis' acids to sequester intracellular amines offers an alternative to achieve an efficient utilization of TTAB by *P. putida*. The major bacterial response in TTAB-containing media is to produce a highly negatively charged membrane by an increase of phosphatidic acid and phosphatidylglycerol to neutralize the positive charge of TTAB. In the presence of Al_3Cl , an increase in phosphatidylcholine (PC) was detected, which led us to propose a physiological role for PC as a temporary reservoir for Al^{3+} through the formation of Al^{3+} :PC complexes. We have cloned and sequenced a gene in this strain that encodes a phosphatidylcholine synthase (PCS) and characterized a *pcs*-deficient mutant. In the mutant, detectable levels of PC were not found, and the mutant was much more sensitive than the wild-type strain when challenged with Al^{3+} supporting that the PC acts as a temporary reservoir for available Al^{3+} through the formation of Al^{3+} :PC complexes. These complexes are utilized as an Al^{3+} reservoir in the membrane and the bacteria can sequester the ion to reduce the TMA accumulated inside the cell, permitting the total oxidation of TTAB.

S5-2.

ECOLOGICAL RISK OF AMPHIBIANS IN AGROECOSYSTEMS OF CENTRAL-EASTERN ARGENTINA*Lajmanovich R.**Facultad de Bioquímica y Ciencias Biológicas, Universidad Nacional del Litoral, Argentina.*

In the past 20 years oilseed production in Argentina increased by around 67% and the area planted increased by about 45%. The relative shares of different types of plantings were abruptly changed to focus on one type of crop, the transgenic soybean tolerant to glyphosate. This increase in planted area, is directly related to the increased use of pesticides (herbicides and insecticides), which at nationally level are mainly glyphosate, 2,4 D, atrazine, endosulfan, cypermethrin and chlorpyrifos, among others, as well, also increased use of nitrogen fertilizers. In this context, the destruction of habitats, contamination of water by the rain washed of cultivated fields and eutrophication, are among the most serious problems facing amphibian populations worldwide. Therefore, it is important to develop techniques of biomonitoring that allow the interpretation of trends of affected local populations and in turn, lay out management strategies that aim at conservation of these vertebrates. This compendium will be presented results, experimental and field, of 10 years of research in Santa Fe and Entre Ríos provinces. Among the methodologies used include the use of nondestructive biomarkers, in situ microcosms and detection of pesticide residues. Also I will discuss the importance of amphibians in agricultural systems and their potential role as biological control of pests. The results, so far, allowed to determine the replacement of assemblages in the abundance of species (greater presence of terrestrial species but decrease of aquatic ones) attributable to changes in the landscape, were quantified morphological changes of blood cells, teratology and malformations in individuals from areas of intense agricultural use. Finally, by way of integration, I present a characterization of the ecological risk of 11 common amphibians species based on interspecific variation in basal enzyme activities, in the geographic distribution of each anuran, reproductive cycles and phenology overlap with soybean crop.

S5-3.

MINERAL BIOLEACHING AND HEAVY METALS BIOREMEDIATION*Donati E.**CINDEFI, Facultad de Ciencias Exactas, UNLP, Calle 47 y 115 (1900) La Plata, Argentina.*

Metals can be extracted from ores through the microbial action on the sulfides oxidizing them to sulfate and releasing the metals. This process called bioleaching involved different microorganisms mainly bacteria and archaea capable of oxidizing iron(II) and/or elemental sulfur. Iron(III) and sulfuric acid are produced during those oxidations allowing the attack and dissolution of the sulfides. Mineral bioleaching is industrially applied for recovering copper and other metals like cobalt and nickel. A similar process called biooxidation is used in the commercial pre-treatment of refractory ores; in this way the recovery of gold can be strongly enhanced. The mechanisms used by microorganisms to attack and dissolve metal sulfides are the same which can take place in coal or metal mines closed in a wrong way. Under such conditions, being exposed to water and oxygen, the oxidation of sulfides produces drainages with very low pH values and containing very high concentrations of heavy metals. These acid mine drainages (AMD) show a dark red color due to the high content of iron(III) and they are responsible for serious environmental impact. Most microorganisms living in such conditions have developed tolerance mechanisms in order to survive. Processes to treat metal-contaminated effluents or sediments in order to mitigate their effects on the environment, are based on the mechanisms of tolerance some microorganisms have developed after being exposed to high concentration of metals. Most of those processes imply the mobilization or the immobilization of metals; in some cases, metals can be transformed into less toxic species. Bioprecipitation and biosorption have achieved commercial applications. In the first case, sulfate-reducing microorganisms are capable of generating hydrogen sulfide under anaerobic conditions to precipitate metals as sulfides. In the second one, metals are captured by the microbial surface and even into the cells producing their immobilization.

DESERT: "ALTERNATIVES ADAPTED TO LIVE IN HIM AND THREATS TO THEIR CONSERVATION"

S6-1.

SELECTION PRESSURES, CONVERGENCE AND COEXISTENCE OF DESERT SMALL MAMMALS

Ojeda RA.

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The Desert ecosystem is an ideal scenario in which to test some of the relevant aspects of the theory evolution and ecology. Among these, the homoplasy by convergent evolution is one of the attributes shown by organisms from different lineages in distinct places, and exposed to similar selective pressures. To understand the factors that operate as selection forces on the Monte Desert organisms (e.g. low productivity, low rainfall, etc.), their adaptations, and the diversity and coexistence mechanisms in a community of small mammals from different lineages, is part of the research Program at the GiB. The integrated approach from different disciplines (biogeography, ecophysiology, ecology, genetics, behavior) allows a better understanding of the composition, organization, functioning and dynamics of biological diversity in a desert ecosystem, their similarities and differences with other deserts in the world, and on the other hand, greater robustness in the basis for conservation planning in face of the many and increasing threats to biodiversity.

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S6-2.

LIVING WITH SCARCE AND LOW QUALITY RESOURCES: PHYSIOLOGICAL AND BEHAVIORAL STRATEGIES OF A DESERT DWELLING RODENT

Sassi PL.

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The energy balance depends on the efficiency of organisms to use their trophic resources, and has direct impact on their fitness. It is a parameter conditioned by environmental variations that affect availability and quality of such resources, as well as depends on the digestive tract design. In this sense, it is expected that features associated with food utilization are subjected to strong selection pressures and show some adjustment to the variability of the environment. Thus, in this study I address the following questions: What are the morphological, physiological and behavioral attributes like in a species that faces nutritional trade-offs? What is their capability to settle in to spatial and temporal heterogeneity? To answer them I used as a model a small herbivorous rodent widely distributed in arid zones of South America: the small cavy, *Microcavia australis*. Considering both spatial and temporal scales, four populations occurring in the Monte Desert were selected for developing studies during wet and dry seasons. Using diverse methods I measured traits related to the species nutritional ecology such as: plant cover, plant diversity, trophic niche breadth, and diet quality. Furthermore, behavioral, physiological and morphological responses were assessed, like coprophagy, energy digestibility and intake, digesta retention time, and digestive organs size. Results partly supported expectations from theory, as well as revealed some unpredicted responses from the model species. For instance, the concentration of fiber and nitrogen in the dietary items differed between populations and seasons as expected from productivity. Plant cover, diversity and niche breadth, revealed a complex foraging strategy that encompasses seasonal and spatial changes in the trophic scenario. The cecum, the organ most closely related to cellulose fermentation, was significantly larger in animals facing the lowest quality food in the field. Finally, under experimentally different quality diets, *M. australis* showed behavioral and physiological compensatory mechanisms that could also reveal interpopulation differences. These results suggest that the species is plastic in its nutritional ecology, and may explain its capability to colonize extreme habitats. Its adequacy involves different response strategies that account for the ecological versatility of the species and raises new questions from an ecological-evolutionary perspective.

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S6-3.

DISTURBANCES IN THE DESERT AND THREATS TO CONSERVATION

Borghi CE¹, Campos CM², Andino N¹, Reus L¹, Giannoni SM^{1,2}.

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The desert is a system of low species richness at the local level. However, because of the ecological characteristics (e.g., low primary productivity), high environmental heterogeneity and great climate severity (e.g., extreme temperatures, low water availability, and high radiation) of this system, species of low density (rare species) and endemism at genus and species levels are frequently found. Because of these features, biological diversity of arid zones becomes highly sensitive to disturbances. Although these characteristics would place those species in a high threat category, few works have evaluated the impact of different disturbances on diversity of arid zones in Argentina. On the one hand, among the disturbances affecting biological diversity that have traditionally received most attention are overgrazing, deforestation, agricultural frontier expansion, and petroleum and mineral exploitation. On the other hand, a number of anthropogenic activities that have been poorly studied in South America include road construction and use, urban expansion, tourism development, and the use and contamination of rivers and streams; these activities, which have increased as a consequence of the improvement in the economic situation, disturb our deserts. Besides those factors, there are also some problems that have been poorly or not evaluated, such as the effect of the spread of exotic species on biodiversity, the destruction of biological crusts and natural corridors, the effect of dam construction, and the effect of the introduction of exotic fishes. Hence, the high vulnerability of desert systems, with a high rate of endemisms, is stressed, as well as the scarce knowledge of the effect of traditional and new anthropogenic disturbances on these systems.

BIOLOGY OF THE MALE GAMETE**Sat 1-1.****PARTICIPATION OF SERINE PROTEASES AND INHIBITORS. RELEVANCE IN CRYOPRESERVATION***Cesari A.**Biología de microorganismos y gametas. Instituto de Investigaciones Biológicas. CONICET-UNMDP.*

Seminal plasma contains secretory proteins from the epididymis and accessory glands among other molecules. Proteomics allowed the identification of many serine protease inhibitors in seminal plasma of most mammalian species studied. Still, the identity of their target proteases are not known, neither it is known whether they inhibit proteases on the sperm surface or within the external fluids. However, it has been demonstrated that they bind to the sperm surface and that they are decapacitating molecules. In our laboratory the role of SPINK3 (Serine Protease Inhibitor Kazal type 3) in the signaling pathways during capacitation is being studied. On the other hand, the use of cryopreserved semen is limited in cattle and lab species due to the reduction of the percentage of fertile sperm after thawing. Cryodamage induces a capacitation-like status, since membrane re-arrangements alter sperm membrane permeability, and consequently provoke an intracellular calcium increase. Seminal plasma supplementation after thawing improves semen quality and proteins involved in this effect were identified in our lab, including serine protease inhibitors. We are working in the production of these recombinant proteins in order to evaluate whether they can be applicable to reproduction biotechnology.

Sat 1-2.**IN VITRO MODELS FOR THE STUDY OF CANCER STEM CELLS IN PROSTATE CANCER***Castellón EA, Contreras HH, Huidobro C, Arias J, Valenzuela R, Lillo J, Castillo V.*

The prostate is a major gland of the male reproductive system that provides 20-30% of seminal fluid with enzymes and elements required for sperm function. This organ may present several diseases involving risk to male fertility, including prostatitis, benign hyperplasia and cancer. Prostate cancer is occurring frequently in men close to 50 years with fertility expectations. Current treatments are invasive and in some cases include orchiectomy or androgen deprivation causing, among other several adverse effects, infertility. Currently there are efforts to develop minimally invasive techniques and target therapies. In our laboratory we have developed cell culture systems from prostate tumor explants in which we have studied several aspects of this disease, such as hormone sensitivity, drug resistance and the effect of various compounds with therapeutic potential. We have recently identified tumor-initiating cells or cancer stem cells (CSC) in biopsies and cell cultures of prostate carcinoma. These cells would be primarily responsible for the metastatic potential, recurrence and resistance to the different treatments. We have obtained populations enriched in CSCs through Magnetic Cell-Associated Sorting (MACS) and induction of tumor spheres growth (prostate-spheres). The genetic, molecular and functional characterization of these cells is currently ongoing. The results show several pluripotency genes overexpressed in these cells. These genes may represent appropriate diagnostic and prognostic markers as well as new therapeutic targets in prostate cancer. This is highly relevant considering that this disease is the second leading cause of male cancer death in the world.

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Sat 1-3.**HYPERCHOLESTEROLEMIA MODIFIES SEMINAL AND SPERM PARAMETERS THAT MAY BE RESTORED BY OLIVE OIL ADMINISTRATION***Fornés MW, Sáez Lancellotti TE, Boarelli PV, Cid-Barría JL, Cid-Barría M, Romero A, Martínez V.**Área e Instituto de Histología y Embriología de Mendoza, FCM- UNCuyo; Instituto de Investigaciones, FCM – UDA y CCT-Mendoza, CONICET.*

Rabbits can be transformed into hypercholesterolemic by adding animal fat to the normal diet. This generates an increase in serum cholesterol and several changes, general ones - high blood pressure – tissular - fatty liver, damage to the capillary endothelium - and cellular - fatty degeneration of hepatocytes. But little attention was directed to the link between hypercholesterolemia and reproduction until the development of transgenic mice for receptor LXR - LXR - / -, sensors of intracellular levels of cholesterol - and low reproductive rate of the mutant mice. Not attention was also directed to the increased fat in the diet leads to changes in the male gametes and semen abnormalities, such as the low number of sperm in semen, increasing the number of malformed sperm, increasing cholesterol in the plasma membrane and loss of ability to undergo the acrosome reaction and training. Further changes are checked for example in the mRNA levels of SREBP 1c (Sterol Regulatory Element Binding Proteins 1 and 2: proteins that bind to sterol regulatory elements at the nuclear level). What is even more interesting is that the incorporation of virgin olive oil fat diets prevents the alterations mentioned. The mechanism of this protection is unknown but in the laboratory we observed a decrease in membrane cholesterol indicating that the genesis of the cell (testicular level) is corrected. Further efforts to detect the target cell and the components involved in olive oil are required, among other reasons because it would have positive effects on people with high cholesterol and also in the regional economy of Cuyo.

ICHTHYOLOGY ARGENTINA: TRENDS AND LINES OF WORK IN THE NEW GENERATIONS**Sat 2-1.****MANAGEMENT OF NATIONAL INLAND FISHERIES***Remes Lenicov M.**Dirección de Pesca Continental. Subsecretaría de Pesca y Acuicultura de la Nación-MAGyP.*

Inland fisheries of Argentina have significant importance in economic terms due to the volume exported and the social impact of its contribution to food security and regional household economies. In general terms we can clearly distinguished three types of fisheries: a) subsistence, b) commercial c) sport/recreational. The first two are performed by "artisanal fishermen" while the latter is associated to tourism. At the Río de la Plata river basin, commercial fishing concentrates its efforts on *sábalo*, whose exports in the last 4 years were around 15,000 tonnes per year. At least 20 other species are the subject of commercial fisheries with volumes not exceeding 20% of *sábalo* landings, among them the most important are *tararira* and *boga*. Sport fishing in the NEA focuses its efforts on *dorado* and *surubí*. In the Patagonia region, inland fisheries are mainly sportive and recreational, based on introduced salmonids. Inland resources are administered by the provinces or by international treaties in neighboring courses. Since 2004, under the CFA was created "Commission for inland fisheries and Aquaculture" (CPCyA) in order to harmonize policies at the basin management level and create a federal organism to develop and implementing several measures to fisheries management. The Department of Inland Fisheries has also signed agreements with provincial, national and international governments to undertake projects designed to increase knowledge of fisheries resources. Examples include. 1) Fisheries Management and Conservation of Wetlands biodiversity in the Parana and Paraguay rivers, Argentina (PNUD-GEF; Proyecto FMAM 4205). 2) Assessment of sabalo resource at the lower Paraná-Plata basin (CPCyA). 3) Assessment of fisheries resources in the lower Uruguay and inner Río de la Plata rivers (CARP- CARU). 4) Fisheries biology of main species of Paraná and Paraguay rivers (FAO) 5) Conservation of fish and fisheries from Uruguay river (CARU).

Sat 2-2.**FISH BIOMARKERS AS TOOLS FOR ASSESSING WATER QUALITY***Cazenave J.**Lab. Ictiología, INALI-CONICET-UNL, Ciudad Universitaria UNL, Paraje El Pozo, 3000 Santa Fe, Argentina.*

In recent years, the levels of xenobiotics in aquatic environments have increased due to anthropogenic activities, which could lead to a decrease in natural resources. For this reason, there has been a growing awareness to develop methods for the early detection and assessment of adverse effects of pollutants on the biota. Particularly, fish are affected by all impacts on the water body, reflecting early environmental stress situations. Our work is aimed at evaluating the use of biological responses (biomarkers) in fish to evaluate water quality changes. Thus, we carried out both laboratory and field studies, using a set of biomarkers (morphological indexes, haematological and biochemical parameters, detoxification and oxidative stress markers, histopathological analyses). In fact, we assessed biomarkers responses in 3 fish species (*Prochilodus lineatus*, *Cichlasoma dimerus* y *Piaractus mesopotamicus*) exposed to the organochlorate insecticide endosulfan, under experimental conditions. Results showed that low pesticide concentrations are able to bring about immunological disorders as well as oxidative stress induction. On the other hand, we assessed water quality of the Salado river basin by using biomarkers in *Prochilodus lineatus*. The measured biological responses revealed differences in fish health status at different sampling sites, which may reflect unfavorable environmental conditions for fish life. Summarizing, our experience demonstrates that biochemical, physiological and histological markers are sensitive tools for assessing the sublethal effects of pollutants and environmental quality; so we suggest the use of biomarkers in future freshwater aquatic systems monitoring. In order to continue identifying the most effective tools, we are currently studying the effects of toxic mixtures on fish health and biomarkers responses of fish exposed *in situ* to point sources of pollution.

Sat 2-3.**PHYLOGENY OF FISHES OF THE FAMILY CHARACIDAE (TELEOSTEI, CHARACIFORMES): CURRENT STATUS AND PERSPECTIVES***Mirande JM.**Fundación Miguel Lillo (CONICET). Miguel Lillo 251 (4000), San Miguel de Tucumán, Tucumán, Argentina.*

Objective: To establish the phylogenetic relationships of large clades within the family Characidae, with emphasis on the monophyly and classification of its subfamilies. Methods: Phylogenetic characters comes from morphological observations made by the author and DNA data of public domain. The analyses combine both morphological and molecular data and are made following the parsimony criterion under implied weighting and methods of differential character weighting recently implemented in the phylogenetic software TNT. A nodes stability criterion is used to select between results obtained under different analytical conditions, such as in most recently published phylogenies of this group. Results: Although the analyzed data matrix is still in an incipient state of development and it has a great number of missing data (more than 60% of the entries), the obtained results are relatively congruent with the morphological and molecular published phylogenies, supporting the monophyly of several of the subfamilial groups proposed in the literature. Conclusions: Combination of all the available information in a single analysis is highly desirable and, although there is still many information gaps (species of which their osteology and soft anatomy is completely unknown, or species with absolutely unknown molecular data), the results are highly promising.

Sat 2-4.**ENDANGERED SHARKS: WHAT THREATENS THEM, WHY ARE THEY NECESSARY, AND HOW SHOULD WE CONSERVE THEM?***Lucifora LO.**Centro de Investigaciones Ecológicas Subtropicales (CIES). Centro de Investigaciones del Bosque Atlántico (CeIBA). Casilla de Correo 9. Puerto Iguazú, Misiones. N3370AVQ. Argentina.*

In this work, the main conservation problems of sharks, rays and chimaeras are presented, showing how their abundance declined dramatically during the last decades, how this affects marine communities, which species are priority for conservation, and how should chondrichthyans be conserved. Abundance declines reach 99% and are distributed globally, with well-documented examples in the southwest Pacific, northwest Atlantic, Gulf of Mexico, Caribbean Sea, Southwest Atlantic (including Argentina), and Mediterranean Sea; these declines are mostly a consequence of overfishing. The best-documented cases show that the decline of predatory sharks produces trophic cascades affecting the whole marine community and that the selective exploitation of the largest skates leads to changes in skate community structure. These results indicate that chondrichthyans have important roles in marine ecosystems, since changes in their abundance produce changes in the communities they inhabit, making their conservation a necessity. Life history analyses show that the high vulnerability of chondrichthyans is a consequence of its high age at sexual maturity (higher, on average, than that of bony fishes or mammals) and that the species priority for conservation are those with the highest age at maturity, viviparous reproduction and deep-sea habitat. Analyses of the spatial distribution of chondrichthyan diversity off Argentina show that the highest fishing catches occur on high-diversity areas and that high-diversity areas occur on marine fronts. Globally, the highest species richness of sharks is found on continental shelves between 20° and 30° south or north, not along the equator. Distribution of shark diversity and endemism, representative of all biogeographic units, indicates that a strategy based solely on protected areas would be unrealistic, since a minimum of about 14% of ocean's surface should be protected. Therefore, the best strategy to conserve chondrichthyans should be a combination of protected areas and a reduction of fishing effort.

CONTRIBUTIONS TO THE DEVELOPMENT FOR REGIONAL AGRICULTURE AND FOOD**Sat 3-1.****USE OF FINGERPRINTS TO DIFFERENTIATE THE ORIGIN OF ARGENTINEAN FOODS***Wunderlin DA.**Universidad Nacional de Córdoba, Fac. Cs. Químicas-Dpto. Qca. Orgánica-ISIDSA. CONICET- ICYTAC.*

The use of modern methods based on the measurement of multiple chemical parameters in food (profiling) followed by multivariate statistics helps to construct a fingerprint characteristics of productions areas. When this fingerprint is translated to diverse foods it is possible to assess the origin, quality and, in some cases, the production method used (organic or intensive farming, feed-lot, etc.). We constructed fingerprints for different foods characteristics of Argentina (honey, wine and meat), produced at diverse areas (Buenos Aires, Entre Ríos, Córdoba, San Juan and Mendoza). To do that, we measured metals (major and minor), trace elements (lanthanides, etc.) and stable isotopic ratios. This fingerprint was constructed for soil, irrigation water and foods. For wines samples we also determined volatile organic compounds (VOCs) and polyphenols adding organic composition to the fingerprint. Data matrixes were treated by chemometrics pointing out parameters that enable differentiation between diverse origin, varieties, etc. So far, we were able to differentiate honey from three producing areas in Argentina (Tandil, Gualaguaychú and Córdoba), verifying differences with honey produced in Europe. Also wines from Mendoza, San Juan and Córdoba were distinguished by analyzing their respective fingerprints. Furthermore, differences between the same variety produced in diverse areas was possible, including varieties typical from Argentina (Malbec, Syrah and Bonarda). Finally, we assessed differences in meat (beef) arising from cattle grew at three different locations (Tandil, Gualaguaychú and Córdoba) considering their fingerprints. In this last case, some differences in cattle diet were also observed. We conclude that fingerprint methods are quite useful to evaluate food traceability, helping to the claim of origin of Argentinean foods as well as preventing frauds ensuring the quality and composition of our products.

Sat 3-2.**MICROBIAL INTERACTIONS IN INDUSTRIAL FERMENTATIONS**

Maturano YP¹, Rodríguez Assaf LA¹, Toro ME¹, Nally MC¹, Castellanos de Figueroa L^{1,2,3}, Combina M^{4,5}, Vázquez F¹.

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Mixed culture biotechnology (MCB) could become an attractive addition or alternative to traditional pure culture based biotechnology for the production of chemicals and/or industrial products. Owing to the use of different kind of mixed cultures, process development in mixed culture biotechnology can only be based on natural/ecological selection by manipulating the operation of the bioprocess or by varying the source of the natural inoculum. Compared with pure culture based industrial biotechnology, specific advantages of MCB include: adaptive capacity owing to microbial diversity and the capacity to use mixed substrates. Conducting wine fermentations by controlled inoculation of mixtures of different yeast starter cultures is one strategy to harness the unique activity of such yeasts. Now attracts greater interest because of its potential to introduce specific characteristics into wine and also because winemakers have a more thorough knowledge of the ecology and biochemistry of wine fermentation and how to manage this process. In this context, it may be of interest to create defined mixed starter cultures in which *Saccharomyces cerevisiae* does not overcome the non-*S. cerevisiae* species in the first stages of fermentation. Strategies such as sequential inoculation of controlled fermentation processes resulting in wines with a wide range of flavour compositions. At the Biotechnology Institute, the ability of yeasts to produce extracellular enzymes of enological relevance (β -glucosidases, pectinases, proteases, amylases and xylanases) in pure and mixed *Saccharomyces*/non-*Saccharomyces* was determined throughout fermentations. Microorganisms employed were *Saccharomyces cerevisiae* BSc562, *Hanseniaspora viniae* BHv438 and *Torulasporea delbrueckii* BTd259, in pure and mixed cultures at different inocula proportions. Microvinifications were carried out with fresh must from *Vitis vinifera* L. c.v. Pedro Jimenez, an autochthonous variety of Argentina. Yeasts isolations studied in this work produced throughout the fermentations a broad range of enzymatic activities of enological interest that catalyze hydrolysis of polymers present in grape juice.

Sat 3-3.**ANTI-INSECT PROPERTIES OF ESSENTIAL OILS AND MONOTERPENES. REVALUATION OF ARGENTINE AROMATIC NATIVE SPECIES**

Zygodlo J.

Instituto Multidisciplinario de Biología Vegetal- CONICET Cátedra de Química Orgánica, Facultad de Ciencias Exactas, Físicas y Naturales, Universidad Nacional de Córdoba.

Essential oils, secondary metabolites of aromatic plants, in many countries represent an important part of the traditional pharmacopoeia. Essential oils, known for its medicinal properties and scents are natural mixtures may contain various components in different concentrations. This chemical diversity is the product of its dual biosynthetic origin: the path of mevalonic / 1-deoxy-D-xylulose 5-phosphate and shikimic pathway. This results in a wide range of applications such as deworming, bactericides, fungicides, insecticides and virucidal. Thus, that essential oils are shown as an important source for bioactive principles. This work is a description and analysis of potential mechanisms of biological action of the constituents of essential oils: octopaminergic receptor, the GABA receptor, acetylcholinesterase and P450. The results show the essential oils and their components as excellent insecticides and larvicides. It showed the difficulties to determine structural features that determine the bioactivity and quantitative structure-activity relationships for some terpenes in their interactions with different insect of health and nutrition. This exhibition will highlight the potential use of our flora aromatic and diversity of its essential oils to control insects.

Sat 3-4.**BIOSUPPRESSION GRAPE FUNGAL PATHOGENS IN INTEGRATED MANAGEMENT OF FUNGAL DISEASES**

Nally MC, Pesce VM, Muñoz MA, Radicetti DS, Toro ME, Castellanos de Figueroa LI, Vázquez F.

Instituto de Biotecnología, Facultad de Ingeniería, Universidad Nacional de San Juan.

Introduction: San Juan province is the main producer and exporter of Argentine table grapes and in 2010 export totaled about 51,776 metric tons. However, when fruit is exported to foreign countries prolonged periods of postharvest disease control are required. These pathogens can be controlled on grapes with pre and postharvest fungicidal treatments. The emergence of fungicide resistance and increasing consumer demands for reduction in residues on fruit emphasize the need for alternative disease control strategies. Biological control of postharvest diseases of fruits and vegetables by antagonistic yeasts seems promising in replacing or reducing the use of synthetic fungicides. Objective: Evaluated of population dynamic of biocontrol yeasts in grapes. Materials and Methods: 1- Population profiles of yeasts: The ability of biocontrol yeasts to survive and multiply in grape wounds was determined as follows: grape berries were rinsed with water, wounded, inoculated with 20 μ l of a washed cell suspension (106 cells/ml), placed on plastic trays (80% RH) and incubated at 25°C. The grape sample was placed in 10 ml of sterile 0.05 M phosphate buffer, pH 7.0. Then, dilution series in sterile distilled water were spread onto YEPD-Agar and incubated at 25°C. Colony counts were carried out after 3 days. Results: Of the 59 biocontrol yeasts that were inoculated in wounds of grapes at 25°C, 24 yeasts significantly increased their population in grape Red globe. Conclusions: Many yeast species are able to colonize surfaces for long periods of time, characteristics that are relevant in the selection of biocontrol agents.

1.

AH3 - LUNG STRUCTURES FOR GAS EXCHANGE AND HEMOPOIESIS IN *Pomacea canaliculata* (ARCHITAEENIOGLOSSA, AMPULLARIIDAE)

Rodríguez C. Vega IA, Castro-Vázquez A. IHEM-CONICET y FCM-UNCuyo. E-mail: acaastrovazquez@gmail.com

Ampullariid snails have both a gill and a lung, which allows them a certain degree of amphibious life. The lung is a flattened pouch ventilated through a siphon, and three distinct tissue arrangements can be recognized in its wall: (1) a fibromuscular and vascular structure (about 1 μm thick) occupying most of the roof of the lung, which is heavily loaded with hemocytes and may serve hemopoiesis; (2) a massive layer (about 3 μm thick) of urate-storing tissue which forms all of the floor and part of the roof of the lung pouch; and (3) a thin (about 1 μm) vascular lamina wholly covering the inner surface of the pouch, i.e., both the roof and the floor structures, which may serve gas exchange and is in contact with air. The hemopoietic structure is a complex arrangement of both muscular and connective tissue fibers, with intersped groups of glycogen-storing cells and hemocyte nodules. The urate-storing structure is made by urocytes radially arranged around large vessels (i.e., as perivascular urate tissues found in other organs of this snail). The gas-exchange structure is formed by a single layer of capillary-like vessels, which are also in tight contact to each other, and which cover the entire inner surface, being in direct contact with air contained in the lung pouch. A more detailed study involving 3-D reconstruction of the lung structures possibly serving hemopoiesis and gas exchange is in progress in our laboratory.

2.

AH6 - MICROGRAPHIC & MORPHOANATOMICAL CHARACTERIZATION OF *Schkuhria pinnata* (ASTERACEAE: BAHIEAE)

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Schkuhria pinnata (Lam.) Kuntze ex Thell., commonly known as "canchalagua" or "matapulgas", is a South American annual herb used in popular medicine as a depurative, antiseptic, for slimming, and as insecticide against fleas and lice. It has shown antimalarial and antifungal activities. This work was carried out to contribute to the botanical characterization and quality control of the commercial drug. Stem and leaf anatomy as well as quantitative micrography of fresh and preserved (formalin-acetic acid-alcohol) wild samples were studied. Voucher specimens are preserved at Herbarium UNSL. Leaves 1-2 pinnatisect, segments linear. Capitula narrow, in broad peaks, filarial dotted-glandular. Flowers pistillate 1, bilabiate, and perfect 5-6, wide blade. Cipselae pyramidal, pappus 8 scales or mutic. Leaf with thick cuticle, mesophyll isolateral, parenchymatic sheath around bundles, only midrib protected by collenchymatic caps. Stem costate, collateral vascular bundles and outer sclerenchymatic cap. Micrographic parameters: stomatal number (SN upper surface (us) = 7.83 ± 1.02 ; SN lower surface (ls) = 10.16 ± 1.33); stomatal index (SIus = $11.07-13.04$; SIls = $11.93-15.15$); palisade ratio (PR = $8.37-10.17$); vein-islet number (VIN = 9.33 ± 1.4) and veinlet-termination number (VTN = 7.06 ± 0.81). This study contributes to an effective quality control mainly in drugs crushed, milled or reduced to powder.

3.

AH7 - MORPHOANATOMY OF *Thelesperma mega-potamicum*, "TÉ PAMPA" (ASTERACEAE)

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Thelesperma megapotamicum (Spreng.) Kuntze ("té pampa" or "té indio", Asteraceae: Coreopsidae) is a perennial herb with bicentric American distribution, native to Central and Southern Argentina and Uruguay. It grows from Santiago del Estero to Río Negro. Aerial parts are used in folk medicine (digestive, antispasmodic and for liver and kidney diseases). Macro- and micro-morphological and quantitative micrographic studies were carried out because the lack of bibliographic data about pharmacobotanic characterization. Material was obtained from natural populations in the province of San Luis. Techniques were performed by cut, paraffin embedding, staining, diaphanization and dissociation. Photomicrographs were obtained with a Leitz DMRB microscope with digital camera. Leaf segment transections show a semicircular section with bilateral structure. The vascular bundles and parenchyma sheath present intravascular secretory canals. Stem transections are round; the epidermis shows a thick cuticle while cortical area has angular collenchyma and spongy chlorenchyma. The vascular bundles are arranged in a typical eustela, protected by sclerenchyma. Schizogenous secretory canals alternate with vascular bundles. Medullary parenchyma has large isodiametric cells when young, and when mature is fistulous. Micromorphological data allows characterize the drug, being useful in quality control especially when it is reduced to powder.

4.

AH8 - MORPHOANATOMY OF *Gochnatia glutinosa* (ASTERACEAE: GOCHNATIEAE)

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Gochnatia glutinosa (D. Don) Hook. & Arn. ("jarillilla", "jarilla pispá") is a perennial leafy and resinous shrub endemic to Argentina, growing along "Monte" Biogeographical Province, from Catamarca to Neuquén. Aerial parts are used in folk medicine as pedic deodorant, rubefascient, anti-inflammatory and antioxidant, as well as other species related to "jarillas" (*Larrea* spp., Zygophyllaceae). Given the importance that these species have acquired and lack pharmacobotany information, macro-, micromorphological and quantitative micrographic studies is being carried out. Plant material was obtained from Sierras of San Luis. Techniques were performed by cut, paraffin embedding, double staining with different dyes, diaphanization and dissociation. Photomicrographs were obtained with a Leitz DMRB microscope with digital camera. Leaf transections show amphistomatic (raised stomata) and bilateral structure. Vascular bundles are surrounded by a sclerenchymatic sheath. Stem cuttings show a notable endodermis and vascular bundles protected by sclerenchyma. Both leaves and stems have abundant resin secreting glands and glandular multicellular trichomes of two types: uniseriate multicellular and Malpighian ("T") hairs. These data allows characterize the species, being very useful in quality control especially when the material is finely ground or powdered.

5. AH9 - MORPHOLOGICAL AND IMMUNOHISTO-CHEMICAL DESCRIPTION OF TESTIS VISCACHA (*Lagostomus maximus maximus*) IN RELATION TO AGE

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In the male reproductive system, androgens are essential for activation of the hypothalamic-pituitary, testicular descent, control the number of Sertoli cells, start of spermatogenesis and sexual development. The androgen receptor (AR) plays a key role in androgen action. In our experimental model, viscacha, the adult male has an annual reproductive cycle with a period of maximal gonadal activity in summer and minimal in winter. The objectives of this study were to describe the morphology of testes of immature and adult animals to determine immunohistochemically the expression of AR and relate the results found with the animal's age. Testicular tissue samples from immature and adult viscachas were extracted and processed by conventional optical microscopy. The ARs were immunohistochemically determined by using the antibody AR (N-20): SC-816. Morphological results obtained in testes of immature animals showed a disorganized germinal epithelium constituted of numerous primitive germ cells and Sertoli cells. The diameter of the seminiferous tubule in immature animals was $59.7 \pm 0.75 \mu\text{m}$, achieving a value of 137.6 ± 1.67 in adult animals. The RA in immature animals are mainly in germ cells, whereas in adult animals are found in the Sertoli cells. In conclusion, these results suggest that in the first stage of the maturing testicular, androgens promote the development of germ cells to initiate spermatogenesis. In adults the expression of receptors is higher in Sertoli cells, which will be responsible for ensuring the efficient development of spermatogenesis.

6. AH10 - INTRAMAMMARY INOCULATION OF *Lactobacillus perolens* CRL1724 IN DRY PERIOD DAIRY COWS

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Dry period antibiotic therapy is the practice most used for preventing bovine mastitis. We studied the effect of *in vivo* inoculation of *Lactobacillus perolens* CRL1724 (LP) in the bovine mammary gland. Two healthy dry period cows were used to determine the higher concentration of LP that does not produce inflammation, three quarters were inoculated (IQ) with 1ml of 10^3 , 10^6 and 10^9 CFU/ml and one quarter as control. Other animal was used for histological analysis and was IQ with 10^6 CFU/ml. Daily milks samples were collected and clinical signs analyzed during 7 days. Somatic cells counts (SCC) bacteriology and recovery of LP were determined. Teat canal and cistern samples were removed and fixed in formalin and glutaraldehyde for transmission electron microscopy (TEM). LP in numbers of 10^6 CFU/ml did not produce clinical signs in the udder or changes in milk. The SCC increased up to 48h compared to control. LP was recovered in all IQ and no mastitis pathogen was isolated. No histological differences neither presence of LP was observed between samples of teat canal tissue of IQ and not IQ. PMN cells, hyperemia of blood vessels and adhesion of LP was observed in the cisterns samples. LP adheres to the teat cistern without cell damage or modifications. This study provides information for the design of a probiotic product to prevent bovine mastitis in dry cows.

7. AH14 - EFFECT OF MELATONIN ON PITUITARY FOLLICULOSTELLATE CELLS OF *Lagostomus maximus maximus*

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In several species the folliculostellate cells (FSC) have been described. Aim of this work was to study the effect of melatonin administration on FSC of pituitary pars distalis and pars intermedia of viscacha. Immunohistochemistry and image analysis were used. The percentage of S-100-positive area (total = 1, cellular = 2 and colloidal = 3) and the number of folliculostellate cells (4) were determined in controls and melatonin administrated animals.

	Pars Distalis		Pars Intermedia	
	Control	Melatonin	Control	Melatonin
1	1.08 ± 0.11	0.65 ± 0.05^a	9.50 ± 0.59	5.44 ± 0.35^c
2	0.73 ± 0.06	0.47 ± 0.05^a	7.63 ± 0.52	4.03 ± 0.43^b
3	0.34 ± 0.04	0.20 ± 0.03^c	1.82 ± 0.23	0.33 ± 0.12^c
4	0.41 ± 0.02	0.30 ± 0.02^b	3.22 ± 0.40	1.46 ± 0.19^c

The administration of melatonin caused a significant decrease of immunostaining (**a** = $p < 0.001$; **b** = $p < 0.01$; **c** = $p < 0.05$). Consistent with previously reported seasonal changes, these results demonstrate that the expression of S-100 protein in FSC is regulated by melatonin. These cells are probably involved in the secretory activity regulation of pituitary gland through S-100 protein production.

8. AH15 - IMMUNOHISTOCHEMICAL CHARACTERIZATION OF THE INTERSTITIAL CELLS PRESENT IN THE PINEAL GLAND OF *Lagostomus maximus maximus*

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Since three molecular markers have been found in the interstitial cells of the pineal gland in different mammalian species (rat, sheep, monkey and mouse): S-100 protein (glial marker), glial fibrillary acidic protein (GFAP; astrocyte marker) and vimentin (immature glial cell marker), there is little information about the possible functions of these cells. The current report is part of a program dealing with the control and regulation of neuroendocrine axis and its influence on the reproduction of *Lagostomus maximus maximus*. Here, an immunohistochemical study of the pineal gland's interstitial cells of adult male rodents captured in summer is presented. The interstitial cells were abundant towards the glandular stalk and they were scarce or absent towards distal region. The studied molecular markers were distributed according to the following pattern: S-100 in nucleus and/or cytoplasm, GFAP in cytoplasmic projections and vimentin in the cytoplasm of cells localized close to blood vessels. The cytoplasmic projections of the interstitial cells contacted the pinealocytes and some blood vessels. These findings suggest that the interstitial cells have a neuroectodermic origin and probably are involved in the paracrine regulation into the pineal gland. On the other hand, the presence and localization of vimentin would indicate the presence of a reserve population of glial cells.

9.

AH17 - MORPHOLOGICAL CHANGES IN SEMINAL VESICLE OF THE VIZCACHA (*L.m.m*) IN RELATION TO REPRODUCTIVE CYCLE AND AGE

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The objective of this work is to study morphological variations seasonal in seminal vesicles (SV) of viscacha and describe the SV in immature males. The viscacha is a seasonal reproduction rodent, with maximal activity in summer and minimal in winter. SV of adult and immature male viscachas were processed by optical microscopy and electron microscopy. The mucosa of the SV has 3 cell populations: principal (P), basal (B) and clear (Cl). Activity: P cells are high, the basal nucleus, loose chromatin and nucleoli evident. It contains abundant mitochondria and rough endoplasmic reticulum (RER) in the supranuclear region. Cl cells are located between P cells, have lax chromatin and cytoplasm with little RER and absence of Golgi. B cells are small and arranged in the base of the epithelium. They have lax chromatin and scant cytoplasm. Regression: P cells have irregular nuclei and condensed chromatin peripherally located. Their cytoplasm contains few mitochondria, RER with dilated cisterns and glycogen granules. Cl and B cell no significant changes. In conclusion, during the regression period, SV of adult animals have features of inactive glands. The nuclei of irregular borders, mitochondria and RER decreased and increased cytoplasmic glycogen indicates lower epithelial activity, similar to that observed in immature animals, whose SV are stratified epithelium. It is postulated that the low cellular activity observed in the glandular epithelium of adult animals is due to decreased androgen levels during the regression.

10.

AH18 - VARIATIONS IN THE EXPRESSION OF CARBOHYDRATES IN THE INTESTINE OF *Corydoras paleatus* JENYNS, 1842 (SILURIFORMES, CALLICHTHYIDAE)

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In shallow freshwater, where hypoxia is a high probability, inhabit fishes that breathe atmospheric O₂. Among them is the genus *Corydoras*, teleost of cosmopolitan distribution of South American, which used to air breathing the posterior area of the intestine. Our objective was characterized by traditional histochemical and lectin histochemical the pattern of carbohydrate in the intestinal mucosa. We used adult specimens that were sent from a local fish farm and collected in sources of water near the urban of La Plata. Samples were fixed by immersion in 10% buffered formalin and routinely processed and embedded in paraffin wax. Subsequently, the sections were incubated with biotinylated lectins battery. LSAB system was used for detection, diaminobenzidine as chromogen and haematoxylin as a contrast. The observation was performed using immersion objective. To locate and distinguish glycoproteins (GPs) of the mucosal cells (CM), we used the following histochemical methods: PAS; PAS*S; KOH/PA*S; PA/Bh/KOH/PAS; KOH/PA*/Bh/PAS; Alcian Blue and Toluidine Blue at different pHs. The lectin histochemical analysis of the intestinal mucosa showed that PNA, SJA, LEA and BS-I showed no affinity to any of the structures analyzed. The intensity of the binding in the rest of lectins showed a weak to strong in different regions of the intestine. It demonstrated the presence of terminal residues of glucose, mannose and galactose, being CON A highly homogeneous, while RCA-I, VVA and SBA varied in different regions. The CM for some lectins such as SBA and DBA was heterogeneous, in these cells also showed a wide variety of GPs, which would relate to the different functions of the CM, such as lubrication, protection against pathogens and ionic regulation. These results demonstrate that intestinal mucosa contains a large variety of monosaccharide residues, probably related to functional differences.

11.

AH22 - MICROSTRUCTURE OF THE ENAMEL IN HUMAN TEMPORARY MOLARS

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We analyze the microstructure of enamel according to levels of increasing complexity: crystals, prisms, enamel, pattern and dentition. The objective was to identify the types of human enamel in temporary teeth with a technique applied to the other mammalian enamel and establish functional relationships. Six temporary upper molars cleaved or extracted, were included in acrylic resin, worn, prints with acid and observed in a environmental scanning electron microscope (ESEM) FEI Quanta200. The prisms level identified external aprismatic enamel thickness μ 12-40. In the cusps enamel with bands were observed in arches form since the limit with the dentin towards the apex of the same and radial external enamel. In the internal slopes, bands occupy little thickness and external slopes occupy 2/3 of the enamel, in relation to the radial. In the deep fissures enamel is irregular and there are bands to the surface. It was not found compatible irregular enamel to "knotted" enamel of optical microscopy. Free face bands were frequently found in the third half, in cervical the only type of it was radial. The irregular enamel and with bands are located in the internal portion and the radial in the external. Types of enamel meet function: the radial resists abrasion, and irregular and bands prevent the propagation of fractures. Their combination makes it possible to define patterns and relate them to the functional areas of the teeth.

12.

AH23 - HISTOLOGY OF THE INTERRENAL GLAND OF *Rhinella arenarum* (AMPHIBIA: ANURA): ANTEROPOSTERIOR VARIATIONS

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Interrenal gland of anurans is composed of different cell types: steroidogenics, which synthesize aldosterone and corticosterone; chromaffins, which produce catecholamines; ganglionars, sympathetic neurons; and in some species Stilling cells, of uncertain function. The aim of this work is to analyze the localization of these cell types on the interrenal gland of the male toad *R. arenarum* and its antero-posterior variations. Serial cross and parasagittal sections were stained with hematoxylin-eosin, Masson's trichrom and PAS. Steroidogenic and chromaffin cells were identified by immunohistochemistry with antibodies against the 3β-hydroxysteroid dehydrogenase/ isomerase and tyrosine hydroxylase, respectively. Two steroidogenic cell types were identified: acidophilic vacuolated cells and basophilic highly immunolabeled granular cells. Both cell types are organized in cords. Basophilic cells localize in large central groups that decrease their size in the posterior region. Chromaffin cells are found isolated or in small groups near blood vessels, scattered throughout the interrenal. Ganglionar cells and nerve fibers are found innervating steroidogenic cells. PAS positive cells surrounded by steroidogenic cells could correspond to Stilling cells. In conclusion, two steroidogenic cell types of unknown functionality were found in the interrenal of *R. arenarum* and their proportion varies throughout the antero-posterior axis.

13.

AH24 - HISTOLOGY OF THE STORAGE ORGAN OF WILD MACA, *Lepidium meyenii* (BRASSICACEAE)

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Samples of the underground reserve organs of wild Maca (*Lepidium meyenii*) from Calchaquies Summits of the province of Tucumán were fixed in FAA and embedded in paraffin in order to study their anatomy.

Wild maca storage organ is a contractile root with normal secondary growth. It has a differentiated peridermis from outer cortical parenchyma cells, cortex and star shaped pith, constituted by amiliferous parenchyma with idioblasts of myrosin. Also, it has secondary phloem and xylem with abundant parenchymatous rays. Tissue from contractile area adjusts to the shortening of the root caused by vertical contraction and radial expansion of the inner cortical parenchyma. The mechanism of contraction is discussed.

14.

AH28 - EVALUATION OF SURFACES FOR INTESTINAL EPITHELIAL CELL CULTURE

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The intestinal epithelium is rapidly renewed through a process of differentiation of stem cells that migrate from the crypt along the major axis of the villi. During this process there have been identified several factors influencing this phenomenon, including the extracellular matrix. The study aims to assess different surfaces for intestinal epithelial cell culture. Studied surfaces were polyethylene terephthalate (PET) from Nunc and Grenier Bio-One, collagen type I (Invitrogen) and Matrix cell (BDBioscience). The primary culture was performed according to the protocol developed by Campbell *et al.* 2008. The culture medium used, D-MEM supplemented with 2.5% FBS, insulin 10 mg/ml, transferrin 5.5 mg/ml, selenium 0.67 mg/ml, porcine mucosal heparine 50 µg/ml, penicillin 50 U/ml and streptomycin 50 µg/ml, was incubated at 37°C and 5% CO₂. The medium was renewed every other day and growth evaluation was performed daily using an inverted microscope. Dissimilar results were obtained according to the surface to be evaluated. Cell growth was apparent on PET Nunc surface. We obtained an homogeneous monolayer of cells with epithelial morphology that lasted up to 22 days. On the surface PET Grenier Bio-One, cell proliferation was practically nil. Regarding biological surfaces, in both were cell growth, but only achieve to obtain a homogeneous monolayer with epithelial morphology alone on collagen. We obtained better adherence of cells in the first 24 hours of culture in biological surfaces and longer confluence time in the plastic surface. Subcultures were not positive in none of the tested surfaces. Conclusion: Collagen surface was found to be most suitable for the proliferation of intestinal epithelial cells.

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15.

BA3 - GLUTAMINE PREVENTS INTESTINAL CALCIUM ABSORPTION INHIBITION TRIGGERED BY MENADIONE MAINTAINING THE REDOX STATE

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Glutamine (GLN) is an amino acid with restorative properties of the redox state in tissues exposed to oxidant drugs. We have demonstrated that menadione (MEN) decreases intestinal Ca absorption triggering oxidative stress and apoptotic death of enterocytes. In this work we explored whether GLN could prevent the inhibitory effect caused by MEN on the Ca absorption. Chicks (4 weeks old Cobb-Harding) were treated with 1g GLN/kg bw *per os* and with 2.5 µmol MEN /kg bw intraperitoneally (60 or 90 min later). Both drugs acted together for 30 min. Controls were treated with vehicle. We measured intestinal Ca absorption by "in situ" ligated loop technique. The activities of enzymes superoxide dismutase (SOD) and catalase (CAT) and total glutathione content (GSH) were determined by spectrophotometry. GLN administration 60 or 90 min before MEN injection prevented the fall in intestinal Ca absorption triggered by MEN. GLN increased GSH levels, restoring the control values. GLN avoided the induction of SOD and CAT activities by MEN. These data suggests that GLN is capable to prevent the decrease in the intestinal Ca absorption caused by MEN, probably by the restoration of control values of the GSH content and the activities of SOD and CAT.

16.

BA5 - EFFECT OF FOOD RESTRICTION ON BIOCHEMICAL PARAMETERS OF THE COMMON PIGEON (*Columba livia*)

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Climate change has been associated with a mismatch between the energy requirements of birds and the availability of food resources. Lack of food has implications for the metabolism of animals. Hematology and serum biochemical constituents are useful indicators to test the health and nutritional status of animal populations. In this study were monitored parameters hematological and plasma biochemical during process fast-feedback of the common pigeon to determine changes in them. The pigeons (n: 11) received water ad lib and were subjected to total food restriction until a body mass loss of 30-35% (day F) on the initial day (day 0). After, were fed back to regain their initial weight (day RF). On days 0, F and RF blood samples were taken. Capillary hematocrit was measured using a standard table and was used the Natt and Herrick method for red cell count and blood metabolites in plasma were measured using commercial kits. The erythrocyte count and hematocrit decreased after fasting, corresponding to the condition of the birds and the initial values were recovered after being fed back. The birds showed a profile similar to the classic model of fasting compared to biochemical changes except in glucose levels. These results contribute to the understanding of fasting and feedback processes that birds may suffer as a result of climate change.

17.

BA6 - ULTRASTRUCTURAL ASPECTS AND CELL DEATH BY APOPTOSIS IN THE OVARY OF *Myiopsitta monachus* AND *Zenaida auriculata**Bulfon M, Bee de Speroni N.**Anatomía Comparada. Facultad de Ciencias Exactas, Físicas Naturales - UNC. E-mail: mbulfon@com.uncor.edu*

We analyzed the ultrastructure and cell death by apoptosis in order to provide a contribution to knowledge of the ovary of these birds. The ovarian follicles exhibited ultrastructural changes according to development. A characteristic of primary oocytes and previtellogenic follicle (80 a 170 μm) was the Balbiani's vitelline body. In previtellogenic and vitellogenic follicle ≥ 2 mm, the Golgi apparatus, numerous mitochondria, RER and REL and transosomes, involved in early yolk deposition, were the most evident organelles. In yellow vitellogenic follicles ≥ 4 mm, there were yolk granules and a marked zona radiata. Follicular involution manifested by hiperplasia of the follicular epithelium and decrease of the transosomes, and subsequent rupture of the zona radiata and disrupted organelles. Lysosomes, vacuoles, cellular debris and macrophages characterized the necrosis on the later stages of atresia. The marking of fragmented DNA in atretic vitellogenic follicles ≥ 2 mm of the *Z.auriculata* revealed apoptotic cells in the follicle cells. In the ovary of both species simultaneously developed a mechanism for growth and differentiation and a regression process characterized by cell death by apoptosis and necrosis, among others.

18.

BD3 - INMUNOHISTOCHEMICAL ANALYSIS OF CARBOHYDRATE EXPRESSION DURING RAT EMBRYONIC DEVELOPMENT*Ferretti VA, Segal-Eiras A, Croce MV.**CINIBA, Faculty of Medical Sciences, UNLP, La Plata.*

It has been suggested that the number and complexity of carbohydrate on cell surfaces may have important roles in the embryonic development. The aim of this study was to compare the expression and tissue specific localization of Tn, TF and Lewis x carbohydrate epitopes during rat embryonic development. 8 fetuses of each gestational stage from 13 to 20 days of gestation (D13-D20), 8 neonates and eight adults were studied. Immunohistochemistry was performed employing anti-Tn, anti-TF and anti-Lewis x monoclonal antibodies. Tn was expressed in the renal tubules and intestinal epithelium at the D17. In the salivary glands, the acini showed reaction on D18. The gastric glands and the epidermal tissue showed Tn expression on D19. In neonates and adults, expression in the same tissues was observed, except for the epidermis. TF was detected in the esophageal and gastric epithelia from D16. From D18, reaction was observed in the excretory ducts and acini of salivary glands, in the distal tubules and renal corpuscles and at superficial layers of the epidermis. TF was also detected in intestinal glands and luminal epithelium on D20 while in neonates and adults, it was found in the same tissues, except for the esophageal epithelium. Lewis x was detected from D17 in gastric epithelia, colon, renal tubules and skin and from D18 in small intestine. In neonates and adults, expression in the same tissues was observed, except for the epidermis. The results showed that TF, Tn and Lewis x expression depends on the time of development and also the tissue considered.

19.

BD4 - LEVELS OF ESTROGEN AND PROGESTERONE ASSOCIATED WITH INCUBATION PROCESS IN *Tupinambis merianae**Chamut S¹, García Valdez MV¹, Jahn G², Arce O¹, Manes ME¹.**¹FAZ, UNT. El Manantial (4105) Tucumán. ²IMBECU CRICYT (5500) Mendoza. E-mail: mmanes@faz.unt.edu.ar*

During the reproductive cycle *Tupinambis merianae* displays a set of maternal behaviors related to the incubation of eggs. These behaviors, such as nest construction, the contribution of temperature and humidity to incubation and protective responses are unusual among reptiles. We compare the plasma levels of estrogen and progesterone in females while actively incubating (ovulatory reproductive cycles) with those that are not (anovulatory reproductive cycles followed by follicular atresia).

Studies with RIA showed that incubating females exhibit plasma levels of estrogen and progesterone greater than those that are not incubating. These differences are significant in the case of estradiol. Preliminary histological studies of the gonad during incubation revealed the presence of corpora lutea-like structures. It is proposed that these structures are responsible for the higher plasma concentrations of both hormones.

20.

BD6 - LECTIN-BINDING CHANGES IN EARLY STAGES OF PREGNANCY IN CADMIUM INTOXICATED RATS*Díaz MC¹, González N², Najle R¹, Barbeito CG^{2,3}.**¹Univ Nac Centro Prov Bs As, Fac Cs Vet. ²Univ Nac La Plata, Fac Cs Vet. ³Conicet. E-mail: maridel@vet.unicen.edu.ar*

Cadmium is a non-essential heavy metal for living beings that is toxic to plants, animals and humans. There are no homeostatic mechanisms that regulate its metabolism. The aim of this work was to analyse the effect of a single cadmium chloride dose (10 mg/kg body weight) in the uterine horns and placentas of dams treated on days 4 and 7 of pregnancy and euthanized on days 7 and 10 (groups I and II, respectively). Organ samples were processed conventionally for lectin histochemistry (DBA, Con A, SBA, PNA, UEA-I, RCA-I, and WGA). Results for group I, when compared to controls, showed: 1) reduced PNA, DBA, and RCA-1 staining in the uterine epithelia; 2) increased affinity for SBA staining and reduced affinity for RCA-1, and Con-A in decidua, and 3) reduced RCA-1 labeling in endothelial cells. For group II only an increased Con-A marking in the uterine glands was found when compared to controls. Changes in the carbohydrates pattern of uterine horns and placenta might modify cell interactions and thus contribute to generate embryonic alterations.

21.

BD10 - SEXUALLY DIMORPHIC EXPRESSION OF SOX-9 AND AMH IN THE GONAD OF *Caiman latirostris*

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Caiman latirostris exhibit temperature-dependent sex determination (TSD) and sexual reversion has also been demonstrated after in ovum estrogen/xenoestrogen exposure, called hormone-dependent sex determination (HSD). The presence and levels of expression of sex-determining genes are unknown for this species of caiman. We isolated gonad-adrenal-mesonephros (GAM) complexes and collected serum samples from ten-day-old caimans. We quantitatively compared the mRNA expression of *amh* and *sox-9* on GAM complexes from TSD males (n=10), TSD females (n=10) and HSD females (n=7) (born from E₂-exposed eggs incubated at the male-producing temperature). Serum levels of E₂ and testosterone were measured. *amh* and *sox-9* mRNA expressions were higher in the GAM of TSD males than TSD and HSD females. Testosterone serum levels were significantly higher in TSD males, whereas E₂ levels were higher in both TSD and HSD females. In neonate caimans, *amh* and *sox-9* mRNA expressions and gonadal steroid hormones exhibited sexual dimorphic patterns. No differences in the studied parameters were found between TSD and HSD females.

22.

BF4 - COMPARATIVE STUDY OF ERYTHROCYTE AGGREGATION (EA) IN 100 DAYS RATS OF LINES b AND β

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In this work we compared EA between 100 days rats of line “b” and line “β”, since line “b” animals were eumetabolic upon arriving at this Faculty but next generations developed rise in weight and glucose intolerance. The objective was to analyze the probable evolution of the behavior of 100 days line “b” animals to the exhibited by line “β” animals (rat model of Obesity and Diabetes). We used male rats, line b and β of 100 days (n = 11). The EA was determined by optical method in suspensions of red blood cells (RBs) in plasma and in Dextran 500 2% in saline (Htc: 40%) We determined: T (estimating the size of the aggregates) and V (estimating the initial rate). EA results were analyzed with t Student test, expressed as mean ± SD. T: line b: 1.76 ± 0.08, line β: 1.78 ± 0.08; V: line b: 0.38 ± 0.10, line β: 0.49 ± 0.18. There was no EA in plasma for both lines. The results in Dextran showed significant differences in V parameter but no differences in T parameter. We conclude that 100 days animals of line “b”, which are at puberty period, were beginning to express similarities with those of line “β”, probably related with developed of mechanical changes at cellular membrane.

23.

BF5 - ERYTHROCYTE SHAPE AND OSMOTIC FRAGILITY ASSESSMENT OF THE IIMb LINE OF RATS OF 100-DAY-OLD

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The inbreeding IIMb(b) line of rats was considered normal control of the beta line (obesity and diabetes model). Recently, the IIMb line started presenting variability in glucose tolerance; therefore it was necessary to characterize it in different aspects. One of them is rheology. In this study we investigate the osmotic behavior and the erythrocyte shape (ES) of red blood cells (RBC) suspensions of the IIMb and beta line of rats (100 days). Males from the IIMb (n=6) and beta (n=7) lines were used. The ES was determined by microscopy and osmotic fragility (OF) was studied photometrically at 540 nm, reporting: x₅₀ (concentration of NaCl obtaining 50% of hemolysis) and β (cell population response). Statistic: U-Mann-Whitney test. The results were expressed as median (rank). Differences were considered significant if p<0,05. OF: IIMb line: x₅₀ (mM):74.76 (58.06-84.19)^{ns}; β: 0.064(0.062-0.082)^{ns}; beta line: x₅₀ (mM):67.05(58.25-88.16)^{ns}; β:0,061(0.047-0.07)^{ns}. ES: stomatocytes 4 and 3 in both lines. Although the beta line at 100 days-old presents higher body mass, his metabolic alterations appear at 180 days old. In this study, the obtained results indicate that erythrocyte membrane of both lines (IIMb and beta) do not differ in their geometry and present the same behavior in the osmotic response.

24.

BF6 - PREVENTIVE ACTION OF QUERCETINE ON THE ERYTHROCYTE DEFORMABILITY MODIFICATED BY ARSENIC (V)

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We were previously observed that previous quercetine (Qc) treatment inhibits the effect of arsenic (Asv) on the membrane, protecting the cell against oxidative stress and the red blood cells (RC) maintained the cell shape and response osmotic. Now, we study if the Qc prevents the increment of the erythrocyte deformability (ED) caused for Asv. Washed human RC were incubated as follow: I) in PBS, pH: 7.4 10' (control); II) in 3μM Qc solution, 10' (Qc); III) in 3μM Qc solution, 10' and later in AsV solution (0.32 μM Na₂HAsO₄·7H₂O), 30'(Qc-Asv); IV) in Asv solution, 30' (Asv). In they were determined rigidity index: (RI) (high RI low ED) by filtration through pores of 5μm. Statistic: ANOVA test; values presented as means ± SEM, p< 0.05 was accepted. Controls: 8.30±0.55a,b (n:24); Qc: 7.97±0.59a,b (n:11); Qc-Asv: 8.52±0.50a,b (n:8); Asv: 10.79±0.75b(n:9), (a: ns ; b: p<0.05). The cell shape is one of the factors determining from ED, the results from this study are in concordance with the early. Therefore, we conclude that the incorporation of Qc in the lipidic bilayer avoid hemilayer imbalance preserving the ED and, in consequence, its life span.

25.

BF7 - EFFECTS OF GABAERGIC PHENOLS ON FLUIDITY AND LIPID PEROXIDATION OF MEMBRANE MODELS

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The GABA-A receptor is the main inhibitory receptor in the brain. Several modulatory compounds are potentially useful in human medicine as the widely known phenol propofol. Recently, our group has also described gabaergic activity for other similar compounds such as thymol, eugenol, carvacrol and chlorothymol. Considering that all those compounds are highly lipophilic, the aim of the present work is to study their effects on the lipid bilayers properties as well as their antioxidant abilities. Hence, it was analyzed their effects on membrane fluidity and on the lipid peroxidation in artificial bilayers. The results indicated that: *i-* all compounds were able to increment the bilayer fluidity in a concentration dependent manner, *ii-* thymol and carvacrol demonstrated lesser protection that propofol and chlorothymol against the lipidic peroxidation, while eugenol showed intermediate or high protection values considering the dienes or trienes production respectively. In conclusion, the demonstrated ability of all assayed compounds to interact with membranes as well as the observed antioxidant activity, confirm the importance to consider not only the non-specific modulation that those compounds could exert on specific receptors, but also their role in the main mechanisms implicated in the protection, at neuronal level, against the deprivation of oxygen and nutrients.

26.

BF8 - PHYSICAL PROPERTIES OF SPIDER WEBS: AN ALTERNATIVE METHODOLOGICAL PROPOSAL

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The spider webs are known for their remarkable physical and chemical properties. Numerous studies around the world aim to investigate and elucidate the potential applications of this material. The aim of this work was to develop a methodology that could be carried out in laboratories of modest equipment that in turn produces similar results to those obtained in high-tech laboratories. We examined the biomechanical properties of silk produced by *Uloborus plumipes* using a vernier digital caliper, confetti and precision scales. Strength values (1713 ± 481 MPa), Strain rate (0.22 ± 0.09) and Stiffness (16.3 GPa) show that the design developed and tested in 32 orb-webs, gives similar values to those observed by several researchers. This is the first data of physical parameters on *U. plumipes* webs.

27.

BF12 - PHENOMENOLOGY OF Vero AND HeLa CELLS LOCOMOTION IN COLONY OF DIFFERENT GEOMETRIES

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The study of cell motility paid considerable attention in recent years because it is involved in complex cellular processes implicated in a great number of living systems. The locomotion of Vero and HeLa cells on polystyrene petri dishes were characterized by statistical techniques employing a time-lapse system to capture digital images of either initial quasi-radial and quasi-linear colonies. They were cultured in a chamber at 37°C, maintaining the pH with carbon dioxide independent medium. The trajectories, orthogonal and parallel velocity, the mean square displacement (msd) were determined at each time for a large number of cells in colonies with different cell populations (n). The module of the average cell velocity in the border region increased with n for quasi circular colonies, approaching that of the quasi-linear ones and large n, and was higher for Vero than HeLa cells. For both cells, the analysis of msd data rendered power law dependence with an exponent greater than one. Data suggested cooperative effects and displacement in heterogeneous medium. This movement appears to be mainly related to those interactions that affect the generation of protrusion forces in the colony, a complex process in which chemical signaling results in the production of mechanical energy. These data is relevant for modeling biological systems.

28.

BF14 - CARDIAC SODIUM CHANNEL CURRENT AND PROTEIN EXPRESSION IN CHRONIC HYPOXIA

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Hypoxia increases cardiac action potential duration and may predispose to lethal arrhythmias. Low oxygen pressure (hypobaric hypoxia) augments potential-dependent Na currents (INa) and this effect may be clinically relevant as an arrhythmia-triggering mechanism. This work aimed at obtaining a mathematical function for the relationship between INa and membrane potential based on experimental results and to assess changes in protein expression caused by hypoxia. Rats were exposed to either normal pressure or chronic hypobaric hypoxia. Isolated myocytes were obtained and INa was measured in them under potential clamp conditions before and after applying tetrodotoxin. Tetrodotoxin-sensitive INa was significantly higher in cells from hypoxic rats. With a function linearization method, the mathematical function was $y = 598 \cdot x \cdot 10^{0.01x}$ for controls and $y = 1257 \cdot x \cdot 10^{0.01x}$ for chronic hypoxia. These theoretical functions obtained from experimental results allow calculating INa for a given transmembrane potential. Protein expression analysis was carried out with 12 % polyacrylamide gel electrophoresis in homogenates obtained from myocytes of both groups. It was found that chronic hypoxia caused a dramatic change in protein expression. The different values calculated for the constant term of the mathematical function for the relationship between potential difference and INa in each group are related to the INa increase induced by chronic hypoxia, which in turn may be caused by changes in protein expression, although further work is needed to adequately characterize those changes.

29.

BF15 - DETERMINATION OF THE ENZYMATIC ACTIVITY OF THE SALIVARY ALPHA AMYLASE

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The enzyme alpha-amylase which is in the saliva starts the breakdown of starch, the main source of carbohydrates in the human diet. When the starch is treated with an iodine solution gives a complex of blue colour. Thus, the action of alpha-amylase on polysaccharides may be followed by measuring the disappearing of blue coloration of the complex. The aim of this paper is the determination of the enzymatic activity of salivary alpha-amylase in healthy adult smokers. Saliva samples were collected in a sterile disposable polypropylene tube of 15 ml. The hydrolysis of starch catalyzed by alpha amylase was followed in the presence of a solution of iodine/ iodide by measuring the absorbance at 540 nm versus time. This measurement is based on the fact that iodine is placed inside the helix that amylose forms in the hydrophobic regions, giving a blue coloration. The presence of alpha amylase degrades amylose, producing the disintegration of the helix and as a consequence the disappearing of blue coloration. The curves were fitted with an exponential decay. From the fitting curves, the time constant, that is to say, the time it takes the enzyme to degrade 63% of the starch in the sample was obtained. The time constant was: 14.5, 9.7, 8.8; 7.5 minutes for men and 13.5; 6.3, 5.58, 5.74 minutes for women, while for non-smokers was 4 min. From the results we can conclude that there is an increase in the time of degradation of starch, i.e. lower enzyme activity, in the case of saliva of smokers compared with nonsmokers, which could negatively affect the digestion thereof.

30.

BM4 - DEHYDROLEUCODINE ALTERS THE LAMELLIPODIA FORMATION AND CAUSES ERRATIC MIGRATION OF HeLa CELLS

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Metastasis is the main cause of death in patients with cancer, then, it is relevant to find drugs to attack the cell invasion. Previously, we have demonstrated that Dehydroleucodine (DhL) has an inhibitory effect in the proliferation of HeLa cells. In this work we studied the effect of DhL on the migration of HeLa cells. Wound healing assay was performed in HeLa cells treated with 0 and 20µM DhL. We measured the speed and direction of migration and analyzed the cytoskeleton by immunofluorescence. DhL decreased the migration velocity (40%) and changed the dynamics of the advancing front of the cells. While control cells generated large and wide lamellipodia and migrated perpendicularly to the wound, DhL treated cells generated small lamellipodia and migrated erratically. Also, DhL treatment changed the organization of the cytoskeleton. In control cells the microtubules were concentrated in the central region and actin filaments formed an extensive network in the lamella. In DhL treated cells, the microtubules were uniformly distributed in the cytoplasm and the actin filaments were packed in a fine line in the cell periphery. These results indicate that DhL affects the migration and the cytoskeleton of HeLa cells. DhL would be a potential agent to stop the invasion of tumor cells in the body.

31.

BM6 - RIBOFLAVINS INDUCE DIFFERENT PATTERNS OF CELL DEATH IN SQUAMOUS CARCINOMA CELLS

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Photodynamic Therapy (PDT) is a treatment based on cell death induced by light activation of a Photosensitizer (P) localized in tumor cells. Riboflavin (RF) is an efficient P and could be applied for PDT. Our aim was to evaluate the effect of light-activated RF and RF Ester (RFE) on the human squamous carcinoma cell line SCC13. Cells were cultured in DMEM, incubated with RFL or RFH (50, 100µM) and irradiated with 24-high power LED lamp of 444nm. After that, cells were left in darkness for 6 or 24h. Cell viability was determined by neutral red assay and cell proliferation by Ki-67 immunocytochemistry. Apoptosis was evaluated *in situ* by uptake of Hoechst33342. Morphological studies were performed by high resolution optical microscopy and electron microscopy. Statistics: ANOVA-Tukey. After irradiation, both types of RF induced cell death, with decreased proliferation rate ($p < 0.05$). Treated cells showed apoptotic nuclei by nuclear fluorescence staining. Moreover, some cells exhibited chromatin condensation and several apoptotic bodies; while other cells presented typical ultrastructural morphology of necrosis. We also observed a third type of cell response characterized by cytoplasmic vacuolization and surface blebs without chromatin condensation. These findings were more notorious at 24h after light irradiation. These results demonstrate that both FS could be effective in the application of PDT in superficial skin tumors.

32.

BM7 - POLYMORPHISMS OF PAIN-RELATED GENES IN THE POPULATION OF CORRIENTES CITY

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COMT, OPRM1 and PDYN are human genes involved in pain mechanisms. Their polymorphisms have been associated to different nociceptive phenotypes, contributing to inter-individual differences in sensitivity to painful stimuli. Many of these variations have significantly different population frequencies, adding an anthropological interest. Argentinean population mainly has European and Amerindian ancestry, and the impact of these genetic components on the behavior of pain-related markers is unknown. Our aim was to characterize the genetic frequencies of 4 polymorphisms: 1 PDYN VNTR (*rs35286281*), 2 OPRM1 SNPs (*rs17174794*, *rs2075572*) and 1 COMT SNP (*rs4633*) in an urban population sample of Corrientes City (Corrientes Province, Argentina). We analyzed 100 DNA samples from hospitals by PCR, PCR-RFLP, and electrophoresis. We compared our results to available data from Europeans (Arlequin v3.5). The 4 markers fit Hardy Weinberg Equilibrium (Exact Test, $\alpha = 0.05$; $p > 0.05$) and the most frequent alleles were the ancestral ones. Only *rs2075572* differentiated our population from the rest (Exact Test, $\alpha = 0.05$; $p < 0.05$). This differentiation is probably caused by an Amerindian genetic component introduced during the population admixture process. The study of coding variability is important not only for genotype-phenotype associations, but also for understanding the population admixture process.

33.

BM10 - NOVEL METHODOLOGY FOR DETECTION OF *Botrytis cinerea* FROM APPLE SYMPTOM

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Gray mold caused by *Botrytis cinerea*, one of the major diseases of apple fruits in post-harvest. Their detection in disease early stages and symptomless, is essential to establish control strategies. This study developed a methodology for detection of *B. cinerea* from apple symptomless, through a protocol of extraction DNA and amplification for PCR the IGS ribosomal spacer and transposable elements (*flipper* and *boty*). Red Delicious apples were wounded with a punch, and inoculated with 20 µl of a conidial suspension (10⁶ conidia/mL) of *B. cinerea*; after 3 days of inoculation and without symptoms of disease, the fruits were processed in a mortar in liquid nitrogen to homogenize the samples. Pure cultures of *B. cinerea* (3 days old) were used as positive control and healthy apples as negative control. The extraction method was easy and allowed obtain genomic DNA of *B. cinerea* from samples of apple symptomless, which is of sufficient quantity and quality for amplify by PCR the regions of interest. PCR amplification indicates the presence of both transposable elements and IGS spacer. Fungus genomic DNA was molecularly characterized in subpopulation transposon type. Pure cultures are also observed transposable elements and IGS spacer, while in healthy apples were not detected. In conclusion, the proposed methodology was able to detect the presence of *B. cinerea* in apple symptomless (gray mold) for its possible application in the preventive control of latent diseases.

34.

BM11 - EFFECT OF QUERCETIN AND CATECHIN ON MAPK SIGNALING AND COUPLING OF THE NADPH OXIDASE COMPLEX

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Previous results from our laboratory showed that the antioxidants quercetin (Q) and catechin (C), present in red wine, reduce angiotensin II (AII)-induce vascular smooth muscle cells (VSMCs) proliferation and migration by inhibiting NADPH oxidase, the main reactive oxygen species generator in the vascular wall. We investigate whether the MAPK signaling pathways activated by AII, Erk1-2 and p38MAPK, are modified by the action of antioxidants. In addition we evaluated if the effect of Q and C is related to the translocation of the protein p47 from the cytosol to the membrane, required for activation of the complex NADPH oxidase. Western blots were performed on extracts of VSMCs derived from spontaneously hypertensive rat. Treatment with AII (10⁻⁷M) increased the phosphorylation of both Erk1-2 and p38MAPK. Q, C or their combination had no effect on the activation of Erk1-2. The coinubation of Q (15µM) + C (20µM) significantly inhibited the phosphorylation of p38MAPK and p47 translocation to the cell membrane induced by AII. These results demonstrate that small concentrations of Q and C have a synergistic effect on the inhibition of redox-sensitive signaling pathways. The effect of Q and C is associated with uncoupling of NADPH oxidase, proving to be capable of preventing and/or reverse the vascular changes that occur in hypertension.

35.

BM13 - RATS EXPOSED TO Cd²⁺ IN DRINKING WATER. INJURY PRODUCED IN HEART, A HISTOLOGICAL ASSESSMENT

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Cd has been associated with an increased incidence in stroke and heart failure. The aim of the present work was to assess histological alterations in the rat heart, after Cd exposure. We also evaluated the putative protection of a soy-based diet. Male Wistar rats, 8-10 weeks old, were separated into 6 groups and treated during 2 months as it follows: groups (1), (3) and (5) were fed a casein-based diet; groups (2), (4) and (6) were fed a soy-based diet. Groups (1) and (2) received tap water; (3) and (4) received tap water+Cd²⁺ 15ppm; (5) and (6) received tap water+Cd²⁺ 100ppm. Histological studies were performed using hematoxylin-eosin and Sirius red staining. Apoptosis was assessed by the TUNNEL test. We found that Cd²⁺ exposure causes tissue alterations, with hypertrophy and fusion of muscular cells. We also observed extracellular matrix alterations in the animals exposed Cd. A soy-based diet couldn't prevent the disturbances found in the rat heart. On the other hand, we didn't observed any increase in apoptotic cells in the heart of poisoned rats. We conclude that exposure to Cd-contaminated drinking water alters significantly the cardiac tissue. Probably, this observation would explain the association between Cd exposure and cardiovascular diseases. No protection from a soy-based diet was found.

36.

BM15 - ADULTICIDAL ACTIVITIES FOR CONTACT OF *Aloysia citriodora* (VERBENACEAE) AGAINST *Pediculus humanus capitis* (ANOPLURA, PEDICULIDAE)

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The objective of this study was to determine the contact toxicity of essential oil (EO) isolated by hydrodistillation from *A. citriodora* against human head lice. A Filter-paper contact toxicity bioassay was used and performed in the dark at 31 ± 1°C and 65 ± 5% RH. Batches of 10 adults were exposed to four concentrations (0.042, 0.126, 0.294 and 0.630 mg/cm²), each of which was dissolved in hexane. Lice were observed for evidence of knockdown every 5 minutes for 1 hour. The criterion for knockdown was when an insect remain on its back with no leg movements. The KT₅₀ was calculated using the program MicroProbit 3.0. Data analysis showed that the KT₅₀ value decreased with increasing concentrations of EO. At the highest concentration, the KT₅₀ (11.37 minutes) was significantly lower than the others KT₅₀ values (P < 0.05). These results indicate that EO from *A. citriodora* is effective and could be used as potential *P. humanus capitis* control agent.

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37.

BM16 - EFFECTS OF HYPERTHERMIA ON THE GENE EXPRESSION OF HEAT SHOCK PROTEINS IN hMLH1-DEFICIENT TUMOR CELLS

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Exposure of cells to a transient elevation of temperature induces the expression of heat shock proteins (HSPs). Several tumor cell lines constitutively express HSP27 and HSP72, which are critical for cell proliferation and survival, they have been also associated with DNA repair and drug resistance. However, the relation between HSPs and Mismatch Repair (MMR) has not been established. The aim of the study was to analyze the mRNA expression levels of *HSP27*, *HSP72*, *HSF1*, *hMLH1* and *hMSH2* in colon cancer MMR-proficient (HCT116+ch3) and deficient cell lines (HCT116 and HCT116+ch2). Cells were exposed to a mild hyperthermia (41 and 42°C, 1 hour) and then collected at different times. mRNA levels were determined by RT-qPCR. Constitutive expression of *HSP27* and *HSP72* was higher in HCT116+ch3 cells. After hyperthermia, mRNA levels of *HSP27*, *HSP72*, *HSF1*, *hMLH1* and *hMSH2* significantly increased in all cell lines, but they were higher in HCT116+ch3 cells. Our results suggest that the MMR system may participate in the heat shock response and HSF1 could modulate its activity.

38.

BM17 - EFFECT OF A SPECIFIC INHIBITOR OF AKT IN MACROPHAGES INFECTED WITH *Brucella abortus*

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Intracellular pathogens remain viable in the cells and even multiply. In phagocytosis, several factors are known that could be targeted by the alterations caused by these parasites. The subversion of signaling pathways and membrane traffic is a tool used by intracellular pathogens to avoid degradation in phagolysosomes. *Brucella* invades macrophages and replicates inside phagosomes that interact with early components of the endocytic pathway. Rab GTPases are key host proteins that regulate vesicular trafficking and phagosome maturation. AKT, a Ser/Thr kinase of the host cell, phosphorylates AS160, a GAP (GTPase Activating Protein) of certain Rabs, like Rab11 and Rab14. The phosphorylation of AS160 results in the inhibition of its GAP activity, leaving these Rabs in their active state bound to GTP. We analyzed the effect of a specific AKT inhibitor in macrophages infected with a virulent strain (2308) and two vaccine strains (S19 and RB51) of *Brucella abortus*. We assessed the effect of AKT inhibition in the intracellular destiny of all three strains by transmission electron microscopy. The treatment of infected cells with the inhibitor of AKT resulted in a significant decrease in the bacterial progeny determined by quantification of colony forming units. Our results suggest that *Brucella abortus* uses AKT pathway to activate Rabs involved in the generation of a safe intracellular niche where survive and replicate.

39.

BM19 - NAD(P)H-OXIDASE COMPLEX REGULATION BY HSP70 IN VASCULAR SMOOTH MUSCLE CELLS. LOSARTAN EFFECT

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Involvement of Hsp70 in the regulation of Nox4 and p22 phox NAD(P)H-oxidase subunits by the Rho small GTPases, Cdc42 and Rac signaling pathway after Losartan administration in CMLVs. SHR and WKY VSMC primary cultures from resistance vessels were used. Groups Control (C), Angiotensina II (AII: 100 µmol/L), Losartan (L: 100 µmol/L) y Losartan plus Angiotensina II (L+AII). VSMCs were stimulated with AII for 15, 60, 120, 180 min, 24 and 48 h in the presence or absence of Losartan. Cells remained exposed to Losartan for 45, 90, 150, 210 min or 24 and 48 h. Hsp70 membrane translocation was shown in WKY VSMCs 15 min after AII stimulation, decreasing its expression after Losartan exposure for 60 min. In SHR, membrane translocation of Hsp70 occurred after Losartan exposure for 90 min associated with increased Cav-1 expression. SHR Losartan treatment for 90 min showed co-immunoprecipitation of Nox4 with Hsp70, as well as interaction with decreased p22, Cdc42 and Rho in the membrane fraction compared to AII stimulated VSMCs. Moreover, Hsp70 and p22 colocalization by immunofluorescence was shown. Early activation of MAPK ERK1/2 and p38 signaling pathway was demonstrated in AII 15 min cell stimulation, the opposite effect by Losartan treatment for 45 min. Translocated Hsp70 is involved in the negative regulation of Nox4/p22NAD(P)H-oxidase and in RhoA and Cdc42 absence of activation, suggesting Hsp70 involvement in cytoskeleton stabilization in Losartan VSMCs SHR.

40.

BM23 - SEED PROTEIN ANTIGENIC PROFILES OF CACTOIDEAE AND OPUNTIOIDEAE (CACTACEAE) SPECIES

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The aim of this study was to analyse antigenic patterns of species of Cactaceae to assess their usefulness for taxonomic identification and to clarify the evolutionary relationships within the family. Seeds protein extracts corresponding to 4 species of Cactoideae and 6 species of Opuntioideae were compared. Antigenic profiles were obtained by western blot using the antisera prepared against *Cereus aethiops* and *Opuntia elata* var. *cardiosperma* extracts. Similarity (Jaccard's Index, S_j) and distance (Bray-Curtis Index, D_{BC}) matrices were analysed through clustering method (UPGMA). The patterns obtained using anti-*C. aethiops* presented a total of 40 bands (12.5-113 kDa). Between both subfamilies S_j were 0.412-0.697 and D_{BC} were 0.172-0.387. Between species of *Cereus* S_j was 0.971 and D_{BC} was 0.091. The patterns obtained using anti-*O. elata* var. *cardiosperma* presented a total of 44 bands (13.5-112 kDa). Between both subfamilies S_j were 0.514-0.738 and D_{BC} were 0.128-0.287. Among species of *Opuntia* S_j were 0.744-0.946 and D_{BC} were 0.082-0.168. Species evolutionarily closest to *C. aethiops* and *O. elata* var. *cardiosperma* evidenced higher affinity than the others species. This correlation demonstrates the usefulness of antigenic profiles as complementary method for taxonomic identification and for the interpretation of relationships among Cactaceae.

41.

BM24 - DEHYDROLEUCODINE DELAYS HeLa CELLS PROLIFERATION IN G1 OF THE NEW CELL CYCLE

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Tumors are characterized by an abnormal and uncontrolled cell proliferation. Many efforts are focused to find agents that block the uncontrolled cellular proliferation. Dehydroleucodine (DhL) is a sesquiterpene lactone isolated from *Artemisia douglasiana B*, a plant that grow up in the west Argentine region. In this work we analyzed the effect of DhL in the proliferation events of HeLa cells. Cells synchronized in G1 / S by double thymidine treatment, were stimulated with 10% fetal bovine serum in presence of 0-30 μM DhL for 24 -72 h. We determined the growth rate (GR \pm SEM) every 24 h and analyzed the cell distribution in G1, S and G2/M by flow cytometry analyses. At 72 h of culture, cells treated only with the vehicle (0 μM DhL) showed a GR of 5.6 ± 0.3 , with 5 μM DhL 3.8 ± 0.3 , with 10 μM DhL 2.7 ± 0.4 , with 20 μM 1.7 ± 0.07 , and with 30 μM 0.7 ± 0.05 . The flow cytometry analyses indicated that 5 μM DhL arrested the cells in G2 / M for 8 h. However 20 μM DhL arrested the cells in S phase for 12 h, in G2 / M for 4 h and permanently in G1 of the new cycle. These results indicate that DhL inhibits the proliferation of HeLa cells in a dose-dependent way arresting the cells transiently in S and G2/M and permanently in G1 of the new cycle.

42.

BM26 - VITAMIN A DEFICIENCY ON ENERGETIC METABOLISM OF THE HEART RAT

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We had reported that vitamin A deficiency alters the lipid content and induces lipoperoxidation in the heart. Now, we study the impact of this nutritional deficiency on heart energetic metabolism. For that, activities and mRNA levels of enzymes, and expression of nuclear receptors involved in heart lipid metabolism were determined. Wistar male rats at 21 d age were weaned onto either a vit A deficient diet (-A) or the same diet with 8 mg retinol/kg diet (control, +A). They were fed for 3 months. Also, a -A group was refeed by 15 days with control diet (-A refeed) before sacrifice. Heart left ventricle was used. In -A group the heart content and synthesis of cholesterol and phospholipids were increased. In addition, alterations in mitochondrial proportion of phospholipids and composition of their fatty acids (FA) were observed. The increased activity and mRNA expression of carnitine palmitoyl transferase-I and decreased activity of acetyl-CoA carboxylase suggest an induction of the mitochondrial FA beta oxidation. Also, a decrease in the mRNA levels of retinoid receptors RAR α , RXR α and RXR β , and an increase of PPAR α and PPAR β , which regulate many gene involved in FA oxidation, contribute to explain the effects of vitamina A deficiency on the heart lipids. The vitamin A refeeding of -A rats considerably improves the observed changes. Vitamin A modulates the heart lipid homeostasis and, in particular, the energetic metabolism through mitochondrial FA β -oxidation.

43.

BM27 - TRYPANOCIDAL EFFECT OF QUINONES COUPLED TO STEROIDS AND TERPENOIDS AND THEIR DERIVATIVES OBTAINED FROM PLANTS

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Trypanosoma cruzi is the causative agent of Chagas disease. When cultured, this parasite cycles between epimastigote form and a low percentage is transformed into the infective form trypomastigote. We have found that quinones coupled to steroid and terpene residue, obtained from native plants, such as secochilolilquinone (PQ) and, to a lesser extent, secochilolilhidroquinone (PHQ) affect the growth of epimastigotes at low concentrations. To identify the active groups of these compounds, in the present study we evaluated the effect of different derivatives on *T. cruzi*. Epimastigotes (Dm28C strain) were cultured in Diamond liquid media, in the presence or in the absence of different concentrations (1-10 $\mu\text{g/ml}$) of the compounds. Among the compounds tested, PPBuC, PPU, PPAZ and, to a lesser extent PPQ, PPHQ and PPDl showed antiproliferative effects on parasites. As a continuation of this work, it will be determined the minimum lethal dose for each compound and try to elucidate the molecular targets, and assess the toxicity to mammalian cells.

44.

BM30 - LOSARTAN ON Hsp70/CHIP AND Nox4 EXPRESSION IN PRIMARY CULTURE PROXIMAL RENAL TUBULE EPITHELIAL CELLS FROM SPONTANEOUSLY HYPERTENSIVE RATS (SHR)

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Objective: Here we examined the Angiotensin II AT1 receptor inhibitor Losartan effect on Hsp70, Nox 4 and CHIP protein expression in SHR primary culture proximal tubule epithelial cells (PTCs) Four Groups Control (C), Angiotensina II (AII: 1×10^{-7} y 1×10^{-11} M), Losartan (L: 1×10^{-5} M, and Losartan plus Angiotensin II (L+AII). PTCs were stimulated with AII for 15 and 60 min, in the presence or absence of Losartan. PTCs were exposed to the inhibitor for 90 min Increased expression of AT1 in SHR vs WKY, $p < 0.001$ was shown. After Losartan administration (1×10^{-5} M, 90 min) we observed increased Cav-1 expression in SHR+Los vs SHR $p < 0.001$, associated with higher levels of Hsp-70 protein expression in membrane fraction from SHR+Los vs SHR, $p < 0.001$ Decreased Hsp-70 in SHR+Los vs SHR cytosolic fraction allowed us to confirm Hsp70 translocation to membrane fraction. We have demonstrated decreased Nox4 expression in membrane fraction from SHR+Los vs SHR, 0.24 ± 0.09 vs 1.00 ± 0.05 , $p < 0.001$ $n=3$ and also lower Nox4 protein levels in SHR Los+AII (Los 1×10^{-5} M 90min, AII: 1×10^{-7} M 60min) vs SHR+AII (1×10^{-7} : 60min) $p < 0.001$. Interaction among Nox4, Hsp70 and CHIP was determined by immunoprecipitation and immuno-fluorescence Conclusion: After Losartan administration translocation of Hsp70 to PTC membranes in SHR might exert a cytoprotective effect by down-regulation of Nox4 associated with Hsp70/CHIP upregulation.

45.

BM31 - ANALYSIS OF TELOMERIC SEQUENCES IN THE PROGENY OF BLEOMYCIN-EXPOSED CELLS*Paviolo NS, Vidal Bravo M, Bolzán AD.**Laboratorio de Citogenética y Mutagenesis, IMBICE, C.C. 403 (1900) La Plata, Argentina. E-mail: abolzan@imbice.org.ar*

The chromosome damage and its relationship with the telomeric sequences was analyzed in the progeny of mammalian cells exposed to the antitumoral antibiotic bleomycin (BLM). The aim of the present work was to evaluate if chromosome damage induced at telomeres and interstitial telomeric sequences (ITS) persisted on the progeny of the cell population originally exposed to the mutagen. Fluorescence *in situ* hybridization (FISH) with a PNA telomeric probe was applied on chromosome preparations of Chinese Hamster Ovary cells (CHO cell line) and rat cells derived from adipose tissue (ADIPO-P2 cell line). Cells were exposed in the log phase of growth to 2.5 µg/ml of BLM during 30 minutes at 37°C. Cytogenetic analysis revealed a significant increase in the frequency of chromosomal aberrations after 18 h after treatment in CHO cells treated with BLM compared with unexposed cells. BLM also induced a significant increase in the frequency of chromosomal aberrations 6-10 days after treatment on both cell lines. Our results also indicated that BLM induced delayed chromosomal instability, revealed by amplification of ITS in CHO cells and the presence of incomplete chromosomes in ADIPO-P2 cells.

46.

BM32 - COMPARISON OF SEQUENTIAL ISOLATION TECHNIQUE BETWEEN BOVINE AND PORCINE ENTEROCYTES*Molinero D, Busso L, Bellingeri R, Alustiza F, Picco N, Grosso M, Motta C, Vivas A.**FAV, UNRC. Río Cuarto, Córdoba, Argentina.**E-mail: danimoli86@hotmail.com, lubusso85@gmail.com*

Primary cultures are the best model to study the tissue from which they derive. The objective was to compare the Sequential Isolation Technique (TAS) to obtain primary cultured porcine and bovine enterocytes. These were obtained from segments of 20 cm. of intestine (jejunum-ileum) from pigs and cattle for slaughter. The samples were transported in cold PBS-antibiotic to the laboratory, we proceeded with the TAS along the villus-crypt axis for bovine tissue (TB) at 30 min. and the porcine tissue (TP) at 3 hs. The cells obtained were incubated in Williams E medium, at 37°C and 5% CO₂, and were observed in inverted microscope the development and formation of monolayer cell. We obtained 1.3 x 10⁷ cells/ml for TB and 1.2 x 10⁷ cells/ml for TP. Low contamination with intraepithelial lymphocytes, absence of fibroblasts and a low level of differentiation were observed. Viability as assessed by trypan blue, reached 98% and 95% of TB and TP respectively. With this technique is able to obtain a primary culture of pig and cattle enterocytes with excellent results in high viability and low contamination, where TB had better results in the parameters used.

47.

BM33 - STRUCTURAL STUDY OF *Trypanosoma cruzi* NDPK1 AND ADK1 PROTEINS AS A STEP FOR RATIONAL DRUG DESIGN*Gómez Barroso JA, Milesi S, Pereira C, Aguilar CF.**Lab de Biol Molec Estructural, UNSL, IMIBIO SL - CONICET.**E-mail: jagomez@unsl.edu.ar*

Trypanosoma cruzi is the etiologic agent of Chagas' disease. The objective of our work is the resolution of the three-dimensional structure, by X-ray crystallography, of energetic metabolism involved proteins. The three-dimensional structure resolution of these proteins is an important step for rational drug design based on the structure. In this work we studied two proteins: TcAdK1 (adenylate kinase 1) and TcNDPK1 (nucleoside diphosphate kinase 1). The proteins were overexpressed in *E. coli* as N-terminal poly-histag fusion protein and purified by chromatography. TcAdK1 crystallization assays were realized using different screening methods. TcNDPK1 crystallographic data was processed with CCP4 programs. A three-dimensional TcNDPK1 model was obtained by molecular replacement after several refinement steps. TcAdK1 was overexpressed and purified by affinity and molecular exclusion chromatographic steps. Some TcAdK1 crystals were obtained on two different conditions after screening using 5, 10 and 15 mg/ml of protein concentration values.

48.

BM34 - ULTRASTRUCTURE IN ORAL LEUKOPLAKIAS CLINICALLY CLASSIFIED AS HOMOGENEOUS. CONTRIBUTION TO THE DIAGNOSIS*Micinquevich S, Mayocchi K, Dorati P, Gomez MA.**Asignatura Patología y Clínica Estomatológica FOLP UNLP.**E-mail: susmic2003@yahoo.com.ar*

The oral leukoplakias type homogeneous have been associated with a reduced potential for transformation. Objective: To explore if the ultrastructural analysis may reveal changes early as indicators for the possibility of transformation. We took 5 samples of waxy material file of the Laboratory of Surgical Pathology (FOLP) corresponding to oral leukoplakias clinically diagnosed as homogeneous. There were new cuts shaping two groups: (a) cuts to be processed with routine technical., by embedding them in paraffin and coloring them with hematoxylin and eosin. (b) cuts to be processed for TEM. Results: The MO preparations showed thickening of the keratin and increase the thickness epithelial (acanthosis). In a single case was observed mild dysplasia. Were interruptions, and projections of epithelial cells, evident spongiosis. At TEM the changes were evident in the basal layer. There were few hemidesmosomes defined in some areas, irregular shape of keratinocytes and electrodensity variable. Elongated nuclei with crenaciones peripheral and granules of heterochromatin. It can be concluded that it showed characteristics of cells with high proliferative activity, similar to those observed in dysplasias. These changes could be indicators at the ultrastructural level of the malignant potential of these white lesions.

49.

BM35 - ORAL PATHOLOGIES. MARKER C-MYC BY MOLECULAR TECHNIQUE

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Objective: To evaluate on the basis of scientific evidence the expression of the oncogene c-myc with molecular technique. Correlate the expression with viral infection by HPV took into account waxed material of the Laboratory of Surgical Pathology with the following division: (a) proliferative lesions (LP), condylomata n= 15); b) carcinoma squamous cell (CAR) (n= 15) and (c) controls (C) (n= 10). Total 30 preparations, all with histopathological diagnosis. The technique was used PCR Nested Protocol for viral detection and SSCP for the genotyping of the virus. For the oncogene c-myc technique was used in this RG-PCR based on the amplification of competitive sequences of c-myc and the reference gene b-globin, reading by electrophoretic run in minigeles of poliacrilamida to 6 per cent and stained with silver nitrate. Results: of the LP were amplified 5/15 (33%); CAR buccal 2/15 (13%); and C 2/10 (20%). The viral distribution type-specific to the group a) was: 4+ for the type 6 and 1+ to 16. For group b): a positive for HPV 6, 3+ for the type 11, 5+ for the genotype 16 and a coinfection 16/18. In cervical mucosa have been detected differences of expression c-myc between samples with or without HPV infection in our series the samples corresponding to LP were positive for HPV 33%; for CAR 66% and in C 2%. In this item the result is consistent with findings in cervix. Expression of c-myc was lower than in undifferentiated cells, coinciding with high-risk HPVS. You would think that the amplification, at least in our series, would take place in proliferative lesions non-invasive.

50.

BM36 - PURIFICATION OF *T. Cruzi* BDF2 AND N-TERMINAL FIP1 PROTEINS FOR X-RAY ANALYSIS AND STRUCTURAL STUDIES

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Chagas' disease is a chronic systemic parasitic infection caused by the protozoan parasite *Trypanosoma cruzi*. Available drugs are highly toxic and often ineffective, particularly those used to treat the chronic stage of the disease. The aim of our work is the structural and functional study of proteins which are vital for *Trypanosoma cruzi* as a first step in rational drug design. TcFIP1-Nterm (Factor interacting with Pap) is involved in the maturation of mRNA and is essential for cell viability. TcBDF2 is a nuclear bromodomain protein involved in histone acetylation and could be taking part in a chromatin remodelling complex in *T. cruzi*. Both proteins were overexpressed as fusion proteins with His-tag N-terminal. They were purified by using sequentially: affinity, anion exchange and then molecular exclusion (FPLC). Each stage was analyzed by SDS-PAGE and Western Blot. The results show a high concentration of protein with a high degree of purity which allows structural studies as the next step.

51.

BM37 - THE INTERACTION OF RIBOSOME INACTIVATING PROTEINS WITH P PROTEIN IS A CASE OF CONVERGENT EVOLUTION

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Ribosome inactivating proteins (RIPs) are N-glycosidases that depurinate a specific adenine residue in the conserved sarcin/ricin loop in subunit 28S of rRNA. This proteins has different specificity against eukaryotic and prokaryotic ribosome, and all of this can depurinate nude rRNA (without ribosomal protein), suggesting that different RIPs would interact with different proteins. The interaction with P protein is one of the most studied; some RIPs have been reported to interact with the C-terminal end of this, including TCS, Shiga like 1/2 (SLK 1/2), Maize RIP and ricin and the interaction sites of two of them (TCS and Maize RIP) have been mapped. In this work, we use molecular phylogeny to support the hypothesis that different RIPs have achieved the ability to interact with P protein independently throughout evolution, being a case of convergent evolution. We design tree with maximum likelihood and Bayesian method to explain this hypothesis and design an alignment where we can observe that interaction motives are different and it are in different position along the sequences. Moreover after affinity testing we propose pulchellin like a new RIPs that has the ability to interact with P protein. For this reason recombinant pulchellin was expressed and purified and SLK 2 was cloned too, to try to map possible sites interaction of these toxins with P protein.

52.

BM 38 - PENIAL GLANDS OF THE PATAGONIAN RED SNAIL *Odontocymbiola magellanica* (NEOGASTROPODA, VOLUTIDAE)

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Phylogenetic reconstructions of the Caenogastropoda have paid particular attention to the external morphology of copulatory organs. However, the internal structures (spermiduct, epithelial cover and glands, irrigation and innervations, etc.) are largely unknown, and may be also significant for phylogenetic as well for functional studies. We have recently examined the copulatory organs of two caenogastropod species, *Trophon geversianus* (Neogastropoda, Muricidae) and *Pomacea canaliculata* (Architaenioglossa, Ampullariidae) and have found marked differences in the structural plans of these taxa, which are considerably apart in a phylogenetical perspective. In the current study, *O. magellanica* (Neogastropoda, Volutidae) showed a structural plan similar to that of *T. geversianus*, in that the penis is an outgrowth of the right side of the neck, which bends rearwards and culminates in a small but distinct genital papilla. It showed, however, two distinct superficial glands, named the 'distal' and 'paradistal' glands, extending the first through the distal region of the dorsal aspect of the penis, while the second extends close to the internal border of this organ, separated from the distal gland by the sulcus from which the spermiduct was originated. It should be noted that these are ductless glands similar to those found in the penial sheath of *P. canaliculata*, but which are totally absent in *T. geversianus*. The richness of morphological characters found in the glands of both *P. canaliculata* and *O. magellanica* suggests its usefulness for phylogenetic studies.

53.

BM39 - SIGNALLING PATHWAYS IN RESPONSE TO HYPEROSMOTIC STRESS IN *Trypanosoma cruzi*

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Transmission of Chagas disease depends on *Trypanosoma cruzi* development and differentiation in the rectum of triatomine insects, where increased osmolarity is caused mainly by elevated content of NaCl from urine. Previous result showed that InsP_3 was accumulated in response to hyperosmolarity. InsP_3 accumulation was inhibited in the presence of BAPTA showing calcium requirement for PLC activity. Very little is known about the early biochemical events in this parasite, in response to high osmolarity. In this work, we studied the effect of high osmolarity on the functionality of *T. cruzi* epimastigotes Na^+/H^+ exchanger. We found the alkalization of acidic vacuoles via a Na^+/H^+ exchanger increased in response to 0.5M mannitol. Under the same conditions, cytosolic calcium concentration increased. The both effects were inhibited when the parasites were pre-treated with EIPA. The alkalization and calcium signalling were activated by PMA and 8-Br-cAMP and inhibited by Chelerythrine and KT5720 respectively, suggesting the Na^+/H^+ exchanger is regulated by PKC and PKA. 0.5M NaCl or 1M mannitol also caused an increase in number of intermediate forms. The inhibition of the Na^+/H^+ exchanger by EIPA, delayed these parasite forms. These findings show that hyperosmolarity induces PLC activation mediated by Ca^{2+} release from acidocalcisome. We suggest these events could be key steps in the differentiation process.

54.

BM40 - HIGH-FRUCTOSE DIET ELEVATES REACTIVE OXYGEN SPECIES IN MICE ARTERIAL WALL

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Oxidative stress represents an imbalance between the reactive oxygen species (ROS) production and antioxidants and is associated with several diseases where insulin resistance and hyperglycemia are common events. We evaluated NAPH oxidase activation, a main ROS generation system in vascular wall, using experimental animal model and cell culture. C75BL6 mice were fed with 10% fructose for 1 month to induce hyperinsulinemia. Then, tempol (1 mmol / L), a superoxide dismutase mimetic, was given for one month. Animals were sacrificed and NADPH oxidase (NOX) activity was measured by chemiluminescence in aortic tissue. Fructose increased the activity of NOX, and tempol prevented this increase (1.81 ± 0.34 URL/mg/min vs 1.02 URL/mg/min ± 0.31 P<0,01). In order to establish which cell type was involved in this effect we measure AngiotensinII (AII)-induce ROS production in mouse artery smooth muscle cell and mice macrophages by fluorometry. Tempol and Apocinin (a NOX inhibitor) completely abolished AII-induce ROS generation in both cell types. Our results support the hypothesis that pharmacological intervention aimed to reduce ROS generation would attenuate the changes in the vascular wall that are caused by oxidative stress associated with hyperglycemia.

55.

BT4 - ELIMINATION OF YEAST PHOTODYNAMICALLY SENSITIZED BY A POLYMER SURFACE DOPED WITH PORPHYRIN

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Photodynamic inactivation (PDI) has been proposed for the treatment of pathogenic microorganisms. The combined action of a photosensitizing agent and visible light leads to a lethal damage in microbes. In these systems, the removal of the sensitizer after the treatment may be necessary due to the accumulation of the agent in living organisms. In this work we synthesized bridged silsesquioxane films doped with 5-(4-carboxyphenyl)-10,15,20-tris (4-methylphenyl)porphyrin, in order to obtain a surface with aseptic properties activated by visible light. Absorption and fluorescence studies of surface shows the bands characteristic of porphyrins. Also, the film produces singlet molecular oxygen as the main cytotoxic species. In biological systems, the photodynamic effect was studied in *Candida albicans*. Inactivating action of the surface produces a decrease of ~ 1 and ~ 2 logs in cell viability after 30 and 60 min of irradiation, respectively. This activity was also confirmed by phototoxic studies of delay in the growth curve of yeast. The main advantage of using polymer surfaces is that it can produce PDI of microbes in a liquid suspension irradiating with visible light without polluting the environment with the sensitizer. The difference with other systems is that the film can be removed easily and then it can be reused in new PDI procedures.

56.

BT5 - PHOTODYNAMIC INACTIVATION OF YEAST SENSITIZED BY PORPHYRINS SUBSTITUTED BY AMINO GROUPS

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The development of new therapeutic alternatives such as photodynamic inactivation (PDI) is necessary due to antimicrobial resistance. The PDI is based on the incorporation of the sensitizer in microbial cells and the subsequent illumination with visible light induces lethal damage in cells. In this study the photodynamic activity of 5,10,15,20-tetrakis [4-(3-*N,N*-dimethylaminopropoxy)phenyl]porphyrin (TAPP) and 5,10,15,20-tetrakis [4-(3-*N,N,N*-trimethylaminopropoxy)phenyl]porphyrin (TAPP⁴⁺) were compared on *Candida albicans*. The quantum yields of singlet molecular oxygen were 0.74 and 0.72 for TAPP and TAPP⁴⁺, respectively. The cultures treated with 5 μM porphyrin showed a binding of 1,04 for TAPP and 1.5 nmol/ 10^6 cells for TAPP⁴⁺ after 30 min of incubation. Cell survival with both sensitizers caused a decrease of ~ 5 log, after 30 min of irradiation. The PDI was also confirmed by the delay in the growth curves of *C. albicans*. Studies of fluorescence microscopy showed that the sensitizers are located in an irregular manner into the cells. TAPP⁴⁺ presents positive charges on the periphery of the macrocycle, while TAPP bears aliphatic amino groups. At physiological pH, these amino groups are in equilibrium with the protonated form giving intrinsic positive charges on the agent. This effect can significantly increase its activity in the PDI.

57.

BT6 - ANTIFUNGAL EFFECT BY TETRACATIONIC PHTHALOCYANINES ON *Candida albicans*

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The resistance of *C. albicans* has been increasing even against traditional antifungal agents. The photodynamic inactivation (PDI) has been proposed for the elimination of microorganisms. In this treatment, the sensitizer is accumulated in microbial cells, which in the presence of light are inactivated by the formation of cytotoxic species. In this work were studied the spectroscopic, photodynamic properties and the PDI of *C. albicans* sensitized by Zn(II)2,9,16,23-tetrakis[4-(*N*-methylpyridyloxy)]phthalocyanine (ZnPyPc⁴⁺) and Zn(II)2,9,16,23-tetrakis(*N,N,N*-trimethylamine)phthalocyanine (ZnAmPc⁴⁺). Both sensitizers presented a singlet molecular oxygen production of ~0.6. The cultures treated with 10 µM of ZnPyPc⁴⁺ and ZnAmPc⁴⁺ for 30 min of incubation showed a binding of ~4.80 and ~0.17 nmol/10⁶ cells, respectively. The amount of cell-bound sensitizer diminished with the number of washing steps. Cell survival after irradiation was dependent on the concentration used and the dose of light, producing a decrease of 5 log at 10 µM ZnPyPc⁴⁺ and 30 min of illumination, while the inactivation was lower in cultures treated with ZnAmPc⁴⁺. Moreover, this activity of ZnPyPc⁴⁺ was also confirmed by the delay in the growth curve. Studies indicate that ZnPyPc⁴⁺ presents potential applications as phototherapeutic agent for the inactivation of yeast by PDI.

58.

BT8 - TETRADECYLTRIMETHYLAMMONIUM DEGRADATION BY FREE AND IMMOBILIZED *Pseudomonas putida*

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Tetradecyltrimethylammonium bromide (TTAB) degradation by immobilized *P. putida* A ATCC 12633 and free cells suspension was studied under controlled conditions. For suspended cells culture, exponentially growing cells were added to Erlenmeyer flasks containing M9 medium. For immobilized cells, 20mM HCl-Tris pH 7.4 plus KCl 44mM, NaCl 85mM, was used. In both conditions, TTAB 50, 100 and 170 mg l⁻¹ was added. The degradation process was carried out at 30°C on a rotary shaker at 100 rpm. Samples from the culture were withdrawn under sterile conditions at different incubation period for the analysis of residual TTAB and trimethylamine (TMA), an intermediate product in the degradation of TTAB. The free cells degraded 28 mg l⁻¹ of TTAB after 48 h of incubation from an initial 50 mg l⁻¹ TTAB and they lost their degrading activity when the initial concentration was increased to 100 mg l⁻¹. The alginate-immobilized cells degraded 117 mg l⁻¹ TTAB after 24 h of incubation from an initial 150 mg l⁻¹ of TTAB. An inhibitory effect of the intermediary TMA from TTAB degradation was observed in free cells but it was not presented in the immobilized system. This effect was countered after additions of AlCl₃ 0.1 mM. Overall, these results revealed that the immobilized cell systems are more efficient than suspended cells for TTAB degradation.

59.

BT12 - PRODUCTION OF OXYGEN IN AQUATIC PLANTSBertoluzzo SM², Bertoluzzo MG¹, Agostinis F², Balboa E², López D¹, Rigatuso R¹.¹Fac de Cs. Bioq y Farm., Taller de Física, UNR. ²Fac de Cs Médicas, UNR.

The phytodepuration is to exploit the ability of green plants to metabolize contaminants, which can be nutrients for plants, with the help of solar energy. This paper analyzed and quantified the production of oxygen of *Elodea* in the presence of white light and different wavelengths within the visible spectrum to determine at which wavelength the oxygen generated in photosynthesis is optimum. We work with *Elodea Canadensis*, as this plant produces large amounts of oxygen and absorbs nutrients such as nitrates and nitrites. In order to measure the oxygen production small clear glass bioreactors at laboratory scale were utilized. A given volume of tap water with antichlorine was placed in each bioreactor and a previously weighed piece of *Elodea* was dipped. One of the systems was undergone to radiation of white light for 40 minutes after which the number of bubbles produced in 5 minutes and the total volume of oxygen produced were recorded. The same procedure was carried out with the rest of bioreactors, but subject to different wavelength within the visible spectrum. The results indicated that the bioreactors subject to wavelength corresponding to red provided the greatest volume of oxygen. It is concluded that the photosynthesis in *Elodea* is optimal when the plant is illuminated with red light. This result will be used to build bioreactors with *Elodea* which allow water denitrification at low cost and without creating other pollution.

60.

BT13 - PHENOTYPE AND GENETIC METHODS FOR IDENTIFICATION OF LAB STRAINS PRODUCING BACTERIOCINS

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Lactic acid bacteria (LAB) belong to groups microaerophilic Gram-positive bacteria that have a low G + C and play a vital role in fermentation processes, biopreservation. The detection and identification of various species LAB through rapid methods is important for the control of products and identification of probiotic strains. The aim of this study was to identify the molecular level LAB strains isolated from goat milk showed antimicrobial activity against food pathogens. We evaluate and compare identification results at the genus level using an API 50 CHL biochemical method against a genetic method (PCR) using primers specific for the 16S-23S intergenic regions of rRNA. Six strains were selected for PCR of 23 isolates characterized as LAB. The biochemical identification showed that lcSL3, lcSL11 and lcSL9 strains belong to the *Lactococcus lactis* ssp. *lactis* 2 specie and lcSL5, lcSL23 and lcSL20 strains to the *Lactobacillus paracasei* ssp. *L. paracasei* 1 specie. Verified that with the exception of strain 9, remaining five genera correspond to previously characterized by biochemical method. From this we conclude that although phenotypic identification methods are appropriate, are inadequate and must be confirmed using molecular techniques.

61.

BT15 - ANTIMICROBIAL ACTIVITY OF *Lactobacillus paracasei* and *Lactobacillus plantarum* ISOLATED FROM REGIONAL GOAT MILK

Troncoso N, Collado S, Mitjans N, Bonomi M, Stagnitta P, Rezza I. PROIPRO Project 02-0506 – UNSL.

Lactic acid bacteria produce substances called bacteriocins that can be used in food preservation. *Enterococcus faecalis* and *Listeria monocytogenes* are bacteria pathogens that contaminate food. The aim of our study was to isolate new strains BAL with antimicrobial activity. MRS medium was used and the isolates were characterized by testing catalase, oxidase and Gram stain, was used for typing Kit CH50 API. Antimicrobial activity was determined by the well diffusion method and the peptide nature of the antimicrobial substances by the action of proteases. The same activity was determined using TSC broth with *E. faecalis*, by reading the OD at 625 nm at 0, 3 and 6 h. Four strains were isolated and characterized positively BAL and classified as *Lactobacillus paracasei* ssp *paracasei* 1 (*lcSL50* strains, and *lcSL57 lcSL53*) and *Lactobacillus plantarum* 1 (*strain lcSL59*). These strains showed inhibition with halos between 9-11 mm. The antimicrobial activity after incubation with pepsin was negative, indicating the peptide nature of the inhibitor. It can be concluded that the four isolates are producing bacteriocins and inhibit the growth of *L. monocytogenes* and *E. faecalis*. Further studies on isolated BAL and characterization of bacteriocins, would determine their possible use in the food biopreservation.

62.

BT16 - ISOLATION OF PHENOLIC COMPOUNDS DEGRADING MICROORGANISMS FROM OLIVE-OIL MILL WASTESMonetta P¹, Bressan F², Crespo D², Aguilera M¹, Gouiric S³, Vallejo M³.¹EEA SJ-INTA. ²IMYZA-INTA. ³IBT-UNSJ.

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Modern olive-oil mills generate a semi-solid residue commonly called “alperujo” (AL) that presents serious environmental problems related to its disposal. It high phenolic load constitute an important source of water and soil pollution when it is improperly disposed. In order to obtain useful tools to detoxify and enable the later reuse of AL, microorganisms were isolated from AL and then, their ability to decolorize and degrade it phenolic content was assessed. The isolations were performed at 25°C using conventional culture media. Later, a screening of strains capable to decolorize AL was performed in Petri dishes containing basal medium and aqueous extracts of AL (10% w/v). The visualization of a translucent halo around the colony was considered as positive. In parallel, a strain of *Bjerkandera adusta* was used as positive control. Subsequently, Solid-State Fermentations (SSF) were performed using AL as unique substrate (25°C, moist 40%, pH 5.5; inoculated with four 5mm side agar cubes). After 15 days, phenolic content was evaluated. The results show that 11 yeast and 17 filamentous fungi strains were isolated from AL. Among them, 8 presented ability to decolorize AL water extracts. Finally, the SSF assays showed that 3 strains of filamentous fungi were capable to grow in AL and decrease it phenolic content, suggesting that SSF is an interesting alternative for AL detoxification.

63.

BT17 - GROWTH KINETIC CHARACTERIZATION OF NEW WINE YEASTS FROM SAN JUANMaturano YP¹, Rodríguez Assaf LA^{1,2}, Vallejo MD¹, Zapata MJ^{1,2}, Mestre MV¹, Toro ME¹, Vázquez F¹.¹IBT-FI-UNSJ. ²FCEFYN-UNSJ. San Juan. Argentina. E-mail: paolamaturano@yahoo.com.ar

Saccharomyces cerevisiae is the main yeast in winemaking. Also, non-*Saccharomyces*, may be present and affect wine characteristics. Winemakers prefer controlled fermentations (selected yeasts inoculation). A prerequisite for controlling the characteristics of the wine is verifying its fermentation kinetic. In this work, essential parameters and growth kinetics of three wine yeasts *S. cerevisiae* BSc562, *Hanseniaspora viniae* BHv438 and *Torulaspora delbrueckii* BTd259 (from San Juan), in aerobic and fermentative conditions, were determined. Pedro Jimenez pasteurized must (28°Bx, 300 mg/L assimilable nitrogen, total acidity 5.5 g/L and pH 4.25) was used. Yeasts were inoculated at 3x10⁶ cells/ml and incubated at 20°C. Samples were taken and biomass was quantified by dried weight and colony forming units. For aerobic conditions, all yeasts exhibited logistic growth profiles: lag time (t_L) 2.96h (BSc562), 0.58h (BHv438) and 2.19h (BTd259); maximum specific growth rate (μ_{max}) 0.465 h⁻¹ (BSc562), 0.434 h⁻¹ (BHv438) and 0.467 h⁻¹ (BTd259). Maximum cell concentrations (X_{max}) were attained at 24 h: 190; 99.5 and 527 millions/mL. For anaerobic conditions, t_L increased 10 times for BHv438 and 5 times for BTd259; μ_{max} decreased for BSc562, BHv438 and BTd259 about 10, 15 and 18 times, respectively. These yeasts can be used as starters in controlled fermentations.

64.

BT20 - MULTIWALLED CARBON NANOTUBES FUNCTIONALIZED WITH CHITOSAN, BIOCOMPATIBILITY ASSAYAlustiza F¹, Bellingeri R¹, Picco N¹, Motta C¹, Peralta M¹, Busso L¹, Molinero D¹, Grosso M¹, Molina A², Acevedo D², Vivas A¹.¹Anat., FAV; ²Qca, FCEFYN-UNRC.

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The aim of this study was to evaluate the Multiwalled Carbon Nanotubes (MWCNT) functionalization with chitosan, test its solubility and its biocompatibility in primary culture of spleen lymphocytes from chickens. MWCNT were functionalized in 1% acetic acid solution with 0.1g of chitosan. Near infrared mass spectroscopy was performed. Four Lohman Brown laying hens were used, spleens were removed and lymphocytes were separated with Histopaque 1070 and cultured in RPMI 1640. Trypan blue viability assays were performed before and after 24 h of culture. Experimental design, groups: 1- Control-, 2- with MWCNT, 3- with chitosan, 4- with MWCNT functionalized, 5- Control+ stimulated with *E. coli* F4. MWCNT were functionalized, as determined by IR spectra (1100 cm⁻¹), were solubilized in aqueous media presenting a density of 1.3 mg/ml. Lymphocytes showed an initial 97% viability, group 2 showed 65% viability with insoluble MWCNT precipitates and cellular debris, group 3 showed 95% viability with clusters on non-solubilized chitosan granules, groups 1 and 4 showed 95% viability and regular clusters compared with group 5. MWCNT were successfully functionalized with chitosan, solubilized in aqueous medium and improve its biocompatibility in spleen lymphocytes culture.

65.

BT23 - EMBRYOGENIC CALLUS CULTURES IN *Prosopis caldenia* BURK

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The somatic embryogenic is considered as an efficient form of clonal multiplication, offering a great potential as tool in the forest investigation. The objective of the present work was to select the type of appropriate explant and to determine the appropriate nutritious medium for the induction of organogenyc or embryogenic callus in *Prosopis caldenia* Burk. The used explants was: meristematic tissue, leaves tissue, mature embryos and roots of seeds germinated *in vitro*. The nutritious medium was Murashige and Skoog, with 2 sucrose% and 0,6 agar%. Supplement with different combinations of 2,4-diclorofenoxyacetic acid (2,4-D) and acetic indol acid (NAA). The cultivations stayed in cultivation camera with a temperature of 24+/-2°C; half of them with a fotoperiod of 16 hours of light and 8 of darkness, and the other half in darkness. After 40 days, significant differences were observed at level p 0.05 (Test G) in the formation of greenish friable, yellow calli, for cotyledons and hipocotyl like explantos, incubated under conditions of light and with 2,4-D (2 mg/l) and NAA (0.5 mg/l). The sections histological of these explants showed areas of embryogenic cells. These preliminary studies provide the base to consider this technology like a valuable tool for the conventional techniques of propagation and vegetable improvement of this species.

66.

BT24 - EVALUATION OF INDUCED POLYPLOIDS *IN VITRO* IN *Gomphrena pulchella* MART

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The use of native species in the landscape designs allows to establish gardens and sustainable green spaces. *Gomphrena pulchella* Mart., it possesses interesting ornamental attributes. Presently work was evaluated the effect of different treatments with colchicine in segments nodal micropropagated of this species, in search of polyploid exemplary with characters improved fenotipic of ornamental value. The duplication of chromosomes or polyploidy allow the obtaining of organs of more size, especially leaves and flowers, an escalation of the color, plants more compact and smaller behavior. The micropropagated plants between nutritious Murashige and Skoog medium without hormones was submerged in 1% dimethylsulfoxide (DMSO) solution with the following doses of colchicine (V/V): 0.0%; 0.1%; 0.05%; 0.01% and 0.001% (12 and 24 hours). The level of the polyploidy was evaluated using flow citrometric (Partec, CA). Of the number of plants analyzed by the flow citrometric, 10% of plants chimeric was detected (2X/4X) in the treatments 0.05% and 0.01% with 12 and 24 hours of exhibition. The concentration of 0.05% (24 hours) of colchicine allowed to obtain 60% of plants solid tetraploid (4X), with highly significant differences regarding the rest of the evaluated treatments. Confirmed the effectiveness of the colchicine like polyploidy agent.

67.

BT25 - PHYSICAL CHEMICAL STUDY OF STIMULI IN THE GERMINATION OF *Trichloris crinita*

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In San Luis province the resource forage of more relevance is the natural grasses, dedicated basically to the activity of bovine breeding. The loss of diversity of the herbaceous stratum takes to implement strategies to preserve these threatened species. *Trichloris crinita* is a perennial species, of summery cycle and of medium to high palatability. But it is characterized to possess seeds of low viability, with persistent latency. The objective of the present work was to establish the environmental parameters, nutrition and aseptically appropriate to achieve a germination rate *in vitro* efficient. The seeds were disinfected with alcohol 70%, sodium hypochlorite and methyl bencimidazol-2-il carbamate (carbendazim), in different concentrations and time of exposure. A half solid of Murashige and Skoog medium was used. Four treatments like physical stimuli were applied (cool and humidity) and chemical (AG3, 1 mg/l) to break latency. The smallest rate of contamination was achieved with the following disinfection: alcohol 70% (15 minutes), sodium hypochlorite 40% (15 minutes), carbendazim 1% (15 minutes). The treatment that achieved the significantly higher percentage of germination was the treatment with AG3, in cultivation camera to 24 ± 2°C with fotoperiod of 16 hours (48 mmol. S-1.m-2). A significant progress is contributed regarding the germination requirements for the multiplication of forage native species.

68.

BT26 - *IN VITRO* CALLUS CULTURES IN *Gomphrena pulchella* MART

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The *in vitro* culture allows the massive propagation of plants, germplasm conservation and improvement genetics, being the biotechnology tool of more practical contribution. With the objective of implementing strategies to spread, to improve genetically and to conserve the existent genetic variability in populations of *Gomphrena pulchella* Mart., they were carried out rehearsals to induce the callus cultures *in vitro*. The cultivation *in vitro* of segments nodals obtained starting from young tissue of selected mature individual. The nutritious means was of Murashige and Skoog (50%), supplemented with 30 sucrose g/l, 6 agar g/l and different combinations of 2,4-diclorofenoxyacetic acid (2,4-D). The cultivations were incubated in cultivation camera to 24 ± 2°C with fotoperiod of 16 hours (48 mmol. S-1.m-2). to the 30 days of cultivation it was determined that the treatment with 2,4-D (2 mg/l) it induced the formation of friable calli, and 16% of the explants they formed adventitious roots and stems. This indirect morfogenesis was not observed in the rest of the treatments. It is in the evaluation stage the appearance of varying somaclonals and the capacity *in novo* organogenic in calli with different extension of the period of cultivation between nutritious MS supplemented with 2,4-D. By means of the setting about to the cultivation *in vitro* the possibility opens up of to conserve and to characterize molecularly of new genotypes.

69.

BT27 - EVALUATION OF THE BEHAVIOR OF INDUCED POLYPLOIDS OF *Gomphrena pulchella* MART. DURING THE ACCLIMATIZATION STAGE

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Gomphrena pulchella Mart. is a native species that possesses showy inflorescences, as interesting ornamental attribute. In order to develop strategies of genetic improvement in this species, biotechnology tools have been used to induce mutation and multiplication *in vitro*. The transplanted of the vitroplants to diverse substrates and their adaptation to conditions heterotrophic, they define the success of the micropropagation. This way, the objective of the present work was to evaluate the conditions of acclimatization of exemplary polyploids of *G. pulchella* Mart., obtained *in vitro*. The used substrates were soil, peat, pearl, lombricompost and their mixtures in different proportions. The physical-chemical characteristics of the substrates and their interaction were evaluated with the survival and growth of the vitroplants polyploids. The substrates that favor the vegetative growth and the viability or survival (100%), they are the treatments T1 (soil) and T6 (soil: peat: lombricompost in relationship 1:1:1, v/v). The substrates that favor the floral permanency in the plants, possesses high content of organic matter and readiness of nutritious. This work contributes advances again in the obtaining germplasm with commercial ends and with great potential in the design of sustainable green spaces.

70.

BT28 - POTENTIAL APPLICATION OF A NATIVE MICROORGANISM FROM CORDOBA IN BIOREMEDIATION

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Bioremediation is an important process to reduce or remove organic and inorganic pollutants from the environment. Chromium and phenolic compounds are examples of these hazardous pollutants. In the present work, we analyzed the capability of the rhizospheric microorganism RTS 13 A.3, a Gram negative bacteria isolated from a polluted industrial area near Rio Tercero, to tolerate and remove high concentrations of chromium (VI), phenol and 2,4-dichlorophenol (2,4-DCP), with the aim to evaluate the potential of this microorganism for bioremediation processes. RTS 13 A. 3 was able to grow up to 300 mg/l Cr (VI), 1000 mg/l phenol and 100 mg/l 2,4-DCP in TY medium, while in mineral medium, MM9, the microorganism tolerated smaller concentrations. In the removal assays, a total Cr (VI) reduction was observed in TY medium when the concentration were 5, 10 and 20 mg/l after 1, 3 and 7 days of incubation, respectively. In addition, a 75% of removal was observed in a solution initially containing 50 mg/l after 7 days. In the same period of time, a decrease in the chromium removal was observed in MM9 medium supplemented with glucose 1 mg/L. Regarding to phenolic compounds removal, the microorganism was able to remove 100 mg/l of phenol and 20 mg/l of 2,4-DCP in MM9 medium after 14 days. These results could indicate the potential of RTS 13 A for future applications in bioremediate environmental pollutants.

71.

BT29 - EVALUATION OF THE ANGIOGENIC EFFECTS OF NOVEL NANOSTRUCTURED COMPOSITESHaro Durand L¹, Vargas GE^{1,2}, Cadena V¹, Romero M², Vera Mesones R³, Zago MP¹, Maëkovic M³, Spallek S³, Spiecker E³, Boccaccini AR³, Gorustovich A¹.¹Interdisciplinary Materials Group-IESIING-UCASAL, INTECIN UBA-CONICET, Salta, Argentina; ²UNSA; ³University of Erlangen-Nuremberg, Germany. E-mail: agorustovich@conicet.gov.ar

Collagen materials have been utilized in medicine because of their proven biocompatibility and capability of promoting wound healing. The aim of present study was to evaluate the effect of adding nanoscale silicate bioactive glass particles (n-BG) on the angiogenic properties of bovine type I collagen/n-BG composites. Nano-sized (20-30 nm) BG particles of nominally 45S5 Bioglass[®] composition were used to prepare composite films, which were characterised by SEM-EDS and TEM. The angiogenic response was evaluated using the quail chorioallantoic membrane (CAM) as an *in-vivo* model of angiogenesis. At 24 h post-implantation, 10 wt% n-BG containing collagen films stimulated angiogenesis by increasing 41% the number of blood vessels branch points. In contrast, composite films containing 20 wt% n-BG were found to inhibit angiogenesis. This experimental study provides the first evidence that addition of a limited concentration of n-BG (10 wt%) to collagen films induces a strong angiogenic response making collagen/n-BG composites attractive matrices for applications in tissue engineering and regenerative medicine, in particular for the regeneration of highly vascularised tissues such as bone.

72.

BV3 - COMPARATIVE NUTRITIONAL AND MINERAL VALUE OF *Ilex paraguariensis*, *I. dumosa* AND THEIR MIX

Maiocchi MG, Del Vitto LA, Machevsky EJ, Pellerano RG, Avanza MV, Petenatti EM*.

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“Yerba mate” is a nutritional product traditionally made in South America with leaves and tender stems of *Ilex paraguariensis* A. St.-Hil. (Aquifoliaceae) (IP). Recently Argentinian market has joined *I. dumosa* Reissek (“yerba señorita”) (ID), with lower xanthine content, that appears in mixing with the former, labeled “Suave-Relax” (70% ID and 30% IP) (ID+IP). This study was conducted to compare nutritional and elemental parameters of “yerba mate” prepared from IP, ID, and ID+IP, both in raw drug as the two forms most frequently used in the region: “cebadura” (discontinuous extraction) and “mate cocido” (c. 1.5% infusion). Crude drug and extracts were analyzed using AOAC and IRAM, Kjeldahl, HPLC-UV, and ICP-OES methods. Studied products were within the variability allowed by Argentinean Food Code. Nutritional properties vary mainly in xanthines and minerals, that were greater in IP. Highlighted the low level of caffeine in samples of ID (0.1-0.25g%g) compared with IP (1.25-1.7g%g). Chlorogenic acids ranged from 5.1-6g%g in ID and 14-16g%g in IP. “Cebadura” is source of Mg, Mn, Fe and Cu, and minor source of Zn, Ca, and P, while “mate cocido” shows lower input as the minor serving size. Both species show low content of Na and high concentration of Mn. Regarding ID+IP, analytical values were closely related with their 70:30 ratio.

73.

BV6 - SECONDARY METABOLITES WITH PHYTOTOXIC ACTIVITY IN *Flourensia campestris* (ASTERACEAE)Piazza LA^{1*}, Silva MP¹, López D¹, López Rivilli MJ¹, Turco MD², Cantero JJ³, Tourn MG¹, Scopel AL¹.¹FAUBA Punilla, ²CEPRO COR, ³FayV UNRC.

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As part of our studies on the ecophysiological adaptations of native plants of Argentina we have focused on the elucidation of phytotoxic effects from *Flourensia campestris* (FC). The aqueous extract from FC dry aerial parts showed strong inhibition on the germination and growth of *Lactuca sativa*. Based on bio-guided fractionation of aqueous extracts and spectral means the (-)-hamanasic acid A (7-carboxy-8-hydroxy-1(2), 12(13)-dien-bisabolene (**1**)) was isolated as the most inhibitory active principle (EC₅₀) on germination (2.9 mM) and on root (1.5 mM) and shoot (2.0 mM) growth. As measured by GC-FID, and correlated with a simple designed 2D-TLC, compound **1** was found to be distributed throughout the plant, with a remarkable high concentration (1.6%) in leaves and inflorescences. Leaf essential oils (HD) between 0.5 and 1.5 µl ml⁻¹ did not show herbicidal effects and **1** was not found in them (TLC) nor among volatiles (HS-SPME). Volatiles composition was assessed by GC-FID and GC-MS and led to the identification of 23 compounds (4 monoterpenes and 19 sesquiterpenes), some of them identified as phytotoxic compounds against other species. The high stores of **1** in FC together with its feasibility of being extracted with water strongly suggest that the compound is indeed a potent allelochemical in this species. Species-specific studies are under way to evaluate the potential of **1** as a natural herbicidal compound.

74.

BV9 - ORDERING OF HYBRID CORN USING MOLECULAR AND MORPHOLOGICAL DESCRIPTORS

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Breeders can use molecular techniques as complementary tools in the phenotypic characterization of genotypes of interest. To interpret the genetic relationships can be established separate characterizations of each type of variable, use all the information together, or apply management techniques consensus through harmonization of configurations. The aim of this study was to interpret the genetic relationship between corn hybrids with a consensus ranking among genetic and phenotypic descriptors. Sixteen corn hybrids from five commercial breeding programs were characterized with polymorphic bands from SSR markers, and three phenotypic traits. With phenotypic variables and the four axes of a principal coordinates analysis that explained 70% of the genetic variation, we applied a generalized procrustes analysis (APG) and selecting the minimum path tree (ARM). Using APG sought to quantify the consensus among the different ordinations of hybrids that were obtained through genetic and phenotypic data. The consensus of the order produced was 83%. The ARM consensus configuration allowed us to visualize associations between hybrids according to their origin. The APG was very useful both to order the corn hybrids from different commercial breeders, to find a common space where they best matched the order of the two different sets of descriptors used.

75.

BV10 - SYSTEMIC CHANGES IN THE SOYBEAN (*Glycine max* L.) - *Bradyrhizobium japonicum* INTERACTION

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In the last decades many complex local mechanisms involved in Rhizobium - legume association have been elucidated. Besides this exist a systemic signaling that it hasn't been described in early stages of the symbiotic infection. The objective was analyze redox systemic changes during the early stages of soybean - *B. japonicum* interaction. Soybean (*Glycine max* L. DM4800) plants 12 days old were inoculated with *B. japonicum* USDA138 and the first trifoliolate leaf was used for several measurements. We also determined the involvement of NADPH oxidase through DPI inhibition. We found an increase of superoxide radical and hydrogen peroxide in inoculated plants after 30 and 60 and 120 min respectively. Inoculated and DPI-treated plants at both leaf and root showed hydrogen peroxide measurements without differences with control plants. Moreover we found an activation of enzymatic/ non enzymatic antioxidant system after 30 min post - inoculation, an increment of PAL activity after 30 and 60 min, and an increment of ethylene after 120 min post- inoculation. Our results show that inoculation of soybean with *B. japonicum* induces redox changes in conjunction with early systemic activation of secondary metabolism, and subsequent increases in ethylene content. This oxidative early signaling would be mediated by the NADPH oxidase complex and would have a fundamental importance in modulating of systemic responses induced by symbiotic microorganisms.

76.

BV11 - SYMBIOTIC ASSOCIATIONS WITH ARBUSCULAR MYCORRHIZA IN *Medicago sativa*: RELATION TO STRESSPedranzani HE^{1,2}, Manella Hoyos JL², Quiroga AM¹, Ruiz Lozano JM².¹Lab.Fisiol. Vegetal. FICES. UNSL Argentina. ²Lab. Microbiología de Suelos. CSIC. Granada. España.

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The aim of this study is to investigate the association of arbuscular mycorrhizal (*Glomus intrarradices*) with plants of *Medicago sativa* var. Verdor and analyze their effects on a biotic stress mitigation Seedling of *Medicago* was transplanted at pots of 1000 ml with soil/sand (1:1, v/v) with 10 gr *G.intrarradices* inoculums. The AM and non AM plants grew in a chamber at 23:21°C with 16:8 photoperiod and watered to field capacity: 1) 23°C and soil at field capacity (no stress) 2) 23°C and 7% water for one week (dry) 3) 4°C for 24 and 72 h (cold) 4) irrigated with 200 mM NaCl for two weeks (salinity). Mycorrhization %, stomata conductance (SC), photosynthetic efficiency (PE) and % dry matter in leaves (DM) was measurement. Mycorrhization ranged from 59.5 to 66%. The SC increased in the AM plants without stress and with it, although there were no differences salt stresses. The PE decreased compared to different stresses in non AM plants, while in the AM plants increased or remained indifferent. The cold reduced the % DM in AM and non AM plants. The % DM was significantly increased in the AM plants with respect to non-AM, showed a mitigating effect. We concluded that the symbiosis association with alfalfa, send tolerance to stress better trough the efficiency parameters SC, PE and increased % DS.

77.

BV12 - MYCORRIZAL *Digitaria eriantha* cv. Sudafricana: CONTRASTING RESPONSE TO ABIOTIC STRESSPedranzani HE^{1,2}, Molina Arias SM², Rodríguez Rivera M¹, Ruiz Lozano JM².¹Lab. Fisiol. Vegetal. FICES. UNSL Argentina. ²Lab. Microbiología de Suelos. CSIC. Granada. España.

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Digitaria eriantha cv. Sudafricana has high sensibility to different biotic stresses. In this study the aim was to study the response of different morphometric and physiological parameters in mycorrhizal and non mycorrhizal plants under drought, salinity and cold conditions. Seedling of *Digitaria eriantha* cv. Sudafricana of 17 days was transplanted at pots of 1000 ml with soil/sand (1:1, v/v) with 10 gr *G. intraradices* inoculum. The AM and non AM plants grew in a chamber at 23:21 °C with 16:8 photoperiod and watered to field capacity: 1) 23 °C and soil at field capacity (no stress) 2) 23 °C and 6% water for one week (dry) 3) 4 °C for 72 h (cold) 4) irrigated with 200 mM NaCl for two weeks (salinity). Percentage of mycorrhization, stomata conductance (SC), photosynthetic efficiency (PE) and % dry matter in leaves and roots (DM) were measured. The inoculated plants showed 70% of mycorrhization and didn't decrease with a biotic stress. The SC showed a significant decrease in all stress in AM and non AM plants, however without stress, the SC of AM plant increased. The PE didn't change in control plants (AM and non AM), but under stress increased in all stresses, except in drought. With stress, foliage DM%, increased in non AM plants, but in AM plants didn't change, only in drought. The roots DM %, in AM and non AM plants decreased in salinity and cold, but in drought increased. The AM plants DM% doesn't change showed the mitigation effects of arbuscular mycorrhizal.

78.

BV13 - IMMUNOPURIFICATION OF SECRETORY *Glycine max* PHOSPHOLIPASE A2

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Secretory Phospholipase A2 (sPLA2, EC 3.1.1.4) from soybean (*Glycine max*) was partially purified and biochemically characterized by the authors. PLA2 promotes the formation of lysophospholipids, strong bioemulsifiers. The objective of the present work was: to obtain *Gms*PLA2 pure to homogeneity with the aim of develop structural and kinetic studies. Polyclonal antibody IgG anti sPLA2 *Bothrops alternatus* was covalently immobilized upon Eupergit® C and C250, Degussa. 100 mg dry support was incubated 66 hs at 20 °C with antibody solution (2 mg/mL) in buffer borate. The systems were treated with Tris 200 mM to blocked unreacted support oxiranes. The active PLA2 fraction from affinity chromatography using Cibacron Blue as immobilized ligand, was incubated with Ac-Eupergit C, 400 mg and Ac-Eupergit C250, 600 mg 16 hs at 20 °C. Elution was performed with glycerol (30%) 0.15 M NH₄OH. PLA2 activity upon lecithin liposomes was obtained from apparent absorption at 340 nm. From three independent immunopurification experiments, it was found that Ac-Eupergit C was bound 29 ± 2 PLA2 units (purification factor= 6.1, yield= 19.1%) and Ac-Eupergit C250 was bound 17 ± 1 PLA2 units (purification factor= 5.1, yield= 11.2). SDS-PAGE electrophoresis showed two bands of 11 and 13 kDa proteins related with *Gms*PLA2 isoforms or homologue groups, as we demonstrate by recent molecular biology studies. It is the first time that *Gms*PLA2 was obtained to homogeneity.

79.

BV14 - RESISTANCE INDUCTION IN SOYBEAN: PHYTOALEXINS DETERMINATION

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The resistance induction, appears as a sustainable control strategy in plants. The biosynthesis of phytoalexins is a relevant early response in soybean (*Glycine max*)-pathogen interaction. This project is related with the induction effect of chitosan, a glucosamine polymer. The specific objective was: to develop a sensitive method for phytoalexin determination in soybean leaves. Phytoalexins are extracted by vacuum infiltration with aqueous 40% ethanol. The aqueous phase was extracted with ethyl acetate and the organic phase was dehydrated to dryness. Finally, it was measured the relative intensity at 268 nm of ethanol dilution, applying 2nd Derivative Spectrophotometry by using a HP 8452 diode array spectrophotometer. A correlation coefficient $r = 0.9984$ using genistein as standard, was obtained. Soybean seeds 4990 from INTA (Argentina) were soaked into chitosan 1 mg/mL acetic acid pH 5.0 and control assay. The growing plants were maintained three weeks in controlled conditions. Phytoalexin level of $32 \pm 2 \mu\text{g/g}$ fresh leaves was found in sample treated with chitosan, representing a 39% increase respect to control. Two weeks later, an increase of 108% was registered. This work allowed us to adjust the quantitative determination of phytoalexins and to obtain preliminary results about chitosan as inducer of phytoalexins biosynthesis in soybean leaves.

80.

BV15 - FATTY ACID CONTENT IN NEW REGIONAL VARIETIES OF AMARANTH GRAINS

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One of the risk factors for cardiovascular disease is inadequate diet, the foods rich in saturated and unsaturated fatty acids with trans configuration being the most harmful ones. The solution lies in further investigation on alternative food sources to improve the diet. *Amaranthus* (pseudocereal, dicotyledonous) is one of the oldest New World crops. The purpose of this work was to assess the content of fatty acids in two new regional varieties, *Amaranthus cruentus* var. candil (Acc) and *Amaranthus hypochondriacus* var. dorado (Ahd). The seed oils were analyzed by liquid gas chromatography of the methyl esters of total fatty acid and their chemical composition was compared with maize (*Zea mays*).

The main fatty acids found were linoleic acid (34.70-43.30%), oleic acid (22.80-30.00%), palmitic acid (13.60-16.10%) and arachidonic acid (12.00-10.30%). The total content of unsaturated fatty acids ranged between 78.70 and 81.80% and the total content of saturated fatty acids was between 18.10 and 21.30%.

The amaranth fatty acid profile was found to be similar to that of maize oil, a conventional cereal oil. The use of amaranth can be therefore recommended as an alternative source of oil for the human diet.

81. BV16 - LIPID METABOLISM IN BARLEY ROOTS (*Hordeum vulgare*) GERMINATED UNDER SALT AND OSMOTIC STRESS

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Salt stress is one of the abiotic factors that limit normal plant development and activates signaling processes. Salinity causes both osmotic and ionic effects; the osmotic is triggered by excess salt in the soil, while the ion is due to excessive salt accumulation in the cells. Phospholipids are membrane components that play important roles as mediators in signal transduction. Therefore it was necessary to determine changes at the lipid signals in roots of barley germinated 4 days under salt stress (100 mM NaCl) and osmotic stress (200 mM mannitol). Salt stress increased the levels of phosphatidic acid (PA), while mannitol caused not only increased levels of PA, it also increased the levels of diacylglycerol pyrophosphate (DGPP) due to the activity DAG-k and PA-k, respectively. DGPP phosphatase activity remained unchanged, while the PA phosphatase activity was decreased in both types of stress. PLD activity also increased in treatments with NaCl and mannitol. On this basis, one could say that abiotic stress (salt and osmotic) not only increased the levels of PA in roots by increasing the activity DAG-k but it would also generate a pool of PA from PLD activity. On the other hand, PI3-k and PI4-k activities were affected both in response to salt and osmotic stress, PI3-k would also be involved in the production of active oxygen species (EAOs) via the NADPH-oxidase complex in response to stress by mannitol but not salt stress. Otherwise, increases in PI4P could be mediated by increased levels of salicylic acid (SA). In relation to the results obtained, it is suggested an interconnection between signaling pathways, which are mediated by phospholipids triggered under stress conditions in barley roots.

82. BV17 - ABA EFFECT ON ANTIOXIDANT ACTION AND THE PRODUCTION OF FIELD-GROWN MAIZE PHOTOASSIMILATES IN SEMIARID REGIONS

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The aim of this study is to analyze the response of exogenous ABA application in plants grown under field conditions in semi-arid zones in order to increase maize production, after relieving the detrimental effect of water stress and enhance the availability of carbohydrates. The maize cultivars used was DK 747 MGRR, with 6 replicates of the treatments: Control (foliar spray of water) and ABA 300 mg l⁻¹ (foliar spray of ABA, 99%, Sigma-Aldrich) at V13. The variables analyzed took into account the photoassimilates production and partitioning towards the corncob and the yield components, as well as the antioxidant responses to water stress. This study showed that ABA in field-grown maize under moderate drought allows a greater amount of maize production, an increase in the level of photosynthetic pigments, the carbohydrates remobilization to grain, and the capacity of this transport by an increase in the number of vascular bundles and the phloem area in the corncob peduncles. In the ABA-treated leaves, a lower content of reductant sugars was observed in the R3. The regulation of the antioxidant enzymes activity and expression was modified in the ABA-treated maize plants, mainly in the APx. Evidence obtained in this study suggests that ABA could help improve agricultural production in rain-fed crops.

83. BV18 - PHYSIOLOGICAL AND BIOCHEMICAL MARKERS IN MYCORRHIZATION OF *Jatropha curcas* DURING EARLY DEVELOPMENT

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Jatropha curcas, a species producing oils for biodiesel was studied as a possible host of the arbuscular mycorrhizal *Glomus intrarradices*. We study physiological and biochemical markers at two stages of early development in mycorrhizal plants and controls. Plants of two stages of development were transferred to pots containing 1000 ml of substrate 1:1 (v/v) soil:sand to which was added 10 mg of *G. intrarradices*. M1 was called to plants with incipient development of hypocotyls, cotyledonary leaves and little roots and M2 to larger plants, with developed hypocotyls, cotyledonary leaves and roots larger than 5 cm. In leaves of control plants, M1 and M2 were determined: percentage of mycorrhization, ascorbate, proline and malondialdehyde (MDA) content, stomata conductance (SC), and photosynthetic efficiency (PE). The colonization by *G. intrarradice* was 60% of roots. Ascorbate and PE didn't change. SC, Prolina and MDA contents increased in M1 respect to control. CE y MDA were higher in M1 respect M2 whit significative difference. The increase in physiological markers such as SC and biochemists as proline and MDA, indicate reduced role of mycorrhizal in protecting stress-induced early growth. In the development stage M2, mycorrhizal provide more stability, with values similar to control. PE and ascorbate didn't change in the early stages of growth, and didn't varies by the micorrhizal associations in this species.

84. BV19 - AMINOACIDIC COMPOSITION OF TWO NEW AMARANTH VARIETIES

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Amaranth is a pseudocereal of elevated protein content and well-known nutritional qualities for the human diet. The purpose of this work was to evaluate the nutritional value of the amaranth protein as estimated on the basis of chemical score (CS). New regional varieties were used: *Amaranthus cruentus* var. Candil (Acc) and *Amaranthus hypochondriacus* var. Dorado (Ahd), of wide adaptation for cultivation and important yield. The aminoacidic profile was determined according to AOAC. Calculation of aminoacid score (AAS) was performed, from which the protein CS was derived. The Acc CS was found to be 47 for Acc and 45 for Ahd. It was observed that they are a good source of essential aminoacids according to values recommended by FAO/WHO (2007) except for valine which in both cases is a limiting aminoacid. Both Acc and Ahd showed elevated levels of lysine (47 and 60 mg/g respectively) which is an indispensable aminoacid of deficient content in cereals, including wheat grain. Leucine and threonine, which are usually scarce in this genus, were found in optimal amounts. These features make amaranthus an adequate protein complement for conventional cereals.

85.

BV20 - *Pseudomonas fluorescens* INDUCES METABOLIC CHANGES IN GRAPE PLANTS

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Pseudomonas spp. are plant growth promoting bacteria with beneficial effects on plants by production of phytohormones and induction of metabolic changes. We previously isolated *Pseudomonas fluorescens* from roots of grape cv. Malbec plants and characterized by GC-EIMS ABA, IAA and GA production in chemically-defined medium. The aim of this work was to evaluate metabolic changes in grape plants inoculated or not with *P. fluorescens*. Roots of 15 d-old grape plants grown *in vitro* were inoculated with 100 µl of microbial culture (2×10^7 CFUml⁻¹). After 45 days plants were removed, and leaves were collected for ABA, terpenes and pigment analysis. Inoculation with *P. fluorescens* did not produce morphological changes as compared with control plants, but increased significantly ABA, carotenes and chlorophylls. Also enhanced the synthesis of the monoterpenes pinene, terpinolene and cineole, and the sesquiterpenes farnesol, trans- α bergamotene, α -farnesene and nerolidol, whereas in controls only pinene and nerolidol were detected in trace amounts. The results indicate that *P. fluorescens* promotes increases in ABA and pigment concentration and induced the synthesis of defense-related compounds against potential pathogens.

86.

BV21- SEED BORNE PATHOGENS IN SOYBEAN (*Glycine max*) VARIETIES IN "VALLE DEL CONLARA" (SAN LUIS)

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Determination and control of seed borne pathogens are tools to be applied to reduce risk and increase crop productivity. With this objective, a trial with soybean seeds (varieties Dalia 457 (A), DM 5.4 (B) and DM3702 (C)) harvested in "Valle del Conlara" (San Luis) during the 2010 was performed. A total of 300 seeds per cv were sowed in APG 2% medium (10 seeds/Petri dish of 9 cm Ø). Incubation conditions were 7-9 days at 25°C + / - 2°C. Pathogens identification was based on the reproductive structures and incidence was determined as number of colony-bearing seeds. ANOVA y Tukey test were applied to evaluate varieties performance. Total incidence: 35% B, was significantly different from A (25%) and C (28%). Pathogens identified that could also develop in the field: a) reduce the viability and vigor: *Colletotrichum* spp., *Fusarium* spp., b) not known precise effect: *Alternaria* spp, *Bipolaris* spp, *Stemphylium* spp, c) relate to damage: *Cladosporium* spp, *Chaetomium* spp., *Nigrospora* spp d) inadequate storage conditions: *Aspergillus* spp, and e) failures in germination and decay: *Bacillus subtilis*.

87.

BV25 - DISCRIMINANT ANALYSIS: CLUSTERING AND CLASSIFICATION OF MAIZE INBREDS BASED ON TRAITS ASSOCIATED TO RIO CUARTO DISEASE RESISTANCE

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Discriminant analysis is used for predictive purposes to classify new observations into previously known groups according to phenotypic traits. In addition, it allows identifying the best traits explaining differences among groups. The objective of this work was to obtain a classification function based on phenotypic traits associated to Río Cuarto disease (MRC) resistance. A set of 77 maize inbreds with known origin was evaluated in natural infection for traits associated to MRC disease resistance including plant height, presence and type of enations, leaf length and width, leaf edge, ear curvature, proportion of kernel in ear, and amount of multiple ears. According to the parental origin, the genotypes were defined in three groups (A, B and C). Canonical axis 1 explained 67% of the variation between groups. The discriminant function indicates that the proportion of grains in ear was the most important trait for discrimination on the first canonical axis. With regard to the clustering, 57% of the lines were classified in group A, while the remaining ones were proportionally distributed among groups B and C. Each group showed an acceptable error rate with a total error of 15%. Ten new genotypes of unknown origin were classified by this function into the already established groups maintaining the same total error rate.

88.

BV29 - UVC LIGHT IRRADIATION EFFECT ON GRAPE BERRIES ANTHOCYANINS AND TRANS-RESVERATROL CONTENT

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Anthocyanins (ANT) and *trans*-resveratrol (TR) are grape phenolic compounds which contribute to its nutraceutical properties, being very important for wine industry. UVC light irradiation increases these polyphenols depending on the cv. and the given conditions. The aim of this work was to identify the best treatment conditions of UVC light irradiation on harvested berries of Cabernet Sauvignon (CS), Tempranillo (T), and Malbec (M) in order to increase ANT and TR contents. Grape berries were irradiated with 240 W at 20 and 40 cm from the light source, for 30, 60 and 120 seconds. Polyphenols extracts were analyzed by HPLC. UVC irradiation modified ANT profile of the three cultivars. The treatment for 30s at 40 cm on T increased the 3'4'5'-OH ANT over the 3'4'-OH and the methylated over the non methylated ones. These changes on the ANT profile switch berries tonality to bluish colors due to F3'5'H enzyme stimulation. Methylation confers more stability to ANT, being this appreciated to preserve color during wine aging. On CS the treatment for 120s at 40 cm decreased these ratios, leading to reddish colors and not such stability on the ANT. TR was increased by 30s 40 cm. The ratios on M were not affected.

89.

BV30 - GRAIN STRAINS IDENTIFICATION IN TWO DIFFERENT TRITICALE SETS

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Triticale grain (*X Triticosecale* Wittmack) is suitable for feed manufacture of cattle, pigs and poultry. Selection of grain material at early stages of a breeding program should take into account their behavior in various environments (localities and years). Two groups of lines introduced from the CIMMYT, and 3 triticale checks (Eronga-CIMMYT, Quiñé-UNRC and Tizné-UNRC) were compared through augmented design in Río Cuarto, Córdoba (2008-2010) and Santa Rosa, La Pampa (2008). Graphical analysis of the effect genotype + genotype x environment interaction (GGE biplot) was used. The analyzed characters were: days to flowering, growth habit, height (cm), yield (g/m²), 1000-grains weight (g) and test weight (kg/hL). Strains presented short or intermediate-short life cycle, 84.5 ± 13.2 cm height (RV = 75.5-93.0 cm) and erect or semierect growth habit. Strains overcome the checks in yield and 1000-grains weight. Several strains were grouped near the best check (Eronga) in test weight. The GGE biplot of these last characters did not allow to grouping materials according to its origin. Thus, the top 15 strains in all environments for each productive trait were identified. Four strains with good behavior in yield, 1000-grains weight and test weight, three in yield and 1000-grains weight, two in yield and test weight and one in 1000-grains weight and test weight were chosen. This group of strains will be included in comparative yield trials to analyze their behavior in different years and localities.

90.

BV31 - FORMS OF POLLINATION AND SEED PRODUCTION IN TRITICALE

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Triticale (*X Triticosecale* Wittmack) is a duplicate hybrid between autogamous wheat and allogamous rye. It is considered an autogamous crop but it is likely that the influence of parental rye result in some degree of outcrossing. In this study different kinds of pollination were analyzed in Tizné-UNRC, Yagán-INTA, Don Santiago-INTA and Eronga-CIMMYT. Three sowing dates and a randomized block design were used at 2009. The treatments were: natural pollination, forced selfing and induced cross-pollination. The number of seeds per spike, the relation between grains per spikelet and 1000-grain weight was considered. The results were analyzed through Kruskal-Wallis non-parametric method. In Tizné and Eronga, natural pollination and forced selfing did not affect significantly the number of seeds per spike and the relation grains per spikelet, but the induced cross-pollination reduced significantly the number of seeds per spike ($H_{Tizné} = 27,84^{***}$; $H_{Eronga} = 12,41^{**}$) and the relation grains per spikelet ($H_{Tizné} = 24,68^{***}$; $H_{Eronga} = 14,67^{***}$). In the other cultivars, Yagán and Don Santiago, the differences among treatments were non-significant. Regarding 1000-grain weight, the differences among treatments were significant in all cultivars and the treatments natural pollination (25,36±8,34 grains) and forced selfing (21,73±7,36 grains) exceeded significantly the induced cross-pollination (5,94±2,45 grains). These results suggest that outcrossing rate is negligible in the formation of triticale grain.

91.

BV32 - TRITICALE AS COVER CROP IN RÍO CUARTO

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Cover crops are an alternative technology that promotes water use efficiency while improving soil physical properties and fertility. In this study we examined the total production of green and dry biomass, your partition and roots depth in two triticale cultivars: Cayú and Tizné-UNRC, under 2 hidric situations and 2 stages of the production cycle. Sowing was 15/03/2010, in a RCBD with 3 replications. The data were analyzed using ANVA and Duncan test. The interactions were not significant for all traits analyzed. The total green biomass weight was 5276.5 ± 1646.4 kg/ha with significant differences for the situation with supplementary irrigation ($p=0.0023$), whereas the total dry biomass weight was 1751.1 ± 818.8 kg/ha with significant differences in favor of the situation with supplementary irrigation ($p=0.001$) and the late cut in milk-dough grain ($p<0.0001$). In relation to biomass partitioning, the average percentages of each component were: 19.7% leaf, 33.8% stem, 26.6% spike and 33.8% root. The highest green and dry weight of leaf were obtained prior to cutting heading stage ($p<0.0001$), while the dry weight of stem and root was higher in the stage of milk-dough grain ($p<0,0001$). Extra irrigation significantly increased green and dry weight of stems, spikes and roots. The depth exploration of the roots was greater for the irrigation situation. The essay provides preliminary information to determine that triticale is an alternative cover crop in the semiarid-subhumid Pampas.

92.

BV33 - EFFECTS OF SALINITY ON WATER STATUS AND OSMOTIC ADJUSTMENT IN THREE NATIVE SPECIES AND AN EXOTIC ONE OF *Atriplex* spp.

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Halophytic plants are able to accumulate inorganic ions inside their cells, adjusting their osmotic potential and ensuring water input in saline soils. The objective was to evaluate the water status of three native species: *Atriplex lampa*, *A. crenatifolia* and *A. argentina* and an exotic one: *A. nummularia* under saline conditions. Seedlings of uniform height were used, grown in a greenhouse and watered with Hoagland solution. Saline stress was induced with 1, 2 and 4% NaCl solutions and distilled water was used as control. The design was at random with six replications. Stem water potential (Ψ_h) at noon, osmotic potential (Ψ_o), relative water content (RWC) and osmotic adjustment (OA) were registered. The data were analyzed using two-way ANOVA and Tukey test for separation of mean. Significant differences between treatments and species for Ψ_o and Ψ_h were observed. These data decreased progressively in the four species as salinity of the medium was increased, resulting more negatively for *A. nummularia* and *A. argentina*. The RWC did not vary between treatments, but was significant between species. Treated seedlings adequated their potentials Ψ_h and Ψ_o and made greater AO by increasing salinity. These mechanisms would allow them to tolerate stressful conditions and survive in arid and saline environments.

93.

BV34 - EVALUATION OF NATURAL PEANUT NODULAR SENEESCENCE

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In nodules of some legumes it has been demonstrated that during the natural senescence process the nitrogen fixing and the antioxidant defense system activities decreased, while the active species of oxygen and the oxidation of leghemoglobin increase, with the consequent damage of biomolecules. There is little information about nodular senescence in peanut (*Arachis hypogaea* L.). The aim of this study was to determine the time of senescence occurrence in peanut nodules. Disinfected seeds were inoculated with 3 ml of *Bradyrhizobium* sp SEMIA 6144 culture (10^8 cells mL⁻¹). Nodules were collected at 25, 60 and 80 days post inoculation (dpi) and determined: a) percentage of red nodules, b) quantitative and qualitative analysis of H₂O₂, c) total soluble protein and leghemoglobin (Lb) contents. The results showed that, at 25 dpi, 100% of nodules analyzed were red, 54% at 60 dpi and 39% at 80 dpi. The content of H₂O₂ decreased slightly but significantly at 60 and 80 dpi compared to 25 dpi. Similar results were obtained from the histochemical detection. The content of total soluble proteins decreased significantly in function of time. At 25 dpi, the Lb content was 1.02 mg g⁻¹ fresh weight nodule. Based on these results we can infer that, although the natural senescence in the nodules of peanut seems to start at 80 dpi (similar to what occurs in other legumes), this process would not be accompanied by an increase in the H₂O₂ content.

94.

BV35 - REORIENTATION OF LEAVES OF *Larrea cuneifolia* Cav.

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Paraheliotropism is a phototropic movement that generates changes in leaf orientation in response to the incidence of sun light. It is associated with hydric stress and arid environments. *Larrea cuneifolia* has leaves with its leaf lamina parallel to incident light rays at midday. We propose that if the orientation of *L. cuneifolia* is a reversible phenomenon attributable to paraheliotropism, mechanically disoriented leaves will return to their natural orientation. Plants of *L. cuneifolia* (n=70) were distributed equally between the following treatments: frond bent with a press to the east (FBE), frond bent to the west (FBW), turned 90° from north to east (T90NE), turned 90° from north to west (T90NW), turned 180° (T180) and two controls: with a press (C+) and without press (C-). Orientation of the leaves was statistically evaluated at three periods of time (day 0, day 7 and the last measure, day 155). Angles (n=3 from each plant) from North were taken with a compass. The experiment took place at the National Park Sierra de las Quijadas, (32°47'S 67°10'O). T90NE and T90NW, reoriented their leaves to 17° from north at day 155 (P < 0.01). *L. cuneifolia*, show a response compatible with paraheliotropism. It is unknown though whether the observed phenomenon is linked to factors such as temperature, water stress or both.

95.

BV36 - HPLC & FTIR PROFILES OF *Melissa officinalis* AND *Nepeta cataria* (LAMIACEAE) OF ARGENTINEAN MARKETPopovich M¹, Gette M¹, Camí G², Saidman E³, Aragón L³, Petenatti M¹, Petenatti E¹, Del Vitto L¹.¹Proj.22/Q-016; ²Quím. Inorg.; ³Lab. C.C. Medic. UN San Luis.

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Melissa officinalis L. ("melisa" or "toronjil") is a perennial, aromatic herb whose tops (especially the leaves) are applicable as a carminative, digestive and sedative both simple as associated with other herbs. For its part, *Nepeta cataria* L. ("catmint" or "catnip"), by similar properties, often replaces it on the national herb market although chemical composition is not entirely coincidental. Therefore, it is required the setting of some parameters for diacritical differentiation at the level of crude drug, mixtures and extracts. Profiles were obtained with a "Gilson" HPLC with DAD detector and a "Nicolet Protégé 460" FTIR spectrometer with a CsI beamsplitter, from methanol and aqueous lyophilized extracts. Voucher specimens are preserved at Herbarium UNSL. Were detected interspecific, qualitative and semi-quantitative differences in various parameters (retention times, higher frequency bands, modes of vibration, stretching, CO, areas under the curves), even in the case of multi-molecular complex systems like these. The comparison of the spectra allows differentiation between samples of both species, demonstrating the usefulness of these methods to find additional diacritical characters; so becomes an important support for traditional pharmacobotanic and pharmacognostic well-established methods.

96.

BV37 - EFFECT OF WATER STRESS ON STOMATAL DENSITY AND TRANSPIRATION IN MAIZEMontoro A¹, Ruiz M^{1,2}.¹Unidad Integrada INTA-UNSJ. ²EEA San Juan INTA. E-mail: moruiz@sanjuan.inta.gov.ar

The most important factor limiting productivity in many areas of the world is the lack of water. The objective of this study was to evaluate the effects of water stress, applied at two points in the crop cycle, on stomatal density and transpiration rate in commercial maize hybrids and its relation to yield. Seeding was held in EEA INTA San Juan. Three treatments irrigation were applied, control 100% of ETc, stress at flowering (EF) 25% of ETc 15 days before and 15 days after flowering, and filling stress (EL) 25% of ETc 15 days after flowering. Was evaluated leaf area, stomatal density, total number of stomata on both sides of the leaf, stomatal conductance and yield in Kg. EF decreased yields between 50 and 90%, EL did it by 50%. The treatments influenced leaf area, transpiration rate and stomatal density. The first two variables decreased in F while the stomatal density increased. The total number of stomata per leaf was not changed by the irrigation treatments only showed differences between hybrids. Hybrids with more stomata yielded more under the stress condition but less in control.

97.

BV38 - EFFECT OF TEMPERATURE, WATER AND SOIL MICRO-BIOLOGICAL ACTIVITY ON THE LOSS OF MONOTERPENES FROM *Tagetes minuta* L. FRUITSLauret CM¹, Zunino MP¹, Dambolena S¹, Zygadlo JA¹, López ML².
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Evaporation, leaching and biotransformation by soil microorganisms are known pathways of allelochemicals release. The influence of physico-chemical properties of monoterpenes and soil microbiological activity was evaluated on the monoterpenes loss from the essential oil of *Tagetes minuta* L. (Asteraceae) fruits. Hydrocarbon monoterpenes were lost faster than oxygenated monoterpenes under the effect of temperature. In addition, the loss of each compound in the oil occurred in a similar fashion than the expected according to their vapour pressures. In another experiment, the leaching of each compound from fruits by water was slightly different according to their solubility values since tagetone, ocimenes (*cis-trans*) and dihydrotagetone showed an altered order. Finally, soil microbiological activity had a pronounced effect on the content of monoterpenes. Under microbiological active soil treatment fruit material showed decreased contents of monoterpenes, while under sterile soil treatment the contents remained constant. The results suggest that physico-chemical properties and soil microbiological activity are factors influencing the loss of monoterpenes from plant material.

98.

BV39 - LOSS OF MONOTERPENES FROM *Tagetes minuta* L. FRUITS AT EARLY DECOMPOSITION PROCESSLauret CM¹, Zunino MP¹, Dambolena S¹, Zygadlo JA¹, López ML².
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Monoterpenes are secondary metabolites with recognized eco-physiological functions as anti-herbivory, allelopathic and modulators of nutrient cycling. No information is available on the pattern of monoterpenes loss at early decomposition process of *Tagetes minuta* (Asteraceae), a species with presumably important chemoeological activity. We conducted a litterbag study during a short period of time (35 days) in order to detect the loss of monoterpenes from the essential oil stored in the fruits by means of GC studies. The overall decrease of essential oil content was 59% in contrast to a low decrease of biomass (11.4%). Loss of oxygenated monoterpenes from fruit litter was higher (58.8%) than hydrocarbon monoterpenes (54.3%). Major losses of compounds were registered for β -ocimene (60.1%), (E)-ocimene (62.6%), (Z)-ocimene (59%), dihydrotagetone (55.1%) and (Z)-tagetone (57.2%). The pattern of loss for each compound adjusted significantly to linear regressions.

99.

BV40 - EFFECTS OF INOCULATION WITH *Pseudomonas fluorescens*, TUNGSTATE AND DIPHENYLAMINE IN *Arabidopsis thaliana* PLANTSCohen AC, Salvadores C, Bottini R, Piccoli PN.
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Pseudomonas sp. are well-studied group of bacteria that promote plant growth (PGPR). In a previous study *P. fluorescens* was isolated from roots of *Vitis vinifera* (cv. malbec L.) and produce the plant hormone abscisic acid (ABA) in chemical-defined media. In addition, when incorporated diphenylamine (DPA, phytoene desaturase inhibitor) or sodium tungstate (W, an inhibitor of ABA-aldehyde oxidase, with the consequent inhibition of synthesis of ABA in plants), production of ABA in culture media decreased. The aim of this study was to evaluate the effect of inoculation of *P. fluorescens* and the application of W and DPA in *A. thaliana* plants (C). Experiments were carried out on plates with MS modified with 12 h light at 22°C. At 15 days the inhibitors were applied and 3 days later was inoculated with *P. fluorescens* (I). I stimulated leaf area, number of lateral roots, fresh and dry weight of both aerial part and roots. W increased the aerial part. However when applied W + I together, the effect was similar. W increased lateral root length, increasing the fresh and dry weight. W + I further increased the number of lateral roots. The application of DPA only altered the morphology of the root being more branched than C. However, the combination of DPA I was not effective.

100.

CL3 - ESTROGEN AND PROGESTERONE LEVELS IN THE SALIVA OF PREGNANT ADOLESCENTS DURING THE FIRST THREE MONTHS IN RELATION TO GINGIVAL DISEASETosti SB, Baudo J, Cecho A, Domínguez G, Di Salvi N.
FOLP UNLP.

During pregnancy changes in buccal cavity appear. The hormonal changes in this period include an increase of serum levels of Estrogens and Progesterone. The objectives of this study is to determine the relationship between the seriousness of gingival disease and the gestational hormone concentration in saliva. Method and Materials: The research was made over a sample of 30 pregnant adolescents aged between 14 and 19. They were previously trained in an exhaustive buccal cleaning and feeding habits. The hormone determination in saliva samples was achieved by the radioimmunoassay method. The bleeding Mulheman Index was used for the determination of gingival disease degree. Results: Estradiol levels founded in saliva were from 12 to 18 pg/ml and Progesterone from 14 to 18 ng/ml. There was observed a slight edema in marginal gum and intertooth papillae in incisive and molar zones without bleeding in catheterism (0 degree index) in 12 adolescents (40%), and bleeding in catheterism with significant edema (1 degree index) in 18 samples (60%). Conclusions: in the absence of local irritant factor, with an exhaustive buccal cleaning and a correct diet, the gingival inflammation is related to an increase in levels of Estrogens and Progesterone in the saliva.

101.**CL4 - USEFULNESS OF INDIRECT INFLAMMATORY PARAMETERS TO COMPARE TWO INHALED TREATMENTS IN MODERATE ASTHMATICS WITHOUT EXACERBATING BACTERIA**

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Forty six moderate asthmatic subject were treated with fluticasone and salmeterol (group I; n= 22) or ciclesonide and salbutamol PRN (group II; n= 24). Forced spirometry and Impulse Oscillometry were performed to evaluate the ventilator function. Hypertonic saline-induced sputum was studied, looking for etiological agents of asthma exacerbation. Relationship between bacterial infection and number of exacerbations, fungal colonization of the oropharynx and quality of life using ACCT inquiry, were also studied. The ventilatory parameters FEV1 and FVC and impulse oscillometry were within the expected values in both groups. All subjects showed bronchial reversibility >12% and >200ml after 400 µg of salbutamol. In spite of the isolation of *B. catarrhalis*, *S. pneumoniae* and *H. influenzae* in the sputum of both groups, asthma exacerbation was not present during the entire study. Fungal colonization, due to *C. albicans*, ranged between 38% and 42%. There was a significant decrease in eosinophil counts in the two treatments (p>0.001). ACCT was of 25 to 15 with SF and 20 to 15 using C. Results pointed usefulness of indirect evaluation of inflammation parameters to compare different treatments in asthmatics patients.

102.**CL6 - RISK FACTORS IN RENAL STONE FORMATION IN PRE-MENO- POSTMENOPAUSAL WOMEN**

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Physiological disturbances as menopause may create an environment conducive to renal stone formation. The aim was to evaluate metabolic abnormalities in renal lithiasis in pre-meno-post menopausal women. 153 patients with nephrolithiasis were classified: I) Premenopausal: 35-44 yr, n=55; II) Menopausal: 45-54 yr, n=64; III) Postmenopausal: 55-65 yr, n=34. The diet hypocalcic-hyposodic/ 7 days with Calcium 1000mg/day was administered. Urine (24h, 2h) and blood were recollected. Calcium, phosphorus, creatinine, magnesium, alkaline, urea, uric acid, phosphatase, citric and oxalic acid, parathormone, sodium y potasium (fotometric test) were determined. Body mass index (OMS) I vs II, III (p<0.04). Phosphatemia increased I vs II (p<0.036), alkaline phosphatase increased I vs II,III (p<0.01); positive correlation uric acid vs years old ($r=+0.218, p<0.0001$). Urine 24h: phsphorus and creatinine I vs II,III (p<0.001), creatinine clearance I vs II,III (p<0.003); uric acid clearance I,II vs III (p<0.005); citrate/creatinine I vs III (p<0.001), calcium/weight I vs III (p<0.04), calcium/citrate I vs III (p<0.0001). Urine 2h: phosphorus clearance I vs II, III (p< 0.001); phosphate tubular reabsorption I, II vs III (p< 0.04); phosphate renal umbral I vs III (p<0.04); hypocitraturia (p<0,021; VdeK= 0,225). Low urine volume was found in premenopausal women (p<0.002). Our results show the importance of studying nephrolithiasis patients by biochemical parameters to achieve a diagnosis of the metabolic abnormality and introduce a specific therapy to prevent recurrence.

103.**CL7 - DIET AND GALLSTONES RISK. PRELIMINARY RESULTS**

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A case-control study is under development in Rosario, in order to analyze diet influence on gallstones risk. So far, data from 40 cases and 42 controls were collected. Cases are subjects diagnosed with gallstones, some of which have been cholecystectomized. Controls underwent an abdominal ultrasound to rule out the asymptomatic gallstones presence. All participants underwent a personal interview. food consumption, five or more years before diagnosis for cases, and habitual diet for controls were investigated. Odds Ratios (OR) and average food consumption differences between cases and controls, were evaluated. Mean total energy intake, and fatty foods consumption were significantly higher, and dairy products, fruits and nuts were significantly lower in cases. After adjusting for total energy intake, significant differences for vegetables consumption was also found. Gallstones risk (adjusted for total energy intake, age and gender) increases by 11 and 21% for every gram increase in cold meats (OR = 1.11, p= 0.050) and fatty foods consumption (OR = 1.21, p = 0.030), respectively. Furthermore, gallstones risk decreases by 69% (OR = 0.31, p = 0.012) for each extra gram of nuts intake. In conclusion, meats and fatty foods intake increase the risk of developing gallstones, and nuts consumption acts as a protective factor.

104.**CL8 - ESTROGEN AND PROGESTERONE LEVELS IN THE SALIVA OF PREGNANT AND NON PREGNANT ADOLESCENTS**

Tosti SB, Baudo J, Cecho A, Domínguez G, Di Salvi N.

FOLP UNLP.

Adolescence, pregnancy and menopause changes in female bodies are associated with Estrogen and Progesterone levels. These hormones were studied in serum and saliva. At these stages of female life the most common changes in mouth cavity are showed in gingival tissue with different degrees of inflammation. The objective of this research is to determine the levels of female sexual hormones in the saliva of pregnant and no pregnant adolescents. Method and Materials: the sample comprise 60 adolescents aged between 14 and 19 years, 30 pregnant and 30 non pregnant. Before taking the sample there was a pause of 2 to 15 minutes. Stimulation of spitting was avoided to preserve possible crossing reaction with the hormones in research. Hormones were measured by Radioinmunoassay. The saliva sample was kept at room temperature and a biocide was added to prevent contamination and/or bacterial growth. Results: Estrogen levels in the saliva were from 5 to 10 pg/ml in non pregnant and from 8 to 17 pg/ml, in pregnant adolescents. The levels of Progesterone were from 10 to 34 ng/ml in non pregnant and 15 to 48 ng/ml in pregnant. Conclusion: The levels of sexual hormones in the saliva show an increase in pregnant adolescents. The question is if this increase induced higher seriousness of gingival disease.

105.

CL10 - WIDE STANDARD FOR TECHNICAL ASSESSMENT OF THE SECTOR SINUS BONE

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Oral Implantology Dentistry is the science that deals with the replacement of missing teeth by artificial substitutes. The goal is to standardize the technique of obtaining panoramic radiographs, to acquire the degree of distortion in the field of craniofacial sinus compared to bone *seco*. The primary method of choice for this purpose is the panoramic radiography, diagnostic method that allows the global observation of all structures surrounding the jaws. The reading and interpretation of imagery is critical to the success of treatment. Since the image obtained in the relationship between the focus-object distances and object-film is not the same at all points, they show different magnification and distortion factors, which hinder the proper relationship between the realization of structures anatómicas. For work were used 50 skulls obtained from Osteoteca Faculty of Dentistry, UNLP. They were numbered with Arabic numerals. Panoramic radiographs were obtained in the radiology Course Faculty of Dentistry, UNLP were used for measurements of optical magnification element graph type and size Vernier. The results demonstrate the existence of a statistically significant magnification for this sector of the massif facial. It can be concluded that given the magnitude of the distortion is found necessary to use some kind of "tutor" or known as reference in order to calculate arithmetically the real measure of analyte.

106.

CL11 - SMOKING. STUDY OF SALIVARY BIOCHEMICAL PARAMETERS. IMPACT ON ORAL HEALTH

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The oral health status depends on several factors. Direct action of snuff is associated with oral diseases. Determine pH, salivary flow, concentration of total protein (TP) and immunoglobulin A and G in saliva of smokers (F) and nonsmokers (NF). Set Gingival, Plaque, CPTIN and CPO Indexes. Link alterations with possible oral pathology. Saliva samples from healthy volunteers, students of 1st grade of Odontology 18-25 years, F and NF were collected spontaneously. pH was determined with test strips, saliva flow in ml/min; TP by the Lowry method, IgAs and IgG by quantitative IDR. The indexes were determined by clinical examination. No significant differences were found in pH, salivary flow, IgAs and indexes between groups F and NF. There were no detectable levels of IgG in any of the groups analyzed. The habit was significantly more frequent in women and in older subjects. The TP concentration was significantly lower in F compared to NF (81.11±8.53 vs 112.84±9.45* respectively. *P<0.05 were considered statistically significant). Our results indicate that no significant differences found in the analyzed parameters would be related to the low consumption of snuff in the population studied. The lowest values of TP in F, as opposed to IgAs values similar to the NF group leads to consider the possibility of decreased of other salivary proteins such as enzyme. We plan to analyze other components involved in the immune response of the oral cavity associated with smoking.

107.

CL13 - DETECTION OF *Escherichia coli* SHIGA TOXIN-PRODUCING (STEC) IN IRRIGATION CANALS NEAR CULTIVATED AREAS OF THE MENDOZA RIVER

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Among the new emerging pathogens are *Escherichia coli* Shiga toxin-producer and linked with Hemolytic Uremic Syndrome (HUS). The aims of this study were: 1) To establish the frequency of *E. coli* O157: H7 and non-producing *E. coli* O157 STEC in the waters of irrigation canals of the Mendoza River. 2) Detect STEC in irrigation canals near areas of cultivation. 3) To study *E. coli* as an indicator of fecal contamination 4) To relate this species with total coliforms. We investigated 50 samples from the northern oasis of Mendoza River. The bacterium was detected by swabs Moore and Immuno-magnetic beads and the toxin by PCR. Suspicious colonies of *E. coli* O157: H7 were isolated from Cromoagar O157 and sorbitol-MacConkey agar with cefixime-tellurite. The count of total and fecal coliforms was performed by method of most probable number (MPN). The total coliform count was exceeded in 4 samples, while only 8 had no coliforms. In populated areas close to load garbage into waterways, it was high coliform counts and the presence of *E. coli*, but the rest of the samples these counts remained within the limits established. Using the PCR we observed 2 (two) positive samples with presence of Shiga toxin.

108.

CL16 - ASSOCIATION OF TUBULIN TO THE MEMBRANE: IMPACT ON DIABETES AND HYPERTENSION

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Acetylated tubulin is associated with the plasma membrane through binding to P-ATPase modifying the activity of enzymes. In our laboratory we have shown that in erythrocytes of hypertensive and diabetic patients tubulin membrane is increased by more than twice a migration from a sedimentable structure. This increase of membrane tubulin decreases the deformability of erythrocytes and inhibits the activity Na⁺,K⁺-ATPase. The effect of tubulin on the P-ATPase activity is dependent on membrane lipidic composition. In fact, in the presence of basic or neutral lipids the enzyme activity is inhibited, but in the presence of acidic lipids is increased to 27 times. Based on this background was proposed for this work to study the mechanism of translocation of tubulin to the membrane and its effect on ATPase activity. In this paper we show that: 1.- the glucose and increased content of microtubules produce migration of tubulin to the membrane in synergy, through a mechanism that involves the participation of aldose reductase, 2.- glucose causes the deacetylation of tubulin and promotes their migration to the plasma membrane probably due to the dynamism of the microtubule, 3.- erythrocytes of diabetic patients or normal subjects treated with high concentration of glucose found a fraction of tubulin sediments with lower sedimentation coefficient to that found in erythrocytes of control subjects or hypertensive. These results allow us to speculate that the migration of cytoplasmic tubulin to the membrane, in addition to the acetylation of tubulin, it is necessary a change in the dynamics of microtubules. Both diabetes and hypertension are the elements that contribute to the modification of microtubule dynamics, the first high concentrations of glucose and the second modification of different microtubule-associated proteins (MAPs) as previously reported.

109.

CL17 - PREVALENCE OF *Enterobius vermicularis* (NEMATODA: RHABDITIDAE) IN ELEMENTARY SCHOOL OF CORDOBA

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Enterobius vermicularis causes enterobiosis, one of the most frequent intestinal parasitic infections in the world and affecting mainly children. In view of the scanty knowledge on the rate of parasitism in the province of Cordoba, it considered to know the prevalence of *E. vermicularis* in a rural elementary school of Rio Ceballos. The samples of the 37 childrens from 4 to 11 years old were taken by their parents, using Graham's method. It also applied a survey asking about health habits and symptoms of each child. To prove the association between each of the variables recorded and frequency of individuals infected with *E. vermicularis* was used Chi-square (χ^2) test. The relationship between parasitism and age of the individuals was assessed by Student's test. Also were calculated the Spearman Correlation Coefficients. Prevalence of *E. vermicularis* in perianal samples was 48.65%. There was a relationship between parasitism and age of children ($t = -2.72$; $gl = 42$; $P = 0.0094$), not this way with the sex ($\chi^2 = 0.05$; $gl = 1$; $P = 0.8241$) and the grade ($\chi^2 = 0.51$; $gl = 2$; $P = 0.7763$). The most frequent symptoms were itching anal, occurred in 66 % of infected children. The prevalence of *E. vermicularis* was high, higher than the found in a school boarding in Unquillo, Cordoba and than found in other studies in rural and urban areas of Buenos Aires, Salta and Corrientes, and was below that the prevalence found in Mendoza and Tucumán. In Argentina, as in most Latin American countries, prevalence rates vary.

110.

EB4- FOOD COMPOSITION AND CULICIDAE LARVAE PREFERENCE OF TWO INDIGENOUS FISHES IN MESOCOSM EXPERIENCES

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Food preference for one target is a trait characteristic to assess the suitability of a species for biological control. Food composition and culicidae larvae preference of *Jenynsia multidentata* and *Cnesterodon decemmaculatus* were analyzed in mesocosm experiences to determine the relative preference for mosquito larvae. Six enclosures of 22 cm in diameter were placed in pools of 80 x 450 cm of standing water containing great quantity and variety of aquatic invertebrates, including mosquito larvae. Five individuals of each species were placed in each enclosure. The individual fishes were starved for a period of 72 hs before experiments. Experiences lasted 2 hours. Cladocerans were eaten most by both species, followed by culicidae larvae for *J. multidentata* and chironomidae larvae for *C. decemmaculatus*. *J. multidentata* present the higher food items richness and absolute numerosity and frequency of occurrence for culicidae larvae, that will be indicating high suitability of this species for biocontrol. Even though, its greater preference for cladocerans could make us doubt of its capacity for mosquito control. The short lasting of experiences could be also underestimating *C. decemmaculatus* biocontrol skills, as it's been shown in others studies.

111.

EB6 - POPULATIONS OF *Echinopsis leucantha* (Cactaceae) IN THE CENTRAL WEST OF ARGENTINA

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This study seeks to gain knowledge of cactaceae populations to aid in their management. A comparative analysis is conducted between *Echinopsis leucantha* populations in areas with and without water erosion, located in the central west of Mendoza province, Argentina. Density of this cactus was 2.25 times higher in the eroded than in the non-eroded area. The greatest number of small individuals, up to 10 cm in diameter, occurs in the eroded area, and the greatest number of adults in the non-eroded area. In all cases, *Echinopsis* plants are generally exposed on their north-facing side, and protected under shrub canopies. The highest number of *Echinopsis leucantha* in the non-eroded area is found beneath *Larrea cuneifolia* and, in the eroded area, beneath *Larrea divaricata*, under whose canopy highly significant values are reached in terms of protective cover, humidity, and lower radiation and temperature, whereas soil conditions regarding organic matter and mineral nutrients are practically non significant.

112.

EB7 - DISTURBANCE EFFECTS ON ARID GROUNDWATER COUPLED ECOSYSTEMS (MENDOZA)

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Shallow groundwater is an important resource for plants and local people in drylands. The modification of the disturbance regime by different land uses can alter several aspects of ecosystems functioning. We evaluated the effects of traditional livestock posts, generally consisting of a housing unit, corrals, a water well, and domestic animals, on vegetation, water and nitrate dynamics in the Monte desert (Mendoza). We postulated that the removal of the vegetation in livestock posts and the accumulation of urine and dung allow water percolation, facilitating nitrate leaching and local groundwater recharge. We analyzed nitrate and water dynamics mediated by animals and vegetation in three livestock areas and their nearby relatively undisturbed *Prosopis flexuosa* woodlands. We found higher water and nitrate, and lower chloride contents in soil profiles obtained in livestock concentration areas, where bare soil and dung densities were higher. Low soil chlorides indicate that water is percolated to the subsoil and into the groundwater, transporting nitrate to the aquifer. We also found other indicators of human impacts that may be useful to evaluate degradation, such as lower soil crust cover, standing dead wood, grass cover, and *P. flexuosa* seeds. Our results show that human activity, even as traditional and extensive livestock production, affects the interactions among water, vegetation, and nitrate, facilitating the movement of water and solutes to the aquifer.

113.

EB9 - CONTRIBUTION OF EARTHWORMS (ANNELIDA: OLIGOCHAETA) TO ECOSYSTEM PROCESS IN AGRICULTURAL SOILS*Vaquero F, Bedano JC, Becker A.**Departamento de Geología, Universidad Nacional de Río Cuarto.**E-mail: vaquero.flor@gmail.com*

Soil resource is critical for agricultural production and food sovereignty. Earthworms have great influence on soil structure (aggregate and pore formation) and organic matter decomposition (fragmentation, bury and mixing of vegetal debris). Some agricultural practices such as tillage, that needs supplies (biocides) and fertilization, reduces biodiversity and ecosystem services provision. In the Argentinean Humid Pampa there are few studies about these issues. The aim of this study is to evaluate the influence of three agricultural practice systems on earthworm's aggregate production, as an indicator of earthworm contribution to the ecosystem services in soils of central-east Córdoba. Three systems were analyzed: 1- No-till, intensive crop rotation, reposition fertilization, minimal chemical inputs, over 30 years (SD-RyF-30); 2- same management, over 12 years (SD-RyF-12); 3- No-till, with soya monoculture (SD-MC) and no fertilization. Number and weight of biogenic aggregates were registered. Both variables were significantly higher in SD-RyF-30, followed by SD-RyF-12 and last SD-MC. We conclude that practices that include crop rotation, fertilization and low chemical inputs are beneficial for earthworms activity and therefore for their contribution to the associated ecosystem services. Besides, treatment duration was also a determinant factor.

114.

EB11 - VASCULAR FLORA OF "MOGOTE BAYO" NATURAL RESERVE (COMECHINGONES SIERRA, SAN LUIS, ARGENTINA)*Del Vitto LA, Petenatti EM, Petenatti ME, Rodríguez N.**Herbarium UNSL/ Proj. 22/Q-016 SPU-ME, Ej. de los Andes 950, D5700HHW San Luis, Argentina. E-mail: elipete@unsl.edu.ar*

"Mogote Bayo" is an ecological reserve from about 250 ha on western slopes of Sierra de Comechingones (1,000 to 2,300 m) that has been recently added to the San Luis provincial system of protected areas. It includes typical "Chaco Serrano" plant communities, distributed in altitudinal belts, with grasslands, steppes, shrublands, and evergreen and mixed forests. The climate is temperate sub humid mountainous. Rainfall varies from 500 to 600 mm with showers and snowfall in winter. Soils are shallow, highly permeable, and poor in organic matter, strongly sloped and therefore easily eroded. There are almost 500 vascular plant species. Forest belt (up to 1,000-1,500m) shows *Aspidosperma quebracho-blanco* and *Prosopis* spp., and a single canopy on slopes with *Lithraea molleoides* and *Zanthoxylum coco*. *Minthostachys verticillata* is an understory aromatic. Shrubby belt extends above 850 m in most windy exposures, up to 1,500-1,600 m, with *Heterothalamus alienus* and *Eupatorium buniifolium* var. *buniifolium*. High meadows are present at the top of the slopes in the mountains and high plains, between 1,500 and just over 2,000 m, with *Nassella*, *Festuca* and *Stipa* species, and some herbaceous dicots. Coppices of *Polylepis australis* and *Maytenus boaria* appear in the upper area of the gorges. A short number of exotic plants have naturalized in the region.

115.

EB12 - COMPARISON OF BIODIVERSITY FORMICA BETWEEN EDGE AND CENTER PEANUT CROP*Vilches JY¹, Quirán EM¹.**¹Facultad de Ciencias Exactas y Naturales. U.N.L.Pam. Av. Uruguay 151. L6300CLB Santa Rosa, La Pampa, Argentina. E-mail: juli_vilches@hotmail.com; emquiran@yahoo.com.ar*

In recent years, in the province of La Pampa, there is a marked increase of crops, causing a proliferation of harmful species. As good control of these pests, we can find the formica, being considered one of the most groups successful in terrestrial ecosystems. Is for these characteristics which seeks protection of its biodiversity, being important for the proper functioning of the agro ecosystem. The objective of this study is to evaluate the diversity of formic, on the edge and center of the peanut crop, located in Maisonave, (64°S and 35°W) northeast of La Pampa province (Argentina). Sampling was conducted in the period December 2008 to March 2009, on plots of one hectare. Three transects were laid out, separated by 25 meters in each plot, where they were placed pitfall traps (pitfall) of a liter, at a distance of 25 m each, with a mixture of water, detergent and salt for 5 days. The material obtained was preserved in 80% alcohol, was analyzed in the laboratory and the chair was placed in Invertebrate Biology II, Faculty of Natural Sciences, the UNLPam. Statistical analysis was performed with Past (2009). The results show a dominance of *Solenopsis saevissima* species in the center of the crop.

116.

EB13 - COPEPODA IN GROUNDWATER: RESPONSE TO POLLUTANTS ASSOCIATED TO ORGANIC MATTER*Tione ML^{1,2}, Blarasin MT¹, Bedano JC^{1,2}.**¹Fac. Cs. Ex F-Q y Nat. UNRC; ²CONICET.**E-mail: ltione@exa.unrc.edu.ar*

Groundwater invertebrates, including Copepoda, have a potential value as environment quality indicators. In this work, we analyze the response of Copepoda living in a fluvio-eolic aquifer (Río Cuarto, Córdoba) in relation to groundwater geochemistry and land use features. 25 wells were sampled and major ions and dissolved organic carbon (DOC) were measured. Also, 150 litres of water were sampled in each well for the collection of invertebrates. Different land uses were taken into account. In general, it was observed that DOC was lower than 1.0 mg/L, but in 7 wells was higher (1.1-3.2 mg/L). DOC was positively and significantly correlated with total dissolved salts (TDS), HCO₃⁻, Na⁺, Cl⁻, SO₄⁼, K⁺ and NO₃⁻. Copepoda was found in 10 wells located near livestock and/or on-site sanitation systems. Copepoda was positively and significantly correlated with DOC, TDS, SO₄⁼, NO₃⁻ and Cl⁻, indicating a clear association to pollution sources. The abundance of Copepoda was highest (91 individuals/150 L) in a well close to a feed lot, with a DOC value of 2.9 mg/L. We conclude that land use influences groundwater geochemistry and the presence/abundance of copepods, which could be used as bioindicators of groundwater quality.

117.

EB14 - CHANGES IN PLANT COVER AND ATTRIBUTES ACROSS A GRAZING GRADIENT IN NORTHEASTERN CHUBUT, ARGENTINABär Lamas M¹, Larreguy C^{1,2}, Carrera A^{1,2}, Bertiller M^{1,2}.¹CENPAT-CONICET, ²UNPSJB. E-mail: barlamas@cenpat.edu.ar

In Patagonia, sheep grazing affects the architecture and dynamics of vegetation through the replacement of species with different morphologies and ecological strategies. We analyzed the variation induced by grazing on plant cover, specific leaf area (SLA) and the height of dominant species in the northeastern Chubut (Argentina). In three sites across a grazing gradient, we assessed the total and relative cover of shrubs and perennial grasses, species richness and species composition. At each site, we measured the height and the SLA in 3 adult plants of each dominant species. The site with low grazing intensity had higher total plant cover and relative contribution of perennial grasses than the other sites. Species richness decreased with grazing intensity and the similarity in species composition was higher than 68% among sites. Plant height and SLA of each species did not vary among sites. We found a general trend of decreasing SLA of perennial grasses and the height of shrubs with increasing grazing intensity. We concluded that grazing was mainly associated with changes in the relative abundance of species and/or biological forms thus affecting the attributes at community level but without modifying them at the species level.

118.

EB15 - GEOGRAPHIC VARIATION IN THE ADVERTISEMENT CALL OF *Hypsiboas cordobae* (ANURA; HYLIDAE)Baraquet M, Grenat PR, Salas NE, Martino AL,
FCEFQN-UNRC. Río Cuarto, Córdoba.

Geographic variation in the advertisement call of frogs and toads is commonly observed among conspecific populations of widespread anuran species. Some authors suggested that these signals, because of the species-specific information they convey, should show little variation within species. Most of these studies analyzed the geographic variation of mate choice signals in order to understand the role of geographic isolation or sexual selection, in the earliest stages of population divergence that might result in speciation. This paper analyzed the geographic variation of advertisement call of *Hypsiboas cordobae* in six populations, covering the area of distribution of that species. Each call was characterized by seven temporal variables and four spectral variables. Nine variables showed significant differences ($p < 0.05$) between populations when compared via ANOVA. The acoustic variables that correlate with temperature were adjusted to 14 °C. Discriminant analyses showed differences between the six populations in study ($p = 0.0003$). Then, we applied a simple regression analysis between each of the variables and latitude of each population, six of the spectral variables showed a significant positive relationship. From discriminant analysis, we calculated Mahalanobis acoustic distances averaging among the six populations, and the Mantel test was applied to estimate the correlation between geographic distance (in kilometers) and the acoustic distances (Mahalanobis), which no showed significant correlation with geographic distance ($p = 0.0635$, $r = -0.498$, $t = -1.5262$). This paper showed a clinal variation from south to north decrease in the dominant frequency of the call. Several authors attribute this change to the negative correlation between SVL and dominant frequency, so this work should be complemented by morphometric studies.

119.

EB16 - COMPARISON OF BODY CONDITION OF *Rhinella arenarum* INDIVIDUALS THAT INHABIT IN RIO CUARTO CITY

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Body condition represents an indicator of environmental stress. The objective was to determine and compare the body condition of *Rhinella arenarum* individual that inhabit sites in the city of Río Cuarto, Córdoba. We chose four sites: an urban lake, Lake Villa Dalcan, two sites of crops called Crop 1 and Culture 2 and Laguna Las Brujas, located on the UNRC. Individuals were collected with pitfall traps and visual surveys for meetings during the years 2008-2010. Each individual is recorded age class (juvenile or adult), snout-vent length (mm) and body mass (g). Body condition was calculated by the residual rate and compared between study sites by age group using the Kruskal Wallis test. Abnormalities were identified using a standard protocol. No significant differences in body condition among adults of different sites ($p = 0.89$), but if you were among between the juveniles ($p = 0.00014$). In Lake Villa Dalcan site was a high rate of morphological abnormalities in individuals (13.64%). The results allow formulating serious questions about the management of these environments and the need for permanent monitoring of them.

120.

EB19 - RELEASE CALL OF *Odontophrynus cordobae* AND *O. americanus*: INTRA AND INTERSPECIFIC DIFFERENCESGrenat PR^{1,2}, Salas NE¹, Martino AL¹.¹Ecología Dpto Cs. Naturales, Fac. Cs. Exa Fco-Qca y Nat. Ruta Nac N° 36-Km 601 Río IV, Argentina. ²Becario CONICET.

Release vocalizations are produced by male anurans in response to male mating attempts. Calls, used in exchanges between males, are more variable in form, as might be expected of communication between rivals, but overall their structure is more similar in closely related species than is that of advertisement calls. We analyzed 145 release calls from 11 individuals of *O. cordobae* and 5 of *O. americanus*. Calls were registered in laboratory conditions (20°C). Release call of the two species differed in six of the seven variables considered. Discriminant Function Analysis based on 7 acoustic parameters correctly classified 89.66% of individuals within the species. The variables with greater weight in the function obtained were pulse rate, the ratio of pulses and pulse duration. When separated the call groups within each species in syntopic and allopatric, we obtained three highly significant discriminant functions. The re-classification obtained in this case was 97.33%, and 100% of the calls of syntopic individuals of both species was classified within their respective group.

The results of this study do not support the convergent character displacement of song release reported by other authors in studies of species that coexist in sympatry. Thus the release call could mean a diagnostic character for distinguishing these two cryptic species.

121.

EB20 - MORPHOMETRIC ANALYSIS OF *Odontophrynus* CRYPTIC SPECIESGrenat PR^{1,2}, Salas NE¹, Martino AL¹.¹Ecología, FCEFQyN, UNRC. Río IV, Argentina. ²CONICET.

We analysed two morphologically cryptic species, *O. cordobae* and *O. americanus*, in order to assess the degree of sexual dimorphism, the intraspecific morphometric variation and morphological differentiation between the two species. We measured 15 morphometric variables on 211 individuals from 21 localities of Córdoba. We found sexual dimorphism in six parameters in *O. cordobae* and three variables in *O. americanus*. Diploid and tetraploid males were significantly differentiated in six morphometric variables. Discriminant Function Analysis (DFA) including all populations showed a positive classification of individuals within their respective species of 76.37%. DFA based on four groups (allopatric and syntopic of each species) showed a positive classification of individuals within their respective group of 73.45%. Allopatric individuals of each species segregate visibly, as allopatric and syntopic individuals within the corresponding species. Syntopic individuals of *O. cordobae* and *O. americanus* showed the lowest percentage of positive classification. Because the observed differences in morphometric characters in sympatry between diploid and tetraploid are no greater than those observed in allopatry, the results deviate from the expected results under the hypothesis of character displacement. These results may suggest that external morphological characters would not have a major influence on the recognition and choice of conspecific males by females.

122.

EB22 - FISH AS BIOINDICATORS THE AQUATIC SYSTEMSPollo FE¹, Salas NE¹, Mancini MA², Martino AL¹.¹Depto de Cs. Naturales, Fac. Cs. Exa Fco-Qca y Nat; ²Acuicultura Fac Agro-Vet, UNRC. E-mail: faviopollo@gmail.com

The water quality of urban lake depend, among other causes, activities that take place in the lake and its drainage basin. Often, these ecosystems become important reservoirs of xenobiotics. The aim of this study was to determine the frequency of micronuclei (MN) and nuclear abnormalities (NA) levels of fish from Villa Dálcar urban lake (33° 07' W, 64° 20' S, Río Cuarto, Argentina). The species studied were *Cyprinus carpio*, *Cheirodon interruptus* and *Astyanax eigenmanniorum*. We captured 98 specimens of three species, with values of 5,56 (± 1,42), 5,38 (± 0,26) and 5,72 (± 0,36) cm in total length. Blood samples were extracted by cutting the caudal peduncle, after anesthesia. The blood smears were fixed and subsequently stained with Giemsa and observed under a microscope with an increase of 1000X. The frequency of micronucleated erythrocytes (MN) and nuclear abnormalities (NA) was calculated on 1000 red blood cells. The results were: *C. carpio* (0.05 ± 0.12 MN; 2.12 ± 1.88 NA), *C. interruptus* (0.04 ± 0.09 MN to 5.62 ± 6.32 NA) and *A. eigenmanniorum* (0.07 ± 0.22 MN; 8.77 ± 7.46 NA). These results allow us to infer the existence of genotoxic compounds in Villa Dálcar Lake. More detailed studies should be performed with the *A. eigenmanniorum* as it outlines a priori as a good "sentinel" for the detection of xenobiotics.

123.

EB23 - HEMATOLOGICAL BIOMARKERS AND CYTOMORPHOLOGIC IN THE COMMON TOAD *Rhinella arenarum*

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The differential amount of white blood cells and cytomorphometry were studied in *R. arenarum* as biomarkers of genotoxicity in environments with different degrees of impact. The study was conducted in the pond Las Brujas (UNRC) and the urban lake Villa Dálcar, Río Cuarto, Córdoba. Ten individuals were captured per site and took samples of blood by puncture of angularis vein. The smears were stained with May-Grünwald-Giemsa and observed under a microscope. The total number of white blood cells for the pond Las Brujas was: 382.7 ± 162.7 / 10000 erythrocytes and 364 ± 312.9/ 10000 erythrocytes for Villa Dálcar. The ratio N/L (neutrophils/lymphocytes) was 0.17 for the pond Las Brujas and 0.24 for Lake Villa Dálcar. No significant differences were observed between sites for the variables for the hemogram (U test, p > 0.05), except for monocytes (p = 0.002). For the blood morphometry, no significant differences between sites (U test, p > 0.05) were found. The ratio N/L is similar to other research in *R. arenarum*. The results of this study indicate that hematological evaluation allows for early detection of physiological changes related to processes of environmental pollution

124.

EB24 - ACOUSTIC INTERFERENCE IN ANURAN CALL: STIMULUS-RESPONSE IN *Hypsiboas pulchellus* (ANURA, HYLIDAE)Salinero MC¹, Grenat PR^{1,2}, Baraquet M^{1,2}, Salas NE¹, Martino AL¹.¹Ecología, FCEFQyN, UNRC. Río IV, Argentina. ²CONICET.

Changes in vocal production of a male in response to other male calls are common, especially when the density of callers is high. In this study, we measured the vocal responses of *H. pulchellus* males to different acoustic stimuli from other males. Experimental session was conducted *in situ*. Series of experimental stimuli were emitted from a speaker located about 50-80 cm in front of each individual. To record the responses from the beginning to the end of the call, the recorder was positioned 50 cm away from the focal male. Throughout the procedure, each male was filmed to analyze their behavioral responses. Acoustic analysis showed an alteration in the song of the focal male post-stimulus relative to its pre-stimulus call, mainly by modifying the call rate. We observed a decrease or increase of the inter-call duration or an alteration in duration within the block of each call, with variations in the note duration or in the interval between notes. In all cases, focal males responded by increasing or decreasing its call rate to accompany the stimulus. The responses to those stimuli were the basis for testing the hypothesis that males of *H. pulchellus* could significantly change their pattern of calls, depending on the source of noise interference to which they are subjected.

125.

EB25 - NEW RECORDS OF *Contracaecum* sp. (NEMATODA: ANISAKIDAE) IN FISH OF ARGENTINA CENTRALMancini M¹, Guagliardo S², Tanzola D², Salinas V¹, Morra G¹, Biolé F¹.¹FAV-UNRC (Rio Cuarto). ²UNS (Bahía Blanca). E-mail: mmancini@ayv.unrc.edu.ar

The parasitic diseases caused by anisakids affect a large variety of fish and in some species produce a decrease in the body condition. The aim of this work was to identify the nematodes species in fish from three shallow lakes in central Argentina, which differ in their genesis, water quality and fish richness. Environments are located in San Luis province (L1: 33°59'W, 65°24'S, 20 ha, 4 fish species) and Córdoba province (L2: 34°46'S, 63°38'W, 190 ha, 13 species and L3: 33°48'S, 64°51'W, 420 ha, 3 species). All helminths were *Contracaecum* sp. larvae (Nematoda: Anisakidae), consistent with type 2 third stage larvae described by Moravec *et al.* (1995, *Folia Parasitologica* 42). These nematodes were present in the species *Odontesthes bonariensis*, *Hoplias malabaricus*, *Rhamdia quelen* and *Oligosarcus jenynsii*. According to our experience, *R. quelen* shows the greatest mean abundance (13.7±18.5 nematodes/fish) and prevalence (87.5%). This study confirms that only *Contracaecum* sp. was present in the three environments evaluated. Because of the findings of these parasites is continuously increasing and have been observed in other species, further studies including fish and definitive hosts (birds), should be made for a better understanding of the cycle.

126.

EB26 - THE SPECIES OF *Solanum* L. (SOLANACEAE) FROM SAN LUIS PROVINCE, ARGENTINA

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Solanaceae is a monophyletic family and in Argentina is represented by 33 genera including 328 species, being one of the best represented in argentinian flora, especially in the provinces of the centre of the country. According with previous data, 13 genera are inhabiting in San Luis province, whereas, as results of different activities in the "Estudios de la vegetación de la provincia de San Luis" investigation project, we have found 16 genera. One of them, *Solanum* L., is the best represented one. The aim of this work is to present a floristic survey and analysis of ecological aspects of the species belonging to the genus *Solanum* L., with particular emphasis in their diversity, in the provincial territory of San Luis. The analysis was done using freshly collected material in different parts of the province, and vouchers of the herbaria VMA and VMSL (University of San Luis) and RCV and RIOC (University of Rio Cuarto). The material collected during the investigation travels was incorporated to the RCV and VMA herbaria. Floristic treatments, geographical distribution and identification keys for the entities are presented. The genus is represented by 24 entities inhabiting San Luis territory, three of them (*Solanum argentinum*, *Solanum chenopodioides* and *Solanum salicifolium*) are cited for the first time to the province, whereas *Solanum concarense* and *Solanum incisum* var. *tenuisectum* are strictly endemic of San Luis province. We have not found exotic species of *Solanum*, being all species native from Argentina.

127.

EB28 - REPRODUCTIVE CYCLE OF *Pisidium chiquitanum* ITUARTE, 2001 (BIVALVIA, SPHAERIIDAE) FROM ARROYO USPALLATA, ARGENTINA

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Like the 18 other South American species of the genus, the reproductive biology of *P. chiquitanum*, a brooding and simultaneous hermaphrodite species, is unknown. The aim is to describe the annual gonadal cycle of the species. Between Oct/2007 and Sep/2008, 5-10 adults/month were collected at the outlet of Arroyo Uspallata (Mendoza, Argentina), along with records of water temperature (WT). Clams were fixed *in situ* in Bouin and processed for conventional histology (serial sections of whole animals). Only a fraction of the population showed maturation signs at the same time every month. In gonads of both sexes the maturation initiated in July and increased gradually and steadily between that month and September-November. The frequency of mature gametes began to decline in both gonads in December. Except isolated acini, mature gonads were not found between January-February and June. Preliminary results suggest an annual peak of gonadal maturation in spring driven by an increase in WT from July (13.4°C) to September (15.9°C, mean values), massive evacuations in November-December and hatching between late November and late January. The massive release of embryos would be between mid January and mid autumn, coinciding with a plateau of elevated WT (15.8 to 16°C).

128.

EB29 - GENETIC DIVERSITY OF *Aspidosperma quebracho-blanco* Schltld. POPULATIONS FROM PROTECTED AREAS OF DRY CHACO

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Genetic diversity of *Aspidosperma quebracho-blanco* Schltld. was assessed by RAPDs from populations growing in four protected areas: QL, BV, C and SG, and one unprotected area, SJ. UPGMA cluster analysis using the Jaccard's similarity coefficient generated a dendrogram, which associated the exemplars into five groups. Principal Coordinate Analysis (PCoA) confirms the associations between samples observed in the cluster analysis. AMOVA analyses revealed high genetic variation within populations (78%) and low variation between populations (22%), in agreement with values estimated by Shannon-Weaver index. Genetic differentiation between populations estimated by AMOVA was $\Phi_{PT} = 0.216$ ($P < 0.001$). Nei's G_{ST} (0.236) was lower for interpopulation variation. Gene flow $N_m = 1.62$.

The high genetic diversity within populations suggests that populations have the capacity to adapt to environmental changes. These populations are priority areas for conservation of forest genetic resources.

129.

EB30 - GERMINATION EXPERIENCE IN CACTI (*Trichocereus pasacana*)

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Trichocereus pasacana is a native species in northern Argentina and southern Bolivia. It is a giant candelabra-shaped cactus and it can be 12 m high. It is very slow to grow. Nowadays local extinctions are happening due to timber exploitation, and a lot of pests are damaging it. In the natural habitat seeds and sprouts are found under the canopy of the plant and in this way they behave like nourisher to facilitate the cacti growth. Members of the PEAM, having in mind this situation have started an experience to mitigate the problems of this species in its natural repopulation. The experience started with the cacti fruit collection in Jujuy in January 2008. The seeds were kept in a cool and dry place and on 25/11/08 the seeds were sown in four trays containing different substrates (different ratios of commercial cacti soil, gravel and volcanic sand). The trays were placed at room temperature. The watering was made by immersion. On 15/01/2009 a general germination was observed in the four trays without differences between them. In April 371 cacti were transplanted into individual pots. They were 1.2, 0.7 and 1.5 cm of height, diameter and root respectively. In the last evaluation (06/29/11), the 52% of transplanted cacti were growing in different sizes, the largest being about 6 cm high and 4 cm of diameter. Planting trays were kept and continuous seed germination was observed after 2.5 years. This experience proves the high germination rates of cacti. The total cacti born were 642. The second stage of this work aims to take the cacti back to their natural environment.

130.

EB31 - GROWTH ANALYSIS OF YOUNG TREES CULTIVATED IN URBAN CONDITIONS UNDER DEFICIT IRRIGATION

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The urban forests of arid zones have growth patterns related to the availability of water, which is in turn increased by some other variables. The aim of this paper is to evaluate the growth responses of young trees of different species used in the city of Mendoza (Argentina) under differential availability of water. Four tree-species are analyzed: *Acacia visco*, *Platanus hispanica*, *Fraxinus americana*, and *Morus alba*. The experimental design tests the irrigation responses under controlled conditions in nursery during three growing seasons. Three treatments were applied: T1 Control (reposition of 100% water transpired); T2 Moderate water deficit (reposition 66%) and T3 Severe water deficit (reposition 33%). The quantified growth's variables included the height, the stem diameter, the leaf area and the width of tree-rings. Results show that the growth responds to differential volumes of water and that the responses vary between each specie and the treatment. The ANOVA statistical evaluation ($\alpha \leq 0.05$) supports these results. The four species slow growth under severe water stress (T3). For height and diameter, *A. visco* and *P. hispanica* show no significant difference in T1 and T2, which indicates that a reduction of 33% in irrigation allow a growth comparable to the one achieved under control treatment. *F. americana* presents difference between T1 and T3. However, *M. alba* presents significant difference in the height under the three treatments. The accumulated growth in the tree-rings for the test period indicates that under severe water deficit the four species decrease their radial growth. Data indicate that *P. hispanica*, *F. americana* and *M. alba* are sensitive to drought, while *A. visco* is more tolerant and it have a high degree of adaptation to moderate water stress.

131.

EB33 - EFFECT OF TEMPERATURE ON THE GERMINATION OF *Euphorbia davidii*

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Euphorbia davidii is an annual weed with wide distribution in the center of Buenos Aires province which reducing yield of soybean, sunflower and corn crops (59°52'52"W, 36°46'05"S). To determine the effect of temperature on the germination were used seeds from two harvests (1995 and 2009) that were stored in laboratory conditions. Using 2,3,5 tryphenyl-tetrazolium chloride test, seed viability was 40% and 73.5% for 1995 and 2009 harvest, respectively. Both seed groups were put to germinate on tissue paper inside compartments in a thermal gradient. Temperature range was from 5.5 to 28 C with incrementing of 1.5 C in each compartment, resulting in 16 temperatures assayed. While maximum seed germination of 27.5% at 22 C for 1995 harvest, it was obtained 64% at 16 C for harvesting 2009. These values correspond to 69 and 87% of viable seeds in the harvest 1995 and 2009, respectively. The optimum temperature range for seed germination was from 11.5 to 22 C for harvesting in 1995 seeds and from 13 to 20.5 C for harvesting in 2009. Maximum germination was 17 C in both cases. Few seeds germinated below 8.5 C and above 26.5 C. *E. davidii* can germinate in a wide range of temperature. Determination of temperature of germination is a biological parameter used to developing integrated strategies of management.

132.

EB34 - CACTACEAE FAMILY IN SAN LUIS PROVINCE, ARGENTINA

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San Luis province is located between parallels 31°50' and 36° south latitude, and the meridian 64°55' to 67°15' west longitude. It has an average length of 460 km in N-S direction and 200 km in the E-W direction. The climate has large seasonal differences: cold winters and hot summers. Rainfall is low (between 200-700 mm) and decreases from east to west, it occurs mainly in summer. There are three eco-regions: *Chaco Seco*, *Espinal* and *Monte de Llanuras y Mesetas*. *Cactaceae* family comprises about 200 genera and 2.000 species; it is distributed in warm and semiarid regions of America. In Argentina, there are 37 genera and 210 species. This family has the largest percentage of endemic species in our country. The aim of this paper is to contribute to the knowledge of the diversity of *Cactaceae* family in San Luis Province and to revalue its conservation. The methodology is based on the collection of material and conservation in cactario, identification by classical methods of systematic, bibliographic searches, morphological description, photographs and geographic distribution. The nomenclature of the taxa cited was corroborated by the on-line version of the Darwinian Institute of Botany.

A total of 31 taxa belonging to 12 genera has been identified so far: 25 taxa are endemic, 5 natives and 1 adventitious. The three genera with higher number of species are: *Gymnocalycium*, *Opuntia* and *Tephrocactus*.

133.

EB36 - SAPROLEGNIASIS RECURRENCE IN *Astyanax eigenmanniorum* AND *Astyanax fasciatus* (PISCES: CHARACIDAE)Mancini M¹, Salinas V¹, Morra G¹, Rodríguez C¹, Liendo A², Taricco E³, Ballester S³.¹FAV-UNR. ²Mun. Almafuerite. ³Guardafauna.
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Fish kills or mortality events of few species have occurred seasonally in Piedras Moras reservoir (32°10.13'S, 64°16.50'W, 832 ha), Córdoba (Argentina). The mortality events described here occurred in september of 2011. Fish were caught and 10 samplings of water quality were done. The species most affected were *Astyanax eigenmanniorum* and *Astyanax fasciatus*, which together represented more than 98% dead fish. The specimens showed changes of swimming, lethargy and skin lesions of fungal infection. Laboratory studies confirmed the presence of *Saprolegnia* sp. The records of temperature, pH, dissolved oxygen and water transparency analyzed *in situ* were of 15.7±0.31°C, 7.95±0.16, 8.84±0.13 ppm and 6.5±1.03 m respectively. The oxygen saturation was 95%. Clinical signs, lesions present and the species affected, coincide exactly with an outbreak caused by *Saprolegnia parasitica* in the same reservoir in winter of 2007, due to low temperatures. There were no significant differences of temperature between the two cases ($P>0.05$), however the number of dead fish was markedly lower than in 2007. In addition to low water temperature, winter Saprolegniasis recurrence in *A. eigenmanniorum* and *A. fasciatus*, reinforces the hypothesis that there are components of their biology that would make them more susceptible than other species

134.

EB37 - *Contracaecum australe* FROM *Phalacrocorax brasilianus* IN CÓRDOBA PROVINCE, ARGENTINABiolé F¹, Mancini M¹, Guagliardo S², Tanzola D², Salinas V¹, Morra G¹.¹FAV-UNRC (Río Cuarto). ²UNS (Bahía Blanca).
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Species of the genus *Contracaecum* (Nematoda: Anisakidae) parasitize aquatic organisms that inhabit different environments. Several species of cormorants around the world are definitive hosts of these nematodes. As there is no many records of *C. australe* in cormorant *Phalacrocorax brasilianus* of the central region of Argentina and the discovery of larvae in fishes has increased in recent years, the aims of this study are: to communicate the presence of this species in continental biotope, to describe the specimens and to compare them with previous records. Cormorants were captured in three lentic and one lotic system of the Córdoba province (Argentina), a shallow lake (SL: 34° 46'00 "S, 63° 38' 38" W), a reservoir (RE: 32° 10'40 "S, 64° 17' 12" W), an urban Lake (UL: 33° 06'20 "S, 64° 22'32 "W") and a river (RI: 33° 07'19 "S, 64° 18' 24" W). The parasites were cleared with lactofenol and observed in light microscopy and scanning electronic microscope (SEM). Birds dissected were positive for *C. australe*, with a maximum intensity in the UL of 69 parasites and minimum in of 3 adult parasites in RE. The observations of different morphological details allowed to include them in *Contracaecum australe* species. The wide distribution of *P. brasilianus* contributes with the dispersion and life cycle continuity of these nematodes.

135.

EB39 - DETERMINATION OF CHLOROPHYLL-A CONCENTRATION IN RIO TERCERO DAM USING CBERS-2B SATELLITE IMAGEBonansea M¹, Rodríguez C², Ledesma C².¹Becario CONICET. ²Universidad Nacional de Río Cuarto. E-mail: mbonansea@hotmail.com

Satellite images, provided by remote sensing, can be used as an innovative tool in monitoring, control and management of water resources. The objective was to model the spatial distribution of chlorophyll-a (chl-a) in Río Tercero dam (32,2128 W, 64,4761 S), located in Córdoba. In its coasts there are several large urban settlements which use this resource as a source of water for human and animal consumption, irrigation, power generation, fishing and recreation. Samplings were conducted during fall of 2008 coinciding with the passage of the CBERS-2B Satellite by the study area. 15 sites were selected inside of the dam, in each one geographic location coordinates and physicochemical parameters were measured. In laboratory concentration of chl-a was determined. We used a CBERS-2B image (Date:27/04/2008), which was georeferenced (RMS=0.84). A PCA was performed to identify associations between reflectance data of the image's bands and the values of chl-a measured. The estimated response ($R^2=0,73$) for multiple linear regression model of the parameter chl-a was created, using as independent variables bands 2 and 3 of CBERS-2B satellite. In conclusion, we were able to evaluate the spatial behavior of the chl-a in Río Tercero dam, providing information relating to the distribution of this parameter over the entire reservoir.

136.

EB40 - DETECTION OF ENTOMOPATHOGENIC NEMATODES (HETERORHABDITIDAE) IN TWO DEPARTMENTS OF CÓRDOBA

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Entomopathogenic nematodes belonging to the Families Steinernematidae and Heterorhabditidae are organisms naturally occurring in the soil and they are important regulators of insect populations. The diversity of this group in the province of Córdoba is represented by five species of the genus *Steinernema* and one species of the genus *Heterorhabditis*. The aim of this work was to detect new populations of this group in order to identify native species that can be used as biological control agents. In March and April 2010, samplings were conducted in departments of Tulumba, Ischilín, Cruz del Eje and Totoral. Seven sampling sites were considered; 5 subsamples were randomly taken from each site, covering an area of 8 to 10 m² and collecting an approximate soil volume of 1 kg, between the surface layer and 30 cm in depth. Samples were processed in the laboratory by the insect-baiting technique, using larvae of the lepidopteran *Galleria mellonella*. Entomopathogenic nematodes of the Family Heterorhabditidae were detected in the departments of Totoral and Ischilín. The presence of Steinernematidae had already been reported in Totoral, but the presence of Heterorhabditidae had gone undetected. This is the first record of entomopathogenic nematodes for the department of Ischilín. Although the populations found needs to be identified, the data obtained expand the distribution records for Córdoba and provide new biological control agents.

137.

EB42 - PROTEROCHAMPSIDS, MEMBERS OF TRIASSIC ECOSYSTEMS FROM SOUTH AMERICA

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Proterochampsids are quadrupedal fossil reptiles with a crocodile-like body and notably skull ornamentation. This group is included within archosauriforms and is considered the sister-taxon of Archosauria. Proterochampsids are endemic for the Middle to the Late Triassic of Argentina and Brazil. The best known proterochampsids come from the Chañares (ca. 241 Ma) and Ischigualasto (ca. 231 Ma) formations (Argentina). In this work two species of Proterochampsids are presented, *Proterochampsia barrionuevoi* and *Chanaresuchus sp. nov.* The sediments which contained the specimens here analyzed represent a fluvial sedimentary succession. This evidence indicates that proterochampsids were aquatic or semiaquatic animals. Body mass analyses are being carried out taking the circumference of the humerus and femur and applying Anderson's equation as a mass estimator. The results depicted that proterochampsids had a mass between 1.2 and 28.4 kg. Accordingly, these proterochampsids were medium-sized aquatic animals that inhabited South American Triassic fluvial environments.

138.

EB44 - WATER USE EFFICIENCY AND NITROGEN USE IN ARID ECOSYSTEMS (TELTECA) BY STABLE ISOTOPESAranibar JN, Meglioli PA, Villagra PE, Guevara A¹.

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In arid ecosystems, water is the main resource that regulates primary productivity. Plants have different adaptations to optimize water use such as stomatal conductance and photosynthesis regulation as a function of local meteorological conditions. Nutrient use is optimized based on environment availability and metabolic requirements. We explored water and nutrient uses on phreatophytic woodlands in Telteca in different environmental conditions (dunes/groundwater coupled interdune valleys), (after/before rainfalls), (disturbance/non-disturbance woodlands). We analyzed foliar carbon (C) and nitrogen (N) total concentration and their stable isotope compositions as indicators of nitrogen use strategies and water use efficiency. *Prosopis flexuosa* presented the highest water use efficiency among C₃ plants in dry periods, both in dunes and interdune valleys. Trees in livestock posts had lower water use efficiency than in relatively undisturbed woodlands. We found no clear evidence of atmospheric N₂ fixation, except in some seedlings of *P. flexuosa*. High foliar C:N ratios were observed prior to senescence in woodlands, indicating N conservation mechanisms, whereas lower C:N ratios were observed in livestock posts, indicating a higher N availability. We conclude that foliar stable isotope composition indicate an environment regulated by local meteorological conditions and disturbances.

139.

EB46 - GERMINATION OF A NATIVE WOODY SPECIES WITH HIGH ORNAMENTAL VALUEDe Luca N¹, Cantero JJ^{1,2}, Fernández EM².

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Caesalpinia gilliesii (Caesalpinioideae), woody species with ornamental value, grows spontaneously in Argentina. Its establishment can fail because of the hard and impermeable integument in seeds. Seed collection was conducted in January 2008 in Alpa Corral (32° 41' 53" S, 64° 43' 24" O), Córdoba, Argentina; they were stored for a year in semipermeable container in the refrigerator (5°C). Moisture content and germination were evaluated at 20°C. Treatments were scarified humid (EH) and mechanical (EM) and the control (T). Initial moisture content of seeds was 8,27% ± 0,02. Germination is epigeal, seedlings measured 24cm (14 days). Differences among the treatments were found EM: 96,67%, EH: 98,3% and T: 13,3%. Between EM and EH there were no differences ($p > 0,5185$). After storage, seeds retained their ability to germinate, with 7,48% ± 0,14 of moisture, classified as orthodox.

140.

EB47 - GERMINATION OF *Myrcianthes cislplatensis* "MATO" (MIRTACEAE) OF THE NATIVE FOREST IN THE MOUNTAINS NORTH OF CÓRDOBADe Luca N¹, Cantero JJ^{1,2}, Fernández EM².

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Myrcianthes cislplatensis grows at the mountains north of Córdoba, Argentina, where the native forest area has experienced a drastic reduction. For conservation it is necessary to know the requirements for germination and storage of seeds. Fruits were collected in the Cerro Colorado, Córdoba (30° 05' 53" S, 63° 56' 12" O). At 15 days and 3 months of storage (4°C) the moisture and germination (TG: 20°C) were evaluated, registering normal and abnormal seedlings, hard, fresh and dead seeds, to calculate the percentage (PG) and the mean time of germination (TMG). Before the TG a group of seeds were immersed in water (IH) at 25°C for 24hrs. Another untreated group corresponded to the control (T). After 15 days of the collection, the fruits reached 42,37% of moisture. Insect damages were observed in seeds (50%). Germination is hypogeal. The IH did increase the PG (53% vs 15%). TMG was 10. At three months, the moisture content was 31,5%, the PG fell significantly (5%) and a 95% of seeds was dead, so they might be recalcitrant seeds.

141.

EB52 - TROPHIC ECOLOGY OF *Oryctolagus cuniculus* IN THE MONTE DESERT (LEONCITO PARK, ARGENTINA)

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The European wild rabbit (*Oryctolagus cuniculus*) is exotic species and was record recently in a wetland area in the "Leoncito" National Park. We determined the diet composition and assessed the relationship between diet and plant availability. Collection of faeces and vegetation sampling were made in wet and dry season. Diet composition was analysed with microhistological analysis. In wet season, graminoid (*Junco* sp.) and herbaceous (*Descurainia* sp. and *Convolvus arvensis*) were the most abundant cover type, but grasses (*Polipogon* sp. and *Bromus unioloides*) and shrubs (*Adesmia trijuga* and *Lycium chilensis*) were more eaten by wild rabbit. During dry season, graminoid (*Junco* sp.) and herbaceous (*Hirschfeldia incana*) were the most representative plant cover, but the most consumed species were grasses (*B. unioloides* and *Polipogon* sp.) and shrubs (*A. trijuga*). The wild rabbit has a selective diet because their intake of grasses and shrubs was significantly higher than the environmental availability in both season. Our results disagree with others works in Patagonia, where shrubs were consumed only when graminoids and grasses were the lowest plant cover. Probably the inclusion of higher proportion of shrubs in both seasons could be a foraging strategy to ensure a nutritional balance.

142.

EB53 - FIELD PARASITISM OF *Trichopoda giacomelli* (DIPTERA: TACHINIDAE) ON WINTER *Nezara viridula* (HETEROPTERA: PENTATOMIDAE) ADULTSWerdin González J^{1,2}, Gutiérrez MM¹, Ferrero AA¹.¹Laboratorio de Zoología de Invertebrados II. Universidad Nacional del Sur. ²CONICET. E-mail: jwerdin@hotmail.com

In Argentina, *N. viridula* is attacked by *T. giacomelli*, a species which has shown to regulate populations of this soybean pest. This is the first study about the parasitization by *T. giacomelli* on overwintered adults of *N. viridula* from Rivera city (27° 09' 38" S, 63° 14' 48" N), Buenos Aires, Argentina. The studies were carried out from 2005 to 2009. The oviposition pattern, superparasitism and its relationship with the Apparent Parasitism (AP) and the Effective Parasitism (EP) were studied. Along this study, eggs of *T. giacomelli* were found on host bodies. The percentage of parasitism was 13.7% ranging from 8.6% to 18.1% and males consistently had a higher parasitization levels than females. Supernumerary oviposition was found to be common on both genders but males showed a higher incidence of tachinid eggs and higher numbers of eggs per individual. The highest tachinid eggs concentration was found on the thorax, mostly on the pronotum and on the ventral surface for males and females, respectively. A significant correlation between number of tachinid eggs on host body and the EP was found, so field estimation of *N. viridula* adult parasitization could be done using the AP.

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143.

EB55- LIMNOLOGICAL AND ZOOPLANKTONIC PARAMETERS FOR TWO YEARS IN A TEMPORARY LAKE (LA PAMPA)

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Temporary wetlands may show large variations in their physico-chemical and biological parameters in short periods. Thus, short-term studies do not explain their long-term dynamics. In Argentina, although these ecosystems are frequent, they have not received much attention. The aim of this study was to compare the physico-chemical parameters, taxonomic composition, abundance and zooplankton biomass in two consecutive years in a temporary saline lake of La Pampa. Samples were collected seasonally during 2006 and 2007. Salinity was different ($H = 5.55$, $P = 0.0209$) and increased from 23.98 to 36.7 $g\ l^{-1}$, whereas the opposite was observed with transparency, phosphorus concentrations, chlorophyll *a* and suspended solids. The richness was reduced (from six species in 2006 to three species in 2007) and showed a negative correlation with salinity ($r_s = -0.71$, $p = 0.0488$). *Boeckella poopoensis*, *Moina eugeniae* and *Brachionus plicatilis* were recorded in both periods. The total abundance of copepods and rotifers did not differ, in contrast to that observed with that of cladocerans ($H = 5.33$, $P = 0.0209$). Its density was correlated with salinity ($r_s = -0.81$, $P = 0.0149$) and was lower in 2007 as *M. eugeniae* decreased from 466 to 6.6 $ind.l^{-1}$. Only cladocerans biomass was different ($H = 5.33$, $P = 0.021$) due to the decrease in *M. eugeniae* from 3332.4 to 34.7 $\mu g.l^{-1}$.

144.

EB56 - BIOLOGY OF *Moina macrocopa* (CRUSTACEA CLADOCERA), AN INTRODUCED SPECIES IN LA PAMPA

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Two subspecies have been described for *Moina macrocopa*: *M. macrocopa macrocopa*, originally restricted to the Old World, and *M. macrocopa americana*, restricted to North America. Specimens found in Bolivia, Corrientes and Rio de Janeiro were identified as belonging to the former, which led to propose an anthropic introduction. The aim of this study was to study the biological characteristics and distribution of *M. macrocopa macrocopa* in La Pampa province. This subspecies was recorded in five shallow lakes, between 1999 and 2007. The salinity in these lakes ranged from 5.7 to 21.81 $g.l^{-1}$, the water transparency from 0.02 to 1.96 m and the temperature from 9.4 to 26.8°C. The maximum presence of *M. macrocopa macrocopa* was limited to four months and its abundance ranged from 1 to 312.7 $ind.l^{-1}$. Parthenogenetic females predominated (mean: 8.4 eggs), but gamogenetic individuals were found in four lakes. The size ranged between 0.55 and 1.69 mm and the maximum was higher than that previously reported. Although it was not classified as a halophilic species, in La Pampa it was recorded only in saline lakes. Its presence could be due to the introduction during stocking of *Odonthestes bonariensis* from Buenos Aires, where it is used as food for these fish. However, the dispersion would be related to natural factors since all the lakes in which it was found lacked fish fauna.

145.

EB57 - ZOOPLANKTON RICHNESS AND ABUNDANCE IN SIX "MERCEDINAS" LAKES (SAN LUIS, ARGENTINA)Cabrera GC¹, Vignatti A¹, Salinas V², Echaniz S¹, Mancini M².¹FCEyN UNLPam, Santa Rosa, La Pampa. ²FAYV UNRC, Río Cuarto, Córdoba. E-mail: gaby_cab3@hotmail.com

In southern San Luis province in central semiarid Argentina, more than one hundred shallow lakes are located in arid watersheds between dunes and grasslands. These shallow lakes are known as "mercedinas", for their proximity to Villa Mercedes city. The aim of this study was to determine the zooplankton taxonomic composition, abundance and its relations with limnological parameters and the fish fauna of six of these lakes. The samples were taken in October 2007. The maximal depth ranged from 2.3 to 3.6 m, salinity from 1.3 to 3.3 g.l⁻¹ and transparency from 0.3 to 2.3 m. There were seven fish species. *Jenynsia multidentata* was found in the six lakes and *Odontheistes bonariensis* in four. The zooplankton richness was 20 taxa: five cladocerans, six copepods and nine rotifers. Species common to the six lakes were *Bosmina huaronensis*, *Ceriodaphnia dubia* and *Filinia* sp. The first was most abundant in five, with densities between 15.6 and 113.2 ind.l⁻¹, and the second in only one with 56.3 ind.l⁻¹. This lake was different from the rest: besides the high abundance of *C. dubia*, it showed the presence of *Daphnia spinulata*, both of which are large cladocerans, whose presence could be due to the absence in this lake of planktivorous fish such as *O. bonariensis*.

146.

EB58 - *Artemia persimilis* (CRUSTACEA ANOSTRACA) POPULATION DYNAMICS IN A HYPERSALINE LAKE OF LA PAMPA

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Hypersaline lakes can only be inhabited by organisms that have physiological mechanisms to withstand osmotic stress. Among these organisms is *Artemia*, represented in Argentina by two species: the introduced *A. franciscana* and the native one *A. persimilis*, the only species recorded in La Pampa. Several works have studied some of the biological aspects of *A. persimilis*, but, since most have been carried out in the laboratory, there are few data on their ecology in natural conditions. The aim of this study was to determine population parameters of *A. persimilis* in relation to environmental variables in a hypersaline lake of La Pampa. Seasonal samples were taken during 2007 in La Amarga, a large lake of 11 109 ha, which had a salinity of 115 g.l⁻¹, low chlorophyll *a* concentration (1.65 mg m⁻³) and high transparency (1.54 m). We recorded low densities of *A. persimilis* and the mean was 1.56 ind.l⁻¹ with a maximum of 4.65 ind.l⁻¹ in spring. With the exception of winter, when post-larvae and adults predominated (43 and 50%), during the other seasons the highest proportion was contributed by nauplii, metanauplii and postmetanauplii. No correlations were found between total abundance or different stages and specific environmental parameters.

147.

EB61 - HABITAT CHARACTERIZATION OF *Microcavia australis* (Rodentia: Caviidae) IN A ROCKY MOUNTAIN IN LA LAJA

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The habitat used by *M. australis* (cuis chico) was characterized in a rocky place in the Mount desert of San Juan. We worked on five rocky places in La Laja, Albardon with individual's presence and/or activity signs of *M. australis*. In each rocky place, we recorded its size, percentage of vegetation, rocks, mulch (litter) and bare soil cover. Also, we considered the dimensions of the entrances, overhangs and shelters in each burrow system, the orientation of each entrance and the soils hardness. In the study site, we found little vegetation cover and high coverage of rocks. In the five rocky places we identified 52 principal burrow systems and 63 shelter burrow systems. Both types of systems were differentiated by its depth. The entrances of the principal burrow systems were deeper than the shelters allowing individuals to find more stable microclimate in relation to depth. The shelter burrow systems had a greater number of entrances respect to principal burrow systems which would reduce the predation risk. The largest proportion of used entrances were orientated to the SW and S, which allows the entry of fresh winds in warm seasons to reduce the temperature within the burrow. In the study site, the vegetation cover would not be used as a shelter from predators, but that function would be carried by rocks, which also would function as a temperature shelter. In addition, the individuals of *M. australis* use the cracks and crevices of rocks for all its activities because the soil in the area is very hard to dig their own caves. This work constitutes the first study of *M. australis* associated with rocks.

148.

EB62 - BIODIVERSITY AND PLANTS TAXONOMY: ANALYSIS OF THEORETICAL AND PRACTICAL ASPECTS OF *Tillandsia capillaris* COMPLEX

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Traditionally, conservation biology used the number of species for measure biodiversity and established priority sites considering the taxonomic distinctness and geographical distributional patterns of taxa (e.g., endemism). Different criteria for classifications could change the number of species of a genus and therefore the geographical delimitation of entities and this could influence decision of what to conserve. *Tillandsia capillaris* Ruiz & Pav. complex (Bromeliaceae: Tillandsioideae) has a pronounced morphological variation and has been undergone various classifications. During S. XIX have been described at least 15 species related to the complex and some sub-varieties. In 1935 Smith L.B. recognized one widespread species divided into 5 forms and in 1989 Till W. defined two species with wide and overlapping distribution. We analyzed the pattern of morphological variation of the complex using morphological characters and multivariate statistical techniques. We proposed a new classification that recognizes three taxa in the complex and raise the entities to a higher taxonomic category. This group can be easily recognized in the field and could help in making decision of priority conservation sites. We also think that a very general and simple classification could result in a disadvantageous way for the specie status and for the conservation of biodiversity.

149.

EB64 - RICHNESS OF FISHES IN THE SAN JUAN PROVINCE

Pastor P, Scarabotti P, Fernández L, Cataldo A, Moreno D, Quiroga L, Galvani G, González E, Cortéz R, Marinero V, De Giovanini C, Navas A, Herrera M, Sanabria E.

FCEFN-UNSJ, IHEM-UNCuyo; IZA-UNC; CONICET.

San Juan ichthyology region is located in the Sub Andean-Cuyo, which includes the provinces of La Rioja, Mendoza, La Pampa northwest as well as west and northwest of San Luis. The aim of this work is to update the list fish fauna of San Juan province. No systematic sampling was conducted and we use different techniques of fishing (rod and bait, nets, in other). Results obtained as a list of richness of fish fauna to San Juan which is comprised of a total of 17 species belonging to the taxonomic arrangement follows: Atherinopsidae (2), Characidae (3), Cyprinidae (2), Anablepidae (1) Poeciliidae (1), Percichthyidae (1), Salmonidae (1) Trichomycteridae (3), Pimelodidae (1), Diplomystidae (1), Synbranchidae (1). Of which 11 belong to the native fauna and 6 were introduced for different reasons. This listing is an update of the fish species in the San Juan province, with 11 records listed here first dates for this type of fauna. Greater efforts are needed to understand the diversity of fish species that are part of the rivers and streams in order to have basic information for the conservation of these environments.

150.

EB66 - DIVERSITY OF AMPHIBIANS AND REPTILES FROM SAN JUAN PROVINCE, ARGENTINA: AN UPDATE

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The aim of this work is to present an update checklist of San Juan's herpetofauna. This work is framing in a general project whose goals are to know the biodiversity through surveys of species richness, scientific collection management and evaluation of biological parameters for determinates the conservation status of the species. The checklist of amphibians and reptiles was composed by 85 species: 1 tortoise: Testudinidae. 48 lizards: 7 Leiosauridae, 32 Liolaemidae, 1 Tropiduridae, 1 Scincidae, 4 Phyllodactylidae (=Gekkonidae) and 3 Teiidae. 1 amphisbaenid: Amphisbaenidae. 19 snakes: 2 Leptotyphlopidae, 1 Boidae, 12 Dipsadidae (=Colubridae), 1 Elapidae y 3 Viperidae and 16 amphibians: 3 Bufonidae, 3 Leptodactylidae, 3 Leiuperidae, 4 Cycloramphidae, 1 Ceratophrydae, 1 Hylidae y 1 Ranidae. Species with taxonomic conflicts and uncertain distributions records were commented. Studies with holistic approach are important to identify vulnerable species and these results could be important for future conservation strategies.

151.

EB67 - DIVERSITY OF PROTEOCEPHALIDEAN CESTODES OF SURUBÍES (PISCES: SILURI-FORMES, *Pseudoplatystoma* spp.) FROM THE PARANÁ RIVER, ARGENTINA

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The proteocephalidean cestodes parasitize mainly freshwater fishes. In South America, their major diversity is found in siluriforms. The surubíes inhabit the most important basins (Amazon, Paraná and Uruguay Rivers); in Argentina there are 2 species (*Pseudoplatystoma corruscans* and *P. fasciatum*). Their proteocephalidean richness is high, with 6 cited species in *P. corruscans* and 7 in *P. fasciatum*. The aim of this work is to contribute to the knowledge of proteocephalids diversity. Worms found in the gut were isolated, fixed and processed following helminthological techniques to study internal morphology and the microthrix pattern. In Argentina, *P. corruscans* is parasitized by: *Choanoscolex abscissus*, *Harriscolex kaparari*, *Megathylacus travassosi*, *Monticellia spinulifera*, *Nomimoscolex pertierra*, *Peltidocotyle rugosa* and Proteocephalidea sp. 1, whereas *P. fasciatum* harbours *M. spinulifera*, *Nomimoscolex lopesi*, *N. sudobim*, *P. rugosa*, *Spatulifer rugosa*, Proteocephalidea sp. 2 and Proteocephalidea sp. 3. In Argentina, neither *C. abscissus* nor *Housayella sudobim* were recorded in *P. fasciatum* and these hosts share only 2 species (*M. spinulifera* and *P. rugosa*). The diversity of proteocephalids in these fish increased with increasing capture effort. In conclusion, the proteocephalidean richness is similar in number but different in composition in surubíes from Paraná and Amazon Rivers.

152.

EB68 - INVASION BY EXOTIC PLANT AT LOWLAND AND UPLAND OF THE POTRERO RIVER (POTRERO DE LOS FUNES, SAN LUIS, ARGENTINA)

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Invasion by exotic plants is recognized worldwide as a serious problem for the species conservation. The aim of this report is to explore the factors that could influence the success of invasive plants at highland and lowland of Potrero River (San Luis, Argentina). The methodology used was phytosociological inventories performed at 7 sites, located along an altitudinal gradient at the river banks. At each site two plots were located, one on the first rung of the river (A) and the other at its edge (B), following the criterion of minimum community area. The analyzed variables were: degree of human impact, altitude, habit of the plants and physical-chemical characteristics of soil. Invasive plants were significantly higher in (B) than in (A). These sites also showed a higher percentage of moisture and a lower content of organic matter. The degree of human impact and the elevation showed correlations with the invasion by plants, positive with the first of these variables and negative with the second. Both variables have been cited in connection with the invasion by exotic plants in several occasions. We conclude that, the sites most prone to invasion by exotic plants were low, wet and human impacted, should be especially careful of not to use exotic plant, when it's forested and landscaped, in sites with these characteristics.

153.

EB70 - ONTOGENETIC VARIATION OF THE DIET OF *Rhinella arenarum* IN THE DRY CHACO, SAN JUAN, ARGENTINA

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The aim of this work is describe and provide data on the trophic ecology of *R. arenarum* in Valle Fertil, San Juan. We analyzed 32 specimens: adults 11 (snout-vent length (LHC) = 9.2 ± 8.3 cm.) sub-adults 12 (LHC = 6.8 ± 4.0 cm.) metamorphosed 9 (LHC = 3.3 ± 1.42 cm.). The trophic spectrum in adults consisted of 6 items and the contribution of each category to the diet (IRI) was: Lepidoptera (6861.4), Hymenoptera (515.0), Coleoptera (153.9), Anura (24.7), Orthoptera (7.4), Homoptera (2.9). For juveniles the trophic spectrum was 8 items, according to IRI: Lepidoptera (4236.4), Hymenoptera (599.3), Homoptera (79.4), Coleoptera (43.7), Araneae (25.8), Diptera (17.2), Collembola (1.8), Vertebrata (1.4). For metamorphosed the trophic spectrum was 6 items, IRI: Homoptera (4026.1), Lepidoptera (817.7), Hymenoptera (587.7), Diptera (28.8), Hemiptera (21.1), Larva (4.8); Araneae (4.7). The ontogenetic relationship of the morphological variables: LHC vs. maximum length of the prey, LHC vs. maximum width of the prey and LHC vs. maximum volume of the prey showed significant and positive relationships. Lepidoptera is the most consumed items, for juveniles and adults, unlike the metamorphosed the most consumed items was Homoptera. The selection of prey is strongly influenced by body size of the specimens studied (ontogeny).

154.

EB71 - BIOLOGY AND CONSERVATION OF ANDEAN LIZARDS: STUDY CASE OF *Pristidactylus scapulatus*

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Pristidactylus scapulatus is a lizard that inhabits in central-western Argentina. Currently is categorized as "insufficient known" due to biological aspects still unknown. For these reason, the aim of this work was to provide basic biological data for use in categorization of this species following the SUMIN criteria. During 2008 to 2010 we captured 17 individuals (10 males / 7 females) in the Andes range mountain of San Juan and we obtained data on trophic ecology, thermoregulation, microhabitat use, activity and sexual dimorphism. About diet Coleoptera was fundamental item. Body temperature is explained by the substrate temperature suggesting a tigmothermic strategy of thermoregulation. Regarding microhabitat use, the more frequently used was basaltic rocks exposed to solar radiation. *P. scapulatus* was active during 7 hours per day (12:00 to 19:00hs) suggesting a unimodal activity pattern. The species present a bold sexual dichromatism and males differs morphometrically from females in head, hindlimbs and tail size. Determination of these biological parameters results essentials to determine their conservation status.

155.

EB72 - PRELIMINARY LIST OF BIRDS OF A SECTOR OF VALLE FÉRTIL PROVINCIAL NATURAL PARK

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Biodiversity study in birds is necessary to establish interest for conservation sites and permit evaluates the status of populations. The aim of this work is survey the birds in a protected area from the western Chaco, Valle Fertil, San Juan, Argentina. Transects of sightings were walked in northeast of the park during march and april 2011. As first results, we obtained a list of 54 species. Most of them belong to Passeriformes order. On the other hand, as families Tyrannidae, Emberizidae, Columbidae and Falconidae are highlighted. The most abundant species were *Cyanoliseus patagonus andinus*, *Myiopsitta monachus calita* y *Molothrus bonaerensis*. Conservation priority species are *Vultur gryphus* categorized as "almost threatened" in globally level and *Spizapteryx circumcinctus*, as "Vulnerable" nationwide. We cite for the first time *Columba picazuro* and *Craniolaeca pyrrhophia* for San Juan province and 14 new species for Valle Fertil department. This result represents the first data on Valle Fertil park's avifauna and will be part of base line for the purpose of declare the protected area as "Reserva de Usos Múltiples Valle Fertil".

156.

EB73 - COMPARATIVE REPRODUCTION STUDY OF FIVE LIZARDS SPECIES FROM MONTE, SAN JUAN, ARGENTINA

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Lizards because of their wide distribution and great diversity of living environments have different reproductive patterns. For annual cycles, two strategies have been established: the first of marked seasonality with a peak in spring and summer, and the second characterized by autumn reproduction. During the evolution of reproductive cycles, the factors considered as the main promoters of reproduction are: temperature, humidity, photoperiod and intrapopulation asynchrony (mainly females). But is still limited the knowledge of some population characteristics, such as life expectancy on the reproductive phenology, because others will depend on it: time to reach sexual maturity, fertility and number of reproductive events in each life cycle. We performed a comparative study of five species from three genera (*Liolaemus*, *Homonota* and *Cnemidophorus*) for two consecutive years, analyzing gonadal patterns and cycles. There were gender differences about cycles and patterns, while intrageneric differences (*Liolaemus*) are related to changes in reproductive parameters, such as litter size, number of litters, sexual maturity age and with events such as cycle times and ovulation and fertilization dates.

157.

EB74 - *Lepus europaeus* (European hare): DIET AND FOOD AVAILABILITY IN THE HYPER-ARID MONTE DESERTReus ML¹, Laspina CA¹, Campos VE^{1,3}, Andino N¹, Giannoni SM^{1,3}, Campos CM².¹Inst Museo Cs Ns. UNSJ. ²CCT-CONICET, Mza. ³CONICET. E-mail: reusster@gmail.com

Lepus europaeus is an exotic herbivore that can affect plant communities and compete with native herbivores. We analysed the diet and dietary changes in relation to food availability in the Ischigualasto Provincial Park (San Juan). Collections of faeces for microhistological analysis and plant cover were made in wet and dry seasons in a mesquite woodlands. Plant species were classified in: trees, shrubs, herbaceous, grasses, cactaceae, and epiphytes. We applied MANOVA and the Kruskal Wallis test. The highest plant covers were shrubs and trees. In both seasons the main food included in the diet was shrubs (*Prosopis torquata*, *Cyclolepis genistoides*, *Atriplex* sp., *Bulnesia retama*). Grasses (*Tricloris crinita*, *Cottea pappophoroides*) and herbaceous (*Tribulus* sp.) were also consumed in the wet season and cactaceae (*Tephrocactus* sp., *Echinopsis* sp.) in the dry season. The diet and food availability showed differences between the wet and dry seasons in shrub and cactaceae, and only in the wet season for grasses and epiphytes. The hare has been considered a generalist and opportunistic herbivore because is adapted to using a wide array of food resources and consume herbaceous and grasses when availability are in the wet season. However, diets were primarily based on shrub consumption in both season (higher 50%) in a stable and poor environment in terms of plant availability (21%).

158.

EB75 - CREVICES SELECTION BY *Octomys mimax* (Octodontidae) IN RELATION TO FOOD AVAILABILITYCampos VE^{1,3}, Reus ML¹, Cappa F^{1,3}, Ortuño MN², Beninato V², Giannoni SM^{1,3}.¹Inst. Museo Cs Ns, UNSJ. ²Dpto Biología. UNSJ. ³CONICET. E-mail: valeriaecampos@gmail.com

Octomys mimax its distribution seems restricted to areas with rocky slopes and ravines, is strictly herbivorous, storage and consume *Prosopis torquata*, *Maytenus viscifolia*, *Ramorinoa girolae*, cactaceae, *Halophytum ameghinoi*, *Bulnesia retama*. We assessed if the food availability can explain the crevices selection by *O. mimax*. Plant cover were recorded in columnar cactus slopes (CCS), chical (C) and creosote bush scrub (CBS) in the Ischigualasto Provincial Park (San Juan). We selected 15 active crevices (with faeces and caches) and 15 inactive crevices in each communities. The analysis were performed using generalized linear models, and Akaike Information Criterion. The selected models for CCS included *P. torquata*, *Mimosa ephedroides*, and *Deuterocohnia longipetala*. *P. torquata* was the main resource in active crevices. The selected models for C included *Zuccagnia punctata* and *R. girolae*, both species were higher in inactive crevices. For CBS, the models selected included *M. ephedroides*, *P. torquata* and *M. ephedroides*. *M. ephedroides* was higher in active crevices. Only in CCS, the crevices are in small ravines with high plant cover so this rodent selected crevices in function to food availability (*P. torquata*). In C and CBS, without ravines, benefits other than food might be obtained by crevices selection.

159.

EB76 - IMPORTANCE OF HIGH ALTITUD GRASSLANDS FOR AMPHIBIAN CONSERVATION IN THE HIGH MOUNTAINS OF CÓRDOBA, ARGENTINAVerga EG^{1,2}, Lescano JN^{2,3}, Leynaud GC², Bellis LM^{1,3}.¹Cát. Ecología, FCEfyN, UNC. ²CZA, FCEfyN, UNC. ³CONICET. E-mail: ernesver@gmail.com

The high mountain range of Cordoba is an important area because of its high conservation value. We compare amphibian diversity between streams in grassland and woodland areas. Sampling was conducted in two seasons (2009-10/2010-11) in 11 grassland streams and 7 woodland streams located above 1800m.a.s.l. We compared species richness (using rarefaction curves) and Shannon index H' (using randomization test Solow) among the rivers of both situations (grassland and woodland). We recorded a total of 232 individuals belonging to 5 species. Amphibian richness (R) and diversity (H') was higher in grassland streams than woodland streams (R: 5, R: 3 and H': 1.14, H': 0.78 p=0,003, respectively). The lower diversity in woodland streams compared to grassland streams can be due to the topographic features of the steep slopes where the forests are found and could act as a constraint to the dispersal of most amphibian species in the area. However, close canopy forest in the woodland streams may result in a decrease in water temperature and food availability, essential variables for larval develop, and therefore in a poorer environment in terms of amphibian diversity when compared to the open areas in grassland rivers. These results highlight the importance of grasslands for amphibian conservation in the high mountain range of Cordoba, Argentina.

160.

EB77 - ECOLOGICAL VARIATIONS OF TWO BIRD ASSEMBLIES IN THE MOUNTAIN FRONT OF THE ANDES OF SAN JUAN

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We studied the structure and dynamics of two assemblies of birds in the Andes of San Juan, Argentina. Ecological's structural's parameters were evaluated during an annual cycle of activity in two environments with different water availability. Were established dominance hierarchies, logging frequency, migration, trophic guilds, activity temporal and variations of daily activity in relation to environmental variables. 54 species were recorded. In terms of dominance hierarchies are could be observed to *Turdus chiguanco* as the species best represented. In terms of frequency of registration, most species belong into the category "Accidental" for both environments. According to migration status, the most of the species were occasional visitors. Insectivorous trophic guild was the best represented. There were no significant differences in the hour of sampling with respect to the abundance of individuals and species in any of the three periods of the two environments. The relationship between daily activity and environmental variables showed no significant difference, given this, the birds were not affected by environmental variables measures.

161.

EB81 - DIVERSITY AND CONSERVATION OF BIRDS OF CALINGASTA, SAN JUAN, ARGENTINA

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An annotated list of avifauna of Calingasta, San Juan province, is presented, in order to contribute to knowledge and conservation. Records were taken at April, September and December during 2009 and 2010 through censuses by counting transects and observations stations. Frequency of birds observations was calculated. An update of birds diversity by comparison with geographical distributions taken from field guidelines and publications were conducted, and comments on its conservation we provided. Two hundred and seven species belonging to 42 families, of which 142 species are confirmed records (37 families and 123 species own records). According to frequency of observations, only Cordillera Frontal's birds were categorized as "abundant", whereas the most of species were assigned to "rare" in all the studied areas. Two conservation concern species are mentioned in the area. Seven species for San Juan province and 64 species for Calingasta department were new records. The information obtained contributes to the knowledge of biodiversity and conservation status of birds of Calingasta department, useful to fix management guidelines for scarcely studied assemblages.

162.

EB83 - TROPHIC ECOLOGY OF *Liolaemus cf. ruibali* (IGUANIA: LIOLAEMIDAE) IN SAN JUAN PROVINCE, ARGENTINA

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Knowledge of dietary habits is important to understand the natural history, vulnerability and strategies used to obtain resources by the species. The aim of this study is to provide preliminary information about feeding ecology of a *Liolaemus cf. ruibali* population in San Juan Andes mountains. We analyzed and compared the diet of 12 specimens captured during April 2009 and December 2009. Each item was identified and analyzed in qualitative and quantitative sense: IRI according to volume, number and frequency was estimated. *L. cf. ruibali* presents an omnivorous diet, prey items may vary according to seasonal period. At first period, Hemiptera was "fundamental" prey, while Coleoptera and Formicidae were "secondary" and "accessory" preys respectively. The second period was different due to Coleoptera was "fundamental" prey, Formicidae "secondary" and Diptera was "accessory" prey. On the other hand, plant material consumed was not different between periods (Mann-Whitney, $P > 0.7$), being present in most stomachs in 70% of total consumption. We conclude that *L. cf. ruibali* is an omnivorous lizard, with a diet consisting invariably of arthropods and plant structures, whose items are time-varying.

163.

EB87 - USEFULNESS OF DNA BARCODES IN THE PHYLOGENY AND SPECIES IDENTIFICATION OF *Hypostomus* (SILURIFORMES: LORICARIIDAE)

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The Loricariidae family is one of the most diverse in number of genera and species in the Neotropical Region. The genus *Hypostomus* includes approximately 130 species that inhabit a variety of environments reaching much of this region. The wide distribution, species richness and the limited available information of *Hypostomus* became this genus difficult to taxonomic determination. In this sense the molecular tools allow a new focus on the systematics. Here, we analyzed the species diversity of *Hypostomus* through DNA barcodes. Sequences of the D-loop marker, whose phylogenetic information has already been checked, has been used as a control. We amplified 876bp of DNA barcodes and 547bp of D-loop markers and rebuilt the phylogeny of *Hypostomus* from each dataset. The topology of the obtained trees has been the same for both genes and the specimens of each species have differentiated by a considerable genetic distance in both cases. Thus, we demonstrated that DNA barcodes are a great tool in the species identification of *Hypostomus*, but it also contains information about the evolutionary history of the genus.

164.

EB89 - ENERGY STRATEGY OF HYLIDS (ANURA: HYLIDAE) IN PARANÁ RIVER FLOODPLAIN DURING LOW TEMPERATURE SEASON

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We studied diet and fat body (FB) development in four hylids actives during cold season (31°37'S, 60°41'W) [*Dendropsophus nanus* (Dn), *Hypsiboas pulchellus* (Hp), *H. punctatus* (Hpu) and *H. raniceps* (Hr)]. We analyze trophic niche overlap (O_{jk} : 0-1), prey volumes and FB variability among species. Frogs diet varied from the one described for warm season. Trophic overlap was high in Dn/Hpu (O_{jk} =0.89), Dn/Hp (O_{jk} =0.78) and Hp/Hpu (O_{jk} =0.67). Differences in FB weight and preys volume among species were statistically significant. Paraná River hylids sowed different strategies for securing energy supply during cold season activity. Hp, with less development of FB, feeds during winter. Dn used well developed FB and feeds little during coldest months. Finally, Hpu and Hr have a midway strategy, with the utilization of fat reserves and an intermediate feeding activity to deal with low temperature season.

165.

EB90 - PRELIMINARY DATA OF DIVERSITY OF WALKERS ARTHROPODS IN LAS FLORES GULLY, SAN JUAN

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Arthropods are the representatives of taxon diversity and evolution, and participate in key roles in the ecosystem. However there is a lack of general knowledge about their distribution and abundance. The aim of our study was to determine the diversity of walkers arthropod in the Las Flores gully, San Juan province. Pitfall traps were used to capture, which were planted during the spring-summer seasons. The traps were removed at the end of each month and collected for further analysis. In the laboratory were identified and counted the fish caught. For walkers arthropod diversity index was calculated Simpson ($D = 0.411$) and Margalef index ($I = 4.01$). The most abundant group was Hymenoptera with 22 different species were found 6 species of Coleoptera, 2 species of Orthoptera, 7 species of Araneae, 1 species of scorpion, 1 species of the Order Solifugae, 1 species of the order Phasmatodea, order Acariina 3 species and 1 species of springtail order. While the data reported so far are for a single month, the results would indicate a high diversity of arthropods to the place, also representing new records of the same presence and distribution of the species found. The high diversity of species of Hymenoptera can be attributed mainly to extreme weather conditions; high socialization ants play a critical role in ensuring the survival of individuals in the group. Future studies should consider using harvesting techniques that incorporate flying arthropods.

166.

EB91 - EFFECTS OF REPRODUCTIVE CONDITION IN DIET IN *Liolaemus parvus* (LIOLAEMIDAE) IN SAN JUAN, ARGENTINA

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Composition analysis of the diet not only provides information on the type of prey which feeds but also reports on the strategy used for feeding. The aim of this study is to provide the first data on diet of *Liolaemus parvus*. We compared the diet of this species between two periods: reproductive (PR) and post-reproductive (PPR). Thirty-three individuals have been analyzed and was calculated the relative importance index (IRI). Degree of similarity in diet composition through Sørensen index and dominance through Simpson index between periods were calculated. To determine amplitude of trophic niche Shannon-Wiener diversity index was used. Percentage of plant consumption between both periods was compared. Diet in the (PR) consist in Formicidae as "secondary" item, whereas in (PPR) Formicidae was "primary" and Coleoptera was "secondary" item. Differences in plant materia consumption with a higher percentage in the (PR) ($P < 0, 01$) were founded. There was overlap among the item-prey, Sørensen ($I_s = 0.66$) between periods. Dominance in certain items there was in reproductive period (PR). No differences in trophic amplitude for both periods were founded. *L. parvus* is generalist and omnivore species that consumes arthropods generally in both periods.

167.

EB93 - DAILY ACTIVITY OF TWO SPECIES OF LIZARDS DURING AUTUMN SEASON IN THE AUSTRAL PUNA, SAN JUAN

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Sympatric species divide resources in the dimensions: trophic, spatial and temporal of ecological niche. The aim of this work is report about the daily activities of *Phymaturus cf palluma* and *Liolaemos parvus* in the highlands of San Juan. This study was conducted in the reserve "Don Carmelo", located in the Ullúm department at 3500 meters. Two transects were constructed 300 meters long each. The transects were traveled at a rate of one hour, beginning at 9 am and ending at 19 pm. They record the frequency of individuals differentiating adult and subadult records. In addition, the air temperature recorded every 10 minutes. The frequency of both subadult and adults of both species were positively and significantly correlated with temperature environment (*P. cf palluma*: Spearman_{Subadult}: $R=0.47$; $p<0.004$; Spearman_{Adult}: $R=0.87$; $p<0.0009$. *L. parvus*: Spearman_{Subadult}: $R=0.69$; $p<0.02$; Spearman_{Adult}: $R=0.80$; $p<0.004$). The Activity of *P. cf palluma* was unimodal and *L. parvus* showed similar activity for both age groups coincide with its peak at 1 pm. Necessary more studies in the extreme stages of the annual activity to understand the use of temporary niche of these species.

168.

EB94 - Scarabaeidae PRESENTS IN LOTS OF GRASSLAND AND WEEPING LOVE GRASS (USA), SAN LUIS, ARGENTINA

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The way of greatest impact to increase livestock productivity potential of the central semi-arid region, is to increase forage productivity through the sustainable management of grassland and forage crops. Unfortunately, there are very few works analyzing regional sanitary problems for them. This work is a communication of preliminary data aiming to survey soil insects (Coleoptera:Scarabaeidae) found in fields of grasslands and weeping lovegrass. Sampling was simple random and was carried out biweekly from November 2010 to March 2011 - in San Luis province fields of dune area of grassland and "isletas de chañar" - Larvae present in each sampling unit (25 cm x 50 cm and a depth of 30 cm), were extracted and identified using the Alvarado key modified by Frana, J. (Frana, 2003) and the key for Melolontidae for Argentine (Morón, 2006). Up to now the following species present both grassland and weeping lovegrass were identified: *Anomala testaciespennis*, *Cyclocephala pútrida*, *Diloboderus abderus*, *Philochloenia bonariensis* y *Liodenys* sp.

169.

EB96 - ACRIDIDAE PRESENTS IN ALFALFA CROP AND GRASSLAND IN SAN LUIS, ARGENTINA

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The “tucuras” are potential pests for agricultural crops, pasture and natural grasslands, destroying a significant amount of forage and therefore impacting on livestock production. For the province of San Luis the following species of “tucuras” have been reported *Chromacris speciosa*, *Dichroplus pratensis* and *D. Vittatus*; a few *Spyllina spp.* y *Neopedies spp.* in 1941: *Staurorhectus longicornis*, *Scyllina variabilis*, *C. speciosa*, *D. pratensis*, *D. vittatus*, *Rhammatocerus pictus*, *Zeniopoda tarsata* in 1981. The present work was performed on alfalfa and grassland in “the pampas” western district, with the objective of making an update register of Acrid’ species. Specimens were identified using the taxonomic key from Salto (1999). The following species of the *Acrididae* family were identified: *D. elongatus*, *Baeacris punctulatus*, *Ronderosia bergi*, *D. pratensis* on alfalfa crops and on grasslands: *D. pratensis*, *R. bergi*, *D. conspersus*, *B. punctulatus*, *Allotruxalis strigata*, *C. miles*, *D. elongatus*, *R. pictus*, *Borellia pallida*, *S. longicornis*, *Z. tarsata*. The survey was carried out during the campagne 2010-11, using drag net entomology. Of 15 species observed only five of them agree with those identified in previous studies.

170.

EB99 - THERMAL TOLERANCE OF TWO SPECIES OF ANURANS: IMPLICATIONS IN GLOBAL WARMING

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Global climate change will affect more to the communities of subtropical and tropical ectotherms that communities are distributed at higher latitudes. The aim of this study is compare the thermal safety margin (TSM) and heat tolerance (HT) of tadpoles of *Rhinella spinulosa* and *Odontophrynus cf occidentalis* in an altitudinal gradient. We record the critical thermal maximum (CTmax) and selected body temperature (Tsel) of both species. Environmental temperature was obtained with the data logger (Tenv). We calculated the TSM (CTmax-Tenv) and HT (Tsel-Tenv). The TSM reported significant differences being species (Mann Whitney U = 3.87, p < 0.000006) the mean for: *R. spinulosa* = 21.8 ± 0.3°C and *O. cf occidentalis* = 15.2 ± 0.5°C. In addition, we detected significant changes in the parameter HT (Mann Whitney U = 4.15, p < 0.00001) where: *R. spinulosa* = 12.9 ± 0.3°C and *O. cf occidentalis* = 3.8 ± 0.4°C. Amphibian communities in lowland are apparently more susceptible to global warming that amphibian communities found at hightland. In this framework would be less affected by future global warming since it causes the minimum winter temperatures increase the effect being greater at low altitude.

171.

EB100 - SEXUAL DIMORPHISM IN *Phymaturus cf. palluma* (LIOLAEMIDAE) FROM SAN JUAN, ARGENTINA

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Study of sexual dimorphism can offer responses about biological meaning that implies morphological differentiation between males and females. However, these differences can be explained by selective pressures affecting on the degree of sexual dimorphism in species. For these reasons, we studied sexual dimorphism in a population of *Phymaturus cf. palluma* in Calingasta Department, San Juan Province. We measured 10 morphometric variables on 16 specimens (9 females / 7 males) and were analyzed by descriptive statistics, regression analysis and measures of central tendency. Sexual differences in variables values were tested by mean and median comparisons (Mann-Whitney test, ANCOVA), 5% significance level. *P. cf. palluma* was sexually dimorphic in 4 morphometric variables: head length, head width, radius-ulna length and tail length, all of which are higher in males than females. We compare and discuss our results with those found in other species.

172.

EB101 - TROPHIC ECOLOGY OF ANDEAN LIZARD *Phymaturus cf. palluma* (LIOLAEMIDAE) SAN JUAN, ARGENTINA

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Knowledge of trophic parameters in species helps us to know natural history and strategies employed to obtain resources. For these reasons, the aim of this study is to provide preliminary data about trophic ecology of *Phymaturus cf. palluma* in a population from Andes of San Juan. We analyzed and compared diet of 11 specimens captured during October 2009 and April 2010. Items were identified in qualitative sense. IRI was estimated according to volume, number and frequency of occurrence of alimentary items. We calculated percentage of material and nematode parasites in digestive tract. *P. cf. palluma* is an herbivorous lizard whose diet is based mainly on stems and leaves, being its consumption not different between dates. For october 2009, flowers were “fundamental item” whereas flowers and fruits were “fundamental items” for April 2010. Diet showed overlap between months (Jaccard = 0.21). We conclude that *P. cf. palluma* is an herbivorous lizard that feed vegetative and reproductive structures of plants. We compared and discuss our results with other species in terms of herbivory in lizards.

173.

EB102 - DIET OF *Rhinella spinulosa* (Anura: Bufonidae) IN CENTRAL ANDES OF SAN JUAN, ARGENTINA

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Knowledge of alimentary habits is valuable for understanding natural history, vulnerability and strategies used to obtain resources in species. The goal of this paper is to present preliminary data on trophic ecology of a population of *Rhinella spinulosa* in the Cordillera of San Juan. We analyzed and compared stomach contents in 9 individuals captured in December 2010. Preys were identified until order or family level and hierarchical IRI was calculated for each item, according to number, volume and frequency. The diet of *R. spinulosa* consists in arthropods, being Escorpionidae "secondary" item followed by Coleoptera as "accessory" item and Lepidoptera, Larvae, Vespidae, Aranae "accidental" items. On the other hand, rocks were present in 44.4% of stomachs. No correlation was found between volume and larger size of the prey with the snout-vent length (LHC) and the width of the mouth (ANB) (Spearman = $p > 0.05$). We conclude that *R. spinulosa* is a generalist amphibian with "sit and wait" trophic strategy to consume variety of terrestrial arthropods. We discuss the results with others existing for *Rhinella* gender from other latitudes and biogeographic regions.

174.

EB103 - NESTING IN A WILD POPULATION OF ANDEAN CONDOR IN THE CERRO BLANCO WILDLIFE REFUGE, CÓRDOBA

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The reproduction of Andean condors has mostly been recorded in captive birds, while the first studies of wild individuals have taken place in Chile and Rio Negro in Argentina. Our objective, therefore, is to contribute to the reproductive knowledge of wild condors in the central and extra-Andean area of Argentina where this type of information is unknown. For this purpose, a condor nest located in Cerro Blanco Wildlife Refuge in Córdoba was monitored seasonally from Oct 2008 to Dec 2010 for a total of 99 days and 452 observation hours. Courting and copulation was observed during three consecutive years in the same nest and each resulted in one egg in mid Oct, which is later in the year than has been reported in other studies. During a total of 178 observation hours, the female spent more time incubating (40%) and was present more time in the nest without incubating (5.1%) than the male (34% and 2.4%, respectively). Neither parent was present in the nest or incubated the egg 18.5% of the observed time. Two of the eggs were successful (mid Dec 2008 and 2009, respectively), and one failed on mid Dec 2010. Once the egg hatched and during a total of 274 observation hours, the male spent more time with the chick (1.7%) and fed it more times (71%) than the female (1.3% and 29%, respectively). The chick was left alone 97% of the time. Knowledge of the reproductive biology of wild condors is an emerging field which requires more studies of this type in other regions of its distribution.

175.

EB105 - MORPHOMETRIC SEXUAL DIFFERENCES IN *Liolaemus parvus* (Liolaemidae) FROM SAN JUAN

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Selective pressures acting differentially on males and females many times can be explained by consider ecological function such as reducing niche overlap and competition between the sexes. For these reason, the aim of this study is to detect morphological differences between sexes in a population of *Liolaemus parvus* in Calingasta department, San Juan, Argentina. We measured 9 morphometric variables in 50 individuals (25 males/25 females): Head length (LCA), head width (ANCA), head high (ALCA), humerus length (LH), femur length (LF), radius-ulna length (LRC), tibio-fibula length (LTF) snout-vent length (LHC) and distance between members (DEM). Dependence of each variable with LHC was tested, when these were significant ANCOVA was performed. LHC differences were tested through ANOVA. And rejection level was 5% in all cases. Significant differences between sexes in 5 morphometric variables (LCA, ANCA, ALCA, LH, and LF) were founded, being all of these in males longer than females. Nevertheless, significant differences were founded in (DEM) being in females longer than males. Our results are discussed in the intra specific competition context and ethological significance as those related to courtship and defense of space.

176.

EB110 - ASSESSMENT OF CHYTRIDIOMYCOSIS IN FERAL POPULATIONS OF BULLFROGS IN PUCHUZUM (SAN JUAN)

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The transport and release of species by human activities for biological control, food, pets, among others, has devastating effects on ecosystems and native wildlife species. Some of the impacts include species extinctions, biotic homogenization, disruption of food webs, changes in primary productivity and new vectors of diseases. The introduced bullfrog (*Lithobates catesbeianus*) is recognized as an asymptomatic vector of *Batrachochytrium dendrobatidis* (*Bd*), the fungal pathogen of amphibians, considered one of the most important emerging diseases in recent times. Given the relationship between the spreading of *Bd* and bullfrogs dispersion suggested on literature, the objective of this study was to determine the presence of *Bd* in feral populations of bullfrogs in San Juan. The sampling was conducted in the Puchuzum locality (Calingasta Department) on summer 2008 and 2010. qPCR analysis was performed to detect the presence of the chytrid fungus infecting frogs. We report a prevalence of 27.27% of the fungus in analyzed bullfrogs. The result obtained in this work represents an important contribution to assist in the elucidation of the role played by the bullfrog in the dispersion of chytridiomycosis in native anurans of this region.

177.

EB113 - DISTRIBUTION OF SPECIES OF ORCHIDACEAE IN THE PROVINCE OF SAN LUIS (ARGENTINA)

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Seven terrestrial species of the family *Orchidaceae* are cited for the province of San Luis. They are: *Sacoila lanceolata* (Aubl.), *Habenaria gourleiana* Gillies ex Lindl., *Habenaria hexaptera* Lindl., *Pelexia bonariensis* (Lindl.) Schltr., *Pteroglossaspis argentina* Rolfe, *Aa achalensis* Schltr. and *Aa hieronymi* (Cogn.) Schltr. Our objective was to estimate the geographical distribution of species in this family to "San Luis" for the purpose of making better management of these areas for the conservation "in situ" of these species. *Sacoila* is the most widespread species found in the "Sierras de San Luis", "Morro" and "Comechingones", as well as in its foothills. *Aa* and *Pelexia* have the same distribution, but the former is easier to find the second, being located on slopes and alluvial valleys. *Habenaria hexaptera* is found in the form of communities in alluvial valleys and hillsides in the "Sierra de San Luis", as *Habenaria gourleiana* not found. *Pteroglossaspis* was found in two valleys of the "Sierra de San Luis" and "Comechingones". We conclude that this family has a distribution characterized by: a) the height (above 700 m), b) tending to cold humid climate, c) high soil stoniness and/or lithosoils, d) on hillsides, valleys and plains barely grazed, which recognizes the vulnerability of these species to trampling and overgrazing.

178.

EB114 - DISTRIBUTION IN ARGENTINA OF THE GENUS *Bryconamericus* (CHARACIFORMES, CHARACIDAE)

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The genus *Bryconamericus* Eigenmann comprises about 50 species of small fishes reaching until 10 cm standard length. Among other characters, they have four teeth in the inner row of the premaxillary and a poorly varied coloration pattern. They live in lotic and lentic environments, being abundant in flooded areas formed by overflow of creeks, rivers and lagoons with dense submerged and floating vegetation. Species of the genus live from Central America to the Sauce Grande River basin (38°56'S-61°23'O) in the South of Buenos Aires Province (Argentina). Thirteen species has been reported from Argentina, occurring in environments in northwestern, northeastern, eastern and center of the Argentinean territory. *Bryconamericus iheringii* is the widest distributed species, living in all the above mentioned areas, from the Bermejo River basin to southern Buenos Aires Province, which is the southern border of distribution of the genus. This wide distribution suggests a high degree of eurytopy. In Argentina, the northern border of distribution of the genus is given by *B. iheringii* and *B. thomasi*, both occurring in the Lipeo River in the Salta Province. Misiones Province is the area with more endemics, with *B. ikaa* and *B. pyahu* reported from the Iguazú River basin, *B. agna*, *B. mennii* and *B. sylvicola* from the Paraná River basin and *B. ytu* found only in localities of the Uruguay River basin. *Bryconamericus eigenmanni* is only found in central Argentina, in Córdoba Province and *B. rubropictus* in the northwestern Salta Province. Based on available material and new surveys, we analyze in this paper the distribution of the genus, providing numerous new localities for different species and a map with the distribution area of *Bryconamericus* in Argentina.

179.

EB115 - ESTIMATION OF BIODIVERSITY OF ARTHROPODS WITH EMPHASIS ON FORMIC (INSECTA: *hymenoptera*) IN AN OPEN JARILLAL SITE OF THE NATIONAL PARK LIHUE CALEL (LA PAMPA, ARGENTINA)

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Arthropods represent the most diverse and evolutionary successful phylum, they play critical roles in the ecosystem, as pollinators agents and seed dispersion. They are also involved regulating population size, acting as biological controllers. The measurement of biodiversity allows to estimate the richness and relative of abundance of species. The province of La Pampa (Argentina) is suffering from advanced destruction of their natural environments with high probability that a percentage of its fauna would be extinguished before being studied. The objective of this study was to estimate the biodiversity of arthropods in an open jarillal site, including pasture (grasslands). Sampling were carried out from December 2008 to March 2009 on two transects of 300m each using the method of canvas over the jarillal and net of trawl on grassland, in two places: with and without disturbance (fire). The obtained material was preserved in alcohol to 80% and was analysed and settled in the subject Biology of the Invertebrate II, Faculty of Exact and Natural Sciences, of the UNLPam. The sults indicated that the orders Hemiptera and Hymenoptera were the most abundant for both sampling sites. The family Formicidae (Hymenoptera) was the most abundant during the sampling months. We watched differences in the abundance of ants species, as regards the vegetal structure of the park, since in the grasslands the most dominant specie was *Camponotus punctulatus*, while in the jarillal it was *Brachymyrmex patagonicus*.

180.

EB116 - FIRST THERMAL DATA OF SCARCELY KNOWN *Amphisbaena plumbea* (SQUAMATA: AMPHISBAENIDAE)

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Many thermal studies in reptiles are mainly based on epigeal taxa. In contrast, the thermal ecology of fossorial reptiles is poorly understood because the observations and captures are difficult. Therefore, our aim is contribute information about thermal relationships in *Amphisbaena plumbea* in lab conditions. During May-2007 the experiment was performed in a sample of amphisbaenids (N=4) placed in open-top terrarium (120x60x40 cm) with sandy-loam substrate (3cm deph) and a thermal gradient produced by a infrared lamps (250-400 W) in a extreme of terrarium. We obtain operative temperatures (substrate: T_s | air: T_a) and body temperature (T_b). Temperatures were taken using ultra-thin catheter thermocouple 5mm inside the cloaca. Operatives temperatures and temperatures of each amphisbaenids was obtained every 10 m for 5 hr. *A. plumbea* was tigmotherm obtaining heat by substrate contact (Spearman, T_s : $r_s = 0.80$, $P = 0,00001$). T_b mean was 28.5 (DE: 3.01°C), this value is remarkable higher than those reported for other amphisbaenids. We conclude that these fossorial reptiles are capable of regulating body temperature in similar manner those epigeal reptiles. We believe that in field conditions may thermogulates by vertical movements from cold deep areas to warm top areas of the soil.

181.**EB117 - THERMOREGULATION AND THERMOREGULATORY EFFECTIVENESS IN *Phymaturus cf. palluma***

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Given their dependence on temperature and solar radiation, the lizards constitute an interesting biological model for determinate individual responses as the ultimate adaptation modulators and, therefore, it is possible to determinate different species life histories and physical environment aspects-related. Therefore, our aim is contribute to knowledge of thermal relationship and their variations in *Phymaturus cf. palluma* that inhabits on harsh environments. Study carried out above 3000 masl, in highlands of Calingasta, San Juan during spring-summer-2008 and fall-2009. Environmental, micro environmental and operative temperatures were studied, and body and selected temperatures in lizards were analyzed through Hertz method. Analysis of temperatures suggest that *P. cf. palluma* present both tigmothermic and heliothermic strategies of thermoregulation, being a “good” thermoregulator regard the thermal resources in the environment, but rarely reaches the preferred temperatures, while Hertz index suggest that *P. cf. palluma* is a moderate thermoregulator. We compare and discuss our results in evolutionary sense with others members in the genus.

182.**EB118 - TEMPORAL AND SPATIAL ACTIVITY IN *Phymaturus cf. palluma* (LIOLAEMIDAE) IN THE SAN JUAN ANDES, ARGENTINA**

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Temporal and spatial activity constitutes two of three dimensions of the resources partition that the species may segregate and explains the distributions and number of ectotherms in variable environments. For these reasons, the aim of this work was to establish space-temporal use patterns and its variations in a population of *Phymaturus cf. palluma* in the Andes of San Juan. Fieldwork was carried out during spring-summer-2008 and fall-2009. Individuals were counted during daily period (10:00-18:00hr) and hour, sex, age group and microhabitat in 6 categories were recorded. Data were analyzed using polymodal decomposition and goodness of fit test (χ^2) through “space-temporal uniformity of utilization” null hypotheses. *P. cf. palluma* presents unimodal activity, and its may modify according the environment conditions. This species is a microhabitat user specialist, capable to choose between two different types of basaltic rocks and can switch between one and the other according thermal requirements in response to micro environmental and seasonal variation.

183.**EB121 - DETERMINATION OF SEXUAL DIMORPHISM IN *Rhinella arenarum* BY MORPHOMETRY GEOMETRY**

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The differentiation morphological between males and female sexually mature is a common feature of animals, reflects the adaptation of both sexes to their different reproductive roles. In general, attributes that enhance the ability of males to locate, attract and retain female are favored by sexual selection. In this paper, we evaluated the cranial morphological variation between male and females of *Rhinella arenarum*. Dorsal photograph was taken of each skull and used nine landmark processed using software package Tps. In males, all the landmarks are extended to the anterior skull producing a more sharpened it. In particular, the landmark that show a greater contribution to the variation is the number 5 (46.6%) located in the squamosal bone and 6 (45.9%) present in the square-jugal. Both related to the jaw suspension. Specifically, in the male these points are farther apart. In other ectotherms, differences in skull shape have been attributed to diet, predation, and reproductive behavior of individuals. The diet composition of *R. arenarum* is same in both sexes. Based on the above and assuming that both sexes are subject to similar predation pressure is likely that the observed difference is based on the further development in males of the muscular structures of the lower jaw. This structure is related to the presence of vocal sac in males, implicated in the acoustic communication in the reproductive period.

184.**EB122 - REDESCRIPTION OF THE TADPOLE OF *Pleurodema nebulosum* (ANURA: LEIUPERIDAE)**

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The specie *Pleurodema nebulosum* distributes in arid areas of western Argentina, from Catamarca province to Río Negro. The objective of the work was to re-describe this tadpole for which 10 specimens were used in stage of development 37. They were collected in temporary ponds, in Sarmiento department, San Juan. The total length is 20.7 ± 1.65 mm. In lateral view the body is gently depressed in the anterior ventral portion. In dorsal view is ovoid and elongated. The snout is rounded and sharp in dorsal view, rounded in lateral view. Nostrils circular and in dorsal position. Rounded eyes, small and dorsolateral, visible in both dorsal and lateral view. Spiracle simple, short, sinistral, in almost ventrally position located in the anterior portion of the body, visible in lateral view with the opening directed toward the left flank, with an absent opercular wall of the tube. The tail represents 58% of total length, and its final portion is sharpened. The dorsal fin (DF) is slightly larger than the ventral fin (VF) with an average mean of $DF/VF = 1.55 \pm 1.13$. Vent tube medial, attached to the ventral fin. Oral disc anteroventral. Labial tooth row formula 2 (2)/2. The present study extends the knowledge of the tadpole of *P. nebulosum* allowing unmistakable identification.

185.

EB124 - ANALYSIS OF ENVIRONMENTAL PREFERENCES OF MATURE AND IMPREGNATED SHRIMP FEMALES *Pleoticus muelleri* (Bate, 1888) IN SAN JORGE GULF, ARGENTINA

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The relationship between the density values of mature and impregnated females of the Argentine red shrimp *Pleoticus muelleri* (Bate 1888) and the environmental variables was analysed. Data came from the research cruises of the Instituto Nacional de Investigación y Desarrollo Pesquero (INIDEP) carried out during November – December 2008 and January – February 2009 in San Jorge Gulf (Argentina). The biological variables considered were the density values of mature and impregnated female shrimp, whereas the environmental variables were depth, bottom water temperature and salinity. Preferential environmental values to mature and impregnated females were calculated and generalized additive models and general linear model were applied. Impregnated females were observed in lower depth and higher bottom water temperature ranges than mature females. In January-February 2009 could establish a significantly increased relationship between mature females and bottom water temperature. The importance of environmental factors such as water temperature, availability of food in the environment and the photoperiod for growth, ovarian development and maturation and beginning of spawning of aquatic and terrestrial crustaceans is well known and studied. The water temperature, especially its increment, to induce the maturation process and spawning in captivity conditions has been widely studied for tropical waters penaeid prawns. The fact that the largest concentrations of impregnated females were found in coastal areas may be related to the type of bottom and the presence of frontal systems favourable for larvae and juveniles development.

186.

EB125 - INTEGRATED ASSESSMENT OF RIVER WATER QUALITY USING BIOMARKERS AT DIFFERENT LEVELS OF BIOLOGICAL ORGANIZATION

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The aim of this study was to evaluate the quality of the lower-middle basin of Suquia River integrating information from different biomarkers. We selected 3 sites along the basin (before Cordoba city, and two once downstream from the city). It was determined a water quality index (WQI), heavy metals and pesticides. The biomarkers used were selected at different levels of biological organization: molecular level (activity of antioxidant enzymes), tissue level (liver and gill histopathology) and community level (variations in fish assemblages and estimation of a biotic index). Molecular and tissue levels were analyzed using *Jenynsia multidentata* as fish bioindicator. The WQI varied between sites. Some heavy metals and pesticides exceeded the permissible limits for rivers. Molecular and histological biomarkers showed different sensitiveness and reflected the same trend registered for water quality conditions. Fish species number, diversity and the BI decreased at the most polluted site located downstream Córdoba city. This urban center together with the effluents from the waste water treatment plant exerts a negative impact on the basin. The present work provides specific and complementary information and constitutes a comprehensive and effective strategy to evaluate the quality of water resources, potential sources of pollution and their effect on biota.

187.

EB126 - FUNCTIONAL GROUPS OF INSECTS AND HABITAT CHARACTERISTICS IN LENCTIC WATER BODIES IN CÓRDOBA, ARGENTINA

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Environmental modifications due to urbanization, such as vegetation removal, soil compaction and impermeabilization, septic water management, may affect the presence of insect fauna and predator colonization in urban water bodies. There are few studies assessing insect communities in Argentine urban lentic bodies that are potential larval habitat of disease vectors such as mosquitoes. Relations between presence of aquatic insect functional groups and water body characteristics were explored in Córdoba city. Entomological samples were collected weekly (December 2009 to March 2010) in 17 water bodies (ditches, ponds, canals), and their environmental characteristics were recorded (plant type and cover, substrate, temperature, pH, depth, etc.). Specimens were fixed and stored in 80% ethanol, counted, identified and assigned to one of 4 functional groups based on their prevailing adaptations for food acquisition (generalists, collectors, scrapers, predators). Out of 12,900 specimens, 98.1% were Diptera (mostly mosquitoes), 1.3% Odonata, 0.3% Heteroptera and 0.3% Coleoptera. Correspondence analysis showed that the presence of predators was related to flooded grasses and weeds, while generalists and collectors were related to emergent plants. Scrapers and predators (except Libellulidae) were associated with intermittent water (<15days of water permanence), while generalists were associated with longer permanence water. Collectors, Chironomidae and Stratiomyidae were associated with intermittent water bodies, Psychodidae and Syrphidae with permanent water and Culicidae did not show a clear pattern regarding water permanence. In conclusion, these results suggest that the presence of vegetation combined with intermittent water permanence sustain a higher aquatic insect diversity in lentic urban water bodies of Córdoba city.

188.

EB127 - *Aedes aegypti* USES WATER HOLDING TREE HOLES AS LARVAL HABITAT IN ORÁN, SALTA PROVINCE

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Aedes aegypti L. is the main vector of dengue worldwide. Periodic outbreaks of dengue are recorded in Argentina, and Salta is one of the most affected provinces. Since there are no available vaccines, prevention is based mostly on vector control. Larval habitats of this mosquito in South America are mainly artificial containers. Recently larvae and pupae were reported in tree holes in Aguaray, Salta province. Knowledge of the use of natural containers by *Ae. aegypti* is relevant from an epidemiological and vector control point of view, and also for a better understanding of biological invasions because it is an exotic species. Trees (1185) in public areas of Orán city were surveyed between January and April 2011. Tree species, presence and characteristics of tree holes were recorded. Every three weeks holes were visited, presence and volume of water were recorded, and larvae and pupae were collected with an aspirator. 3.46% of the trees had water holding holes; 73.1% of them had larvae, mostly (98.44%) *Ae. aegypti*. The average number of larvae (L) per hole was 60 (range 1-578), and 25% of the holes had 50 or more larvae. Average density was 8 L/10ml (range 0.36-595); density of 38% of the positive samples was 5 or more L/10ml. The trees where larvae were more frequently found were *Delonix regia*, *Bauhinia* sp. and *Jacaranda* sp. Water holding tree holes should be taken into account as larval habitat or re-infestation sources during vector surveillance and for vector control activities.

189.

EB128 - AIRBORN FUNGAL SPORES AND POLLEN CONTENT IN THE ATMOSPHERE OF THE CITY OF SAN LUIS, ARGENTINA

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The aim of this paper is to present the first annual aerobiological study of the atmosphere of the city of San Luis. Air samples were taken using a Hirst-type volumetric spore trap sampler (Lanzoni, VPPS 2000) for a period of a year (August 2010-July 2011). The production of aerobiological particles showed fluctuations depending on the flowering seasons of the taxa and on the meteorological parameters. The minimum concentrations were related with precipitation and the peaks of maximum concentration were related with the mean daily temperature and solar irradiance data. During this period Cupressaceae/Taxaceae, Amaranthaceae/Chenopodiaceae, *Celtis tala* and *Larrea divaricata* pollen were the most common pollen. *Alternaria* and *Cladosporium* were the more important in the spore content. The increase of aerobiological particles in September 2010 and February 2011 was associated with the beginning of consultation for allergies. This scientific research reveals a great diversity of aerobiology particles in this outdoor environment, showing a strong dominance of exotic pollen.

190.

EB129 - CONTRIBUTIONS ON DIET IN THE ENDEMIC LIZARD *Liolaemus eleodori* (IGUANIA: LIOLAEMIDAE), SAN JUAN, ARGENTINA

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Knowledge of the trophic parameters is very important for maximizing actions herpetofaunística biodiversity conservation and environment. The aim of this work is to present the first data trophic *Liolaemus eleodori* a population from San Guillermo National Park, Iglesia, Department and determine the existence of temporal variations in the diet. Based on the study of 10 individuals (December 4, 2004 and February 6, 2005) analyzed the stomach contents to identify prey order or family level. We assessed the numerosity and relative frequency of occurrence of different food categories and their respective volumes. For the contribution of each food category was applied to the diet of Relative Importance Index (IRI). It was found that the most important food categories were flowers (IRI = 3225), followed by Coleoptera (IRI = 1168) and Hemiptera (IRI = 832). We observed the presence of two species of nematodes in 3 individuals in different parts of the digestive tract, one in the stomach and the other in the last portion of the digestive tract. From the results it is concluded that *L. eleodori* presents an omnivorous diet type.

191.

EB130 - PRELIMINARY DATA OF A TROPHIC *Phymaturus* POPULATION Puna (*Iguania: Liolaemidae*) THE RESERVE PROVINCIAL SAN GUILLERMO, SAN JUAN, ARGENTINA

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Diet is a fundamental aspect of the biology of an organism, trophic niche is considered along with the temporal and spatial niches of the main ecological dimensions of any animal species. The aim of this study is to present the first data *Phymaturus* trophic puna Provincial Reserve San Guillermo, Department of the Church. The specimens were captured during two periods, December 2004 and February 2005. We analyzed the contents of the digestive tract of 10 specimens. We calculated the number, volume and frequency of occurrence of food items. IRI index was applied to describe the diet. We identified the following items: fruits, flowers, seeds and plant material (stems and leaves). The results allow to suggest that the diet is composed mainly of fruits and secondarily by flowers, seeds, stems and leaves. 60% of stomach contents corresponded to plant material and the remaining 40% consisted of nematodes found at the large intestine in 100% of the sample. We conclude that *P. Puna* herbivore - frugivorous and discuss the presence of parasites in relation to herbivory.

192.

EM5 - NEONATAL TREATMENT WITH BPA ALTERS THE CONTENT OF E₂, P₄ AND GnRH-I IN OVARIES OF ADULT RATS

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BPA is a xenoestrogen used in the manufacture of polycarbonate plastics, epoxy resins and polystyrene. We have previously shown that animals exposed neonatally to BPA have altered serum levels of sex hormones in adulthood: increase E₂ and T and decreased P₄. In this study, Sprague-Dawley female rats were injected daily sc from the day of birth to 10 postnatal with BPA [500µg/50µl (B500) or 50µg/50µl (B50) in castor oil], or oil as a control (C), and sacrificed in the morning of estrus at 110-120 days of age. We evaluated the ovarian content of E₂, P₄, T and GnRH-I by RIA. We found higher ovarian E₂ content (pg/mg ovary: C: 2.11±0.55, B50: 12.00±4.28 (p<0.005), B500: 29.76±7.85 (p<0.0005), n= 8-14) and lower P₄ (pg/mg ovary: C: 531.28±29.57, B50: 351.59±54.81 (p<0.05), B500: 263.44±56.84 (p<0.005), n= 7-12). T content was not significantly different among groups (pg/mg ovary: C: 17.54±4.57, B50: 11.13±3.09, B500: 8.47±1.72, n= 6-13). In B500, GnRH-I content was increased (pg/mg ovary C: 0.54±0.05, B50: 0.69±0.07, B500: 0.97±0.07; B500: p<0.0005 vs. C, p<0.05 vs. B50, n= 9-11). We conclude that in adult neonatally-BPA treated rats ovarian E₂ content, increased, and P₄ content, decreased, are altered coinciding with the serum levels previously reported. Ovarian T content did not change, unlike serum levels, suggesting changes in metabolism and/or origin of this steroid. In addition, BPA-treatment increased the ovarian GnRH-I content. These results suggest that neonatal exposure to BPA produces a clear alteration in ovarian GnRH-I and steroidogenesis in adulthood.
 CONICET-UBA-ANPCYT.

193.

EM6 - NONINVASIVE HORMONE MONITORING IN CHINCHILLA: RANKING THE STEROID EXCRETION IN RODENTSBusso JM^{1,3}, Ponzio MF^{2,3}, Fiol de Cuneo M^{2,3}, Ruiz RD^{2,3}.¹Fac Cs Ex Fis y Nat, ²Fac Cs Méd, Univ Nac Cba, ³CONICET.

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Whether the steroid hormone is primarily passed into urine or feces is species-dependent, and this information is useful to a precise endocrine monitoring; in excretes metabolite concentrations are frequently several times higher than the concentration in blood. Wild *Chinchilla lanigera* is threatened (CITES I), although, domesticated form is widespread in breeding farms. Our experience on its reproductive endocrinology rises from radioinfusion studies of testosterone (in males), estradiol, progesterone (in females) and corticosterone (in both sexes). Testosterone, estradiol and corticosterone metabolites were excreted mainly by urine (84.7±4.2%, 71.7±12.1%, 86.9±0.07% respectively), while progesterone was detected in equal amounts of both excretes. Therefore, we proposed that the urinary route is preferential in *Hystricognathi* (chinchilla and guinea pigs; currently considered cavy-like forms) while in the *Sciurognathi* (squirrel and mouse-like forms, formerly considered *Sciomorpha* or *Myormorpha* suborders). This hypothesis was tested by matching our results and those reported for other rodents subjected to radiolabeled infusions. We consider that this proposal is relevant when the endocrine studies are faced in a new species. Supported by grants from SECyT-UNC, CONICET, FONCyT.

194.

EM18 - EFFECT OF FASTING ON GROWTH PARAMETERS AND HEPATIC HISTOLOGY IN *Cichlasoma dimerus*

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Fish can withstand long periods of food restriction and in some species fasting has a direct effect on somatic growth rate. The purpose of this study was to analyze possible variations of the somatic growth pattern and organosomatic indexes (OI) in response to food restriction. Two groups of fish were fed (F, control) or unfed (UF) for 21 days while recording weight and length twice a week. After this time, blood samples were collected to quantify cortisol and glucose levels and then sacrificed. Liver, spleen and gonads were extracted to determine the OI. Somatic growth parameters were reduced in the UF fish: total and standard length ($p < 0.01$), and body weight ($p = 0.0001$). No differences were detected in the condition factor [$(P/L^3) \cdot 1000$], glucose and cortisol levels ($p > 0.1$). The hepatosomatic index was reduced in UF fish ($p = 0.0001$) and the hepatocyte area/hepatocyte number ratio per selected area (I) smaller ($p = 0.0037$). The exocrine pancreas cells were not as conspicuous in UF as they were in F group. In conclusion: 1) in *C. dimerus* growth is directly affected by the nutritional status, 2) it is proposed that I ratio might be an excellent nutritional status index unlike those classically employed.

195.

EM21 - INFLUENCE OF HYPOTHYROIDISM ON LIPID METABOLISM IN THE HEART OF VIRGIN RAT

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Thyroid state influences cell and plasma lipid composition. The aim of this work was to determine the effects of hypothyroidism on lipid metabolism in the heart of female virgin *Wistar* rats, weighing 150-180g. Hypothyroid state was induced by administration of 6-n-propyl 2-thiouracil (100 mg/L) in the drinking water, for 30 days. Age-matched euthyroid rats were used as controls. Serum lipids were measured by Wiener kits and heart lipids by spectrophotometric assays after lipid extraction (Folch method) and separation of the different lipid fractions (thin-layer chromatography). mRNA expressions of peroxisome proliferator-activated receptor- α (PPAR α) and sterol regulatory element binding protein 1c (SREBP-1c) were determined by RT-PCR, using β -actin as internal control. Heart fatty acid composition was determined by gas-liquid chromatography. Hypothyroid state induced an increase of serum cholesterol and LDLc, a decrease of serum and heart triglycerides (TG) ($p < 0.01$), and an increase of the heart fatty acid unsaturation index (UI). Heart mRNA levels of PPAR α was decreased ($p < 0.01$), without change in SREBP-1c, in relation to control. Hypothyroid state alters serum and heart lipids content, and decreases the expression of PPAR α , which is involved in the regulation of heart energetic balance. In addition, the high UI suggests a low protection against fatty acid oxidation in heart.

196.

EM24 - THE - 629C6A POLYMORPHISM IN THE CETP GENE AND ITS RELATIONSHIP WITH METABOLIC SYNDROME IN FIRST DEGREE RELATIVES OF DIABETIC PATIENTSDella Vedova MC¹, González II¹, Siewert SE¹, Filipuzzi S², Ojeda MS¹.¹Facultad de Química, Bioquímica y Farmacia, Universidad Nacional de San Luis. ²Hospital Provincial de Santa Rosa, San Luis. Prov. San Luis. E-mail: msojeda@unsl.edu.ar

The cholesteryl ester transfer protein (CETP) plays a key role in the metabolism of the HDL particles. One of the polymorphisms studied is the -629C→A, which modulates the transcriptional activity and the CETP levels. It also is a determinant of the plasmatic HDL-c levels. The aim was study the genotype frequencies of the -629C→A polymorphism in the promoter region of CETP gene and its relation to lipid parameters in first-degree relative from patients with T2DM with (w) and without (wo) Metabolic Syndrome. We calculated BMI, HOMA-IR and HOMA- β cell. Biochemical parameters determined were: blood glucose, insulin levels, triglycerides, total cholesterol, HDL-c, LDL-c. We analyzed the -629C→A polymorphism from the promoter by ASO-PCR technique. No significant difference was found among the CC, CA and AA genotypes between (w) and (wo). The levels of HDL-c in AA carriers, was 55.6±6.8 mg/dl in (wo) and 38.3±2.6 mg/dl in (w) ($p < 0.001$); triglycerides values in the AA genotype were 145±12 mg/dl and 215±22.5 mg/dl ($p < 0.05$) for (wo) and (w), respectively. The -629C→A polymorphism in individuals (w) carriers of AA genotype could have a negative effect on HDL-c levels, probably due to high plasma triglycerides levels.

197.

FA3 - CHRONIC STRESS, LIPOPROTEIC METABOLISM AND CARDIOVASCULAR RISK IN FEMALE RATS*Scoppa H, Niebylski A, Bensi N, Bianco M, Binotti S, Puebla M, Torres C, García M, Gauna HF.**Fisiología Animal. FCEFYN. UNRC. Río Cuarto. Cba. E-mail: hscoppa@exa.unrc.edu.ar*

Stress induce metabolic changes in which leads to the stress and atherogenic markers induced by chronic immobilization in female rats. Controls and stressed by immobilization (IMO) during 180 days (2 h per day, 3 times a week) female Wistar rats were used. Blood and aorta samples were extracted. Total cholesterol (Chol), triacylglycerides (TAG), apolipoprotein B (apo B), lipoprotein (a) (Lp (a)) and non-HDL cholesterol (CNHDL) were determined. Lipoproteins were separated by ultracentrifugation and Chol and TAG were quantified. Reactive substances to thiobarbituric acid (TBARS) and nitrotyrosine in aorta were measured. Stressed rats showed greater values of Chol in VLDL and LDL and of TAG in VLDL. ApoB, Lp (a) and CNHDL increased in stressed rats. The increase of TBARS and the positive reaction to nitrotyrosine made it evident the presence of oxidative stress. In conclusion long lasting chronic stress produces a great increment in the atherogenic patterns of plasma, increases the oxidative stress and interfere in the composition of lipoproteins involved in the regulation of lipoproteic metabolism and in the atherogenic process.

198.

FA6 - BLOOD PRESSURE, NATRIURESIS AND OXIDATIVE STRESS IN RATS WITH A GENETIC PREDISPOSITION TO HYPERTENSION AND UNDER STRESS*Torres C, Binotti S, Stagnoli S, Bensi N, Gauna H, Niebylski A.**FCEFYN. UNRC. Río Cuarto. Córdoba.**E-mail: aniebylski@exa.unrc.edu.ar*

The aim of this work was to investigate the blood pressure, natriuresis and oxidative stress in rats with a genetic predisposition to hypertension and subjected to chronic stress. We used 2 groups of male rats: F1 derived from Wistar mothers x spontaneously hypertensive fathers (F1) and male Wistar (W). Half of the animals in each group were immobilized (IMO) 1h/day for 15 days and the rest remained as control (C). On day 15 systolic (SBP), diastolic (DBP) and mean (PAM) blood pressure were recorded. Blood was collected and the heart, kidneys and adrenals were extracted and weighted. Renal malonyldialdehyde (MDA), superoxide dismutase (SOD) activity and plasma total nitrites were determined. F1 rats showed an increase in the adrenals, kidneys, heart, SBP and MAP in response to stress. MDA levels were higher and nitrites lower in F1 than W rats. No changes due to IMO or differences in the activity of SOD were observed. F1 rats showed a tendency to excrete less sodium than W. In W rats, stress increased adrenals, heart and PAS to a lesser magnitude than in F1. IMO increased MDA and nitrite and decreased renal sodium excretion. The largest increase of adrenals and PAS shows a high reactivity to stress in F1 rats. The major heart size reflect the heart's response to the increase in BP. Oxidative stress and sodium excretion would not be involved significantly in this increased stress response in rats F1.

199.

FA7 - APOCYNIN EFFECTS ON BLOOD PRESSURE IN OLD RATS SUBJECTED TO CHRONIC STRESS*Binotti S, Torres C, Cavallera M, Scoppa G, Bensi N, Gauna H, Niebylski A.**FCEFY y N-UNRC. Río Cuarto- Córdoba.**E-mail: sbinotti@exa.unrc.edu.ar*

Aging is characterized by increased prevalence of hypertension and decreased endothelium dependent vascular relaxation-mediated by NO. O_2^- is a major determinant of NO bioavailability. The aim of this work was to evaluate the NADPH oxidase participation in the increase of blood pressure in aging rats in response to chronic stress. 8 months old male Wistar rats received a daily bolus of 30mg/Kg of NADPH oxidase inhibitor, Apocynin (n=18) or the solvent of the drug (S) (n=18) for 15 days. Twelve animals of each group were subjected to immobilization stress (IMO) and the other remained as control (C). The systolic (SBP), diastolic (DBP) and mean (MBP) blood pressure in response to stress and after 6 hs of recuperation (R) were registered by cannulation of the carotid artery. Renal superoxide dismutase (SOD) activity and malondialdehyde (MDA) as thiobarbituric acid-reactive substances were measured. Total serum nitrites levels were determined by the Griess reaction. SBP, DBP and MBP increase in all stressed rats. MBP remained high at 6 hours post IMO. Apocynin prevented the increase in SBP, DBP, MBP and MDA in response to stress. There were no changes in renal SOD activity. IMO decreased while Apocynin increased serum nitrite levels. The blockade of BP increase in response to IMO in aged rats with Apocynin could be due to decreased oxidative stress and increased NO bioavailability.

200.

FA8 - EFFECTS OF FAT DIET ON THROMBOGENIC RISK MARKERS IN STRESSED RATS*Bianco M, Niebylski A, Scoppa G, Bensi N, Gauna H.**Fisiología Animal. FCEFYN. UNRC. Río Cuarto, Cba. E-mail: mbianco@exa.unrc.edu.ar*

The objective was to evaluate the effects of a high-fat diet (HF) on the lipid profile, plasma viscosity, osmotic erythrocyte fragility and the hemostasis in stressed rats. HF Wistar rats were fed with a fat supplement for 12 weeks and another group fed standard diet (SD). Half of each group was stressed by immobilization (IMO) for 14 days. HF controls (HFC) and stress (HFS), SD Control (SDC) and stress (SDS) groups were formed. On day 14 plasmatic total cholesterol (Col), triglycerides (TAG), HDL, LDL cholesterol (HDL-Chol; LDL-chol), viscosity (η), and coagulation time (CT), haematocrit (H), minimum (RGMin) and maximum (RGMax) erythrocyte resistance and plasma proteins (P) were measured. Col and TAG increased in HF and only increased in SD in response to IMO. IMO increased HDL-chol in SD and decreased this lipoprotein in HFS group. All HF rats showed high LDL-chol. The η was higher in IMO and HF rats. No changes in P levels were observed. H increased only in IMO rats. TC decreased in HF diet and in IMO rats. Low RGMax and RGMin were observed in the HFC group. IMO increased RGMin in both diets and RGMax only in DS. The increase in Col, TAG and LDL-chol associated with increased η , erythrocyte fragility and TC reduction in HF, show an increased thrombogenic risk, but not a synergistic effect with the IMO induced changes in lipemia, coagulability and erythrocyte fragility.

201.

FA9 - FREE RADICAL EPR DETECTION IN *Pleoticus muelleri* EXPOSED TO NITRITE AND ITS INTERACTION WITH DIETARY CAROTENOIDSDíaz AC^{1,2}, Velurtas SM¹, Cuartas EI¹, Fenucci JL^{1,3}.¹UNMdP. Funes 3350, Mar del Plata. ²CIC. ³CONICET.

In crustacean high levels of nitrite affect the health and survival of animals. The carotenoids function as an antioxidant, astaxanthin (Axt) is the predominant carotenoid in penaeids, which must be supplied in the diet. The objective was to investigate the interaction between concentration of Axt in diet and antioxidant response to exposure to nitrite on *P. muelleri* postlarvae fed with three diets supplemented with 0, 100, and 300 mg Axt/kg diet. The shrimp were obtained from hatchery-raised postlarvae. Diets were tested in three replicates groups of 90 individuals each, during 30 days. Subsequently, acute toxicity tests were carried out exposing animals at 0, 20, 40, and 80 mg/l of nitrite. Antioxidant activity is quantified with radical 2, 2-diphenyl-1-picrilhidracilo (DPPH), using EPR. The 96-h LT₅₀ values of nitrite were 62.97, 93.98 and 93.93 mg/l for shrimps fed to 0, 100 and 300 mg astaxanthin/kg diet. All analyzed homogenatos showed protective capacity as evidenced by the ability to react with the DPPH. A higher percentage of DPPH decay time was observed in specimens kept at 80 mg/l NaNO₂. The highest protective activity was found in the shrimp fed C100 and C300 (50% of DPPH remnant 10 minutes), compared to the diet control (72%). We conclude that in postlarvae the addition of Axt in the diet increases the survival and the production of protective substances.

202.

FA10 - METABOLIC AND HISTOLOGICAL EFFECTS OF THE NITRITE TO DIFFERENT SALINITIES ON SHRIMP *Palaemonetes argentinus*Espino ML¹, Díaz AC^{1,2}, Cuartas EI¹.¹FCEN, UNMdP. Funes 3350. Mar del Plata. ²CIC.

The aim of this work was to evaluate the effects of the nitrite on morphology and physiology of *P. argentinus*, at different salinities. Shrimp collected from Los Padres lagoon were maintained to three salinity treatments: 0; 6 and 12‰ with concentrations of nitrite between 0 and 300mg/l at intervals of 20mg/l. Two shrimps from each treatment were subjected to routine histological techniques. To metabolic parameters (glucose, protein, cholesterol, and triglycerides) were determining in whole shrimp. In gill the salinity caused edema of the epithelium, nuclei pyknotic, and destruction of cuticle. Nitrite caused disorganization of the epithelium, thickening and disruption of the cuticle in animals kept in freshwater, the shrimp acclimated to 6‰ and 12‰ showed collapse of the epithelium, infiltrated cuticle and loss of the overall structure of the lamella. In the hepatopancreas, the salinity induced retraction of the tubular epithelium and loss of cellular identity. Because of the increased of nitrite in the shrimp kept in freshwater, the cytoplasm protrude into the tubular lumen, at 6‰ was observed deterioration and lack of B cells and at 12‰ no effect was observed. All metabolic variables exhibited a high degree of correlation with the increment of nitrite in all salinities, except proteins in tests with freshwater. In *P. argentinus* salinity affects gills and hepatopancreas histological structure; however the hiperosmotic condition reduces nitrite toxicity.

203.

FA11 - CLIMATE CHANGE, FEEDING AND BLOOD ANTI-OXIDANTS IN THREE ANTARCTIC PENGUIN SPECIESDi Fonzo C¹, Zappala C¹, Rosa GA², Archuby D¹, Ansaldo M^{1,3}.¹IAA-DNA, ²DBBE, FCEyN, UBA. ³Universidad JF Kennedy.

Pygoscellis antarctica (Chinstrap), *Pygoscellis papua* (Gentoo) y *Pygoscellis adeliae* (Adelie) are exposed to severe environmental conditions (extremely low temperatures and high UV radiation) that can generate oxidative stress. Superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) and glutathione S-transferase (GST) activities, as well as reduced glutathione (GSH), lipid peroxidation (TBARS) and protein oxidation (PO) levels, were measured in blood samples from adults and chicks. The adult penguins had SOD and CAT activities higher than the chicks, with concordant responses between the activities of both enzymes. The TBARS and PO responses may be associated with the preference of krill in the diet of Chinstrap and Adelie, while Gentoo penguins complete their diet with fish. The GSH content showed that chicks exhibited higher levels than adults, which agree with the blood GSH level patterns observed in the ontogeny of other animal species. The GST activity negatively correlates with GSH levels, meaning that the latter is used as a substrate in conjugation mechanisms to deactivate peroxides. Only Papua showed differences in GPx activity, which may be associated with the variety of the diet. The analyzed biomarkers are proposed as useful tools for assessing the global Climate Change since that the marked variations registered from year to year, for longer periods of thaw, bring new stress factors from changes in salinity, temperature and food availability for these animals that are at the top of the Antarctic food chain.

204.

FA12 - SEASONAL ANALYSIS OF STEROID-INDUCED MATURATION IN *Rhinella arenarum* OOCYTESArias AJ¹, Zelarayan LI¹, Benitez L², Bühler MI¹.¹Instituto de Biología-INSIBIO. Fac. de Bqca, Qca y Farmacia, UNT. Chacabuco 461. Tucumán. ²Fac. de Ciencias Naturales e IML. UNT. M. Lillo 205. Tucumán. E-mail: lzelarayan@bfqf.unt.edu.ar

Amphibian ovarian oocytes do not usually resume meiosis unless stimulated with steroids. However, denuded *Rhinella arenarum* oocytes undergo spontaneous maturation when cultured *in vitro* without added hormones. The oocyte maturation has been best studied in *Xenopus laevis*, where androgens were the most potent promoters of this process and the most abundant steroid detected in the serum during ovulation. In this report we studied the seasonal role of testosterone (T) in *Rhinella arenarum* oocyte maturation comparatively with progesterone (P4) during three periods of the year: low (PB), high (PA) and high response whit spontaneous maturation (PAE). Follicles were harvested from *Rhinella arenarum* females during different periods: PB, PA and PAE. Totally grown follicles were selected and incubated for 20 hr with P4 or T (10-5-10-8 M). Meiosis resumption was scored by germinal vesicle breakdown (GVBD) after 20 hr of culture. Data were analyzed by ANOVA and Student's t test. Our results showed that P4 and T were capable of promoting maturation *in vitro* in *Rhinella arenarum* oocytes in a doses dependent manner, but the biological response was higher with P4. The results of ANOVA were significant (p < 0.05) for both hormones in all doses, but during PAE oocytes showed a great variability. The results of Student's test (GVBD to each doses) the minimum response occur during PB to both hormones in all doses (p < 0,01). No significant differences were found to P4 for all doses during PA and PAE. In conclusion, the present study suggests that in *Rhinella arenarum*, in a different manner to others amphibian, P4 has an important role than T during oocyte maturation.

205.

FA13 - NEONATAL STRESS INDUCES CHANGES IN THE β -GALACTOSIDASE (β -GAL) DISTRIBUTION IN THE RAT EPIDIDYMIS

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Early experiences induce long-lasting changes. As previously described, neonatal stress (NS) induces resistance to a novel stress, increases socialization, dominance and lordosis in adult rats. In this work we abord the influence of this model on the male reproductive tract, to determine if reproductive function was also affected. NS group (n=16), was exposed to daily sessions of different stressors (postnatal days 2 to 15). Another group (Sep, n=20), was daily separated from the dam for 10 min. Control group (n=23) was kept under standard conditions. In adulthood 50% of rats were submitted to a novel aleatory stress (15 days), and 50% remained undisturbed. Animals were killed by decapitation, and testis, epididymis and adrenal glands were removed. A similar stress-induced adrenal hypertrophy was observed in all groups. Testis weight and epididymal sperm count were not affected as well. However, NS and Sep treatments caused a rearrangement in the epididymal β -gal distribution, with a significant increase in the sperms and proportional decrease in the fluid. This could be explained by an increase in the affinity for the enzyme. Adult stress did not affect β -gal distribution. The higher content of β -gal in sperm of NS and Sep rats suggests a reproductive improvement caused by these treatments.

206.

FA17 - EFFECT OF STARVATION ON MUCOSAL MORPHOLOGY OF SMALL INTESTINE IN *Passer domesticus*

Funes SC, Chediack JG, Filippa V, Cid FD, Mohamed F, Caviedes-Vidal E.

Lab. Biología Integrativa. IMIBIO-SL. CONICET. Cátedra de Histología. UNSL. San Luis. Argentina.

Starvation induces rapid and dramatic changes in the digestive system. The small intestine (SI) is the organ that is most affected, and to date, little is known about the effect of phase III starvation in the intestinal histology in no migratory small wild birds. Our objective was to determine the effect of starvation on the small intestine histology. To test our goal, eight *Passer domesticus* were acclimated to laboratory conditions with water and food *ad libitum*, then four animals were fasted (phase III of starvation) and four were used as controls. The SI was removed and sectioned in three portions (proximal, medial and distal) and fixed in Bouin solution. Then, the intestinal pieces were dehydrated and embedded in paraffin. Histological studies were performed on 5 μ m sections, stained by hematoxylin-eosin coloration, and examined by a light microscope. Our results showed a significant decrease ($P < 0.05$) on almost all intestinal parameters in fasted animals compared with controls. Perimeter reduction was around 15%, 30% on mucosal thickness and villus height (except distal portion), 27% on villus width, 25% on enterocytes height and 12% on width. In addition, a reduction in crypts abundance and mitotic cells abundance in crypts of the fasted animals were observed. Starvation in passerines birds produces gut mucosa atrophy and a phenotypic change in enterocytes. Probably, one mechanism involved is a decrease in cell proliferation.

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207.

FA18 - DAILY VARIATIONS OF BLOOD BIO-CHEMICAL PARAMETERS IN *Passer domesticus*. EFFECT OF FASTING

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In mammals and birds starvation is characterized into three sequential phases (F1, F2 and F3) that are defined by physiological changes, such as rate of mass loss or changes in biochemical parameters. In small birds these changes occur in few hours, and are not known how these parameters change during day. Our objective was to determine the daily variations of biochemical parameters and the effect of fasting. To test our goal, for daily variations experiment twenty *Passer domesticus* were acclimated to laboratory conditions with water and food *ad libitum* and then four groups were established (8:00, 12:00, 16:00 and 20:00 hs). For fasting experiment, five groups were established (Control, F1:4hs, F2:8 y 12hs, F3:32hs). We measured hematocrits (Hto) and plasma concentrations of triglycerides (TG), uric acid (UA), total protein (TP) and albumin (Al). We found daily variations (without fasting) in TG, UA and Al, but not in TP and Hto. TG and UA show a peak at 16:00 h, and Al increase during the day until 20:00hs. During fasting, profiles of TG, TP and Al plasma levels and hematocrit, are similar during F1 and F2, but change at F3 (32 hs of fasting). UA change during all phases of fasting, showing a peak at 32hs of fasting. The last phase of fasting produces drastic variations in metabolites profiles in plasma, mainly in uric acid and hematocrit.

208.

FA19 - VARIABILITY IN LEUCOCYTE PROFILES IN *Passer domesticus*. EFFECT OF FASTING

Ronchi GD, Chediack JG, Cid FD, Padrones N, Arias R, Caviedes-Vidal E.

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Fluctuations in food supply are the rule in nature. The animal immune system changes during fasting period. In birds, starvation is characterized into three sequential phases (F1, F2, F3) that are defined by physiological changes, among biochemical parameters. A classical method to determine immunological state is measuring leucocyte profiles. Our objective was to determine the effect of fasting on distribution of leucocyte types and the development of heterophils/lymphocytes (H/L) ratio in *Passer domesticus*. To test our goal, twenty *Passer domesticus* were acclimated to laboratory conditions, water and food *ad libitum*, and regulated temperature. Since, repeated blood sampling affect hematological parameters we established four independents groups of birds: control and three fasting groups F1: 4hs, F2: 12hs and F3: 32hs. Blood samples were collected from brachial vein and smears were stained by the May Grünwald Giemsa technique. A total of 100 leucocytes were classified by slide. The differential count included relative percentages of lymphocytes (L), heterophils (H), eosinophils and monocytes. We found a higher number of lymphocytes and eosinophils at 32 hs of fasting, but not for heterophils. The ratio H/L, which is indicative of stress, increases at 32 hs of fasting. In conclusion, chronic fasting (F3) produce a stress situation and changes immunological status of animals.

209.

FA20 - AROMATASE ACTIVITY IN BIDDER'S ORGAN OF *Rhinella arenarum* (AMPHIBIA, ANURA)

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The Bidder's organ (BO) of male true toads of Bufonidae family is located in the anterior pole of the testis and it has been compared to a rudimentary ovary because of the presence of previtellogenic follicles. The presence of several steroidogenic enzymes has been detected in BO from adult males of *Rhinella arenarum* and, since testes lack aromatase activity, plasma estradiol may be synthesized by BO. The purpose of this study was to analyze annual variations in plasma estradiol levels and also in aromatase activity in BO. Enzymatic activity was determined in homogenates of BO and plasma estradiol levels were measured by radioimmunoassay. Lowest levels of plasma estradiol are registered during the pre reproductive season (PreR, $0,39 \pm 0,01$ ng/ml), they increase during the reproductive season (R, $0,90 \pm 0,02$ ng/ml) and achieve highest levels during the post reproductive season (PostR, $1,34 \pm 0,04$ ng/ml). Moreover, during PreR season total aromatase activity is significantly lower than during the rest of the year (PreR, $3,66 \pm 1,46$ pmol*min; R, $13,56 \pm 4,15$ pmol*min; PostR, $16,20 \pm 9,47$ pmol*min). Taken together, these results suggest that the BO is the main source of plasma estradiol.

210.

FA21 - EXTRACELULAR DIGESTION OF PROTEINS ALONG THE INTESTINE OF THE APPLE SNAIL *Pomacea canaliculata*

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The digestive physiology of this snail was revisited because it was previously reported the absence of protease activity in the intestinal content of ampullariids and the endosymbiont isolated from the midgut gland (MGG) had an important intrinsic protease activity. Specific protease activity (mIU/mg of protein) changed along the intestine of *P. canaliculata*. It was significantly higher in the coiled gut content than in the crop and stomach contents (ANOVA, $p < 0.05$). Zymography of these contents showed proteases of 32 and 130 kDa in the crop and stomach and 32, 150 and 200 kDa in the coiled gut. Aprotinin (serin-protease inhibitor) inhibited the 60% of protease activity in crop and stomach contents. This inhibition was positive in all soluble proteases found (30, 130 and 150 kDa). The optimal temperature and pH for protease activity were 30-35°C in all content studied (crop, stomach and coiled gut) and pH 8.5 in stomach contents and pH 9.5 in crop and coiled gut contents, respectively. Leu-N-aminopeptidase specific activity was gradually increased from the crop to the coiled gut, with the significantly higher specific activity in the MGG (ANOVA, $p < 0.05$). We suggested that this snail can carry out protein digestion using soluble and membrane associated proteases (leu-N-aminopeptidases).

211.

FA23 - THE PHYSIOLOGICAL RELEVANCE OF MIDGUT MEMBRANE PROTEINS IN THE PROCESS OF LIPID TRANSFER IN A VECTOR OF CHAGAS' DISEASEFruttero L¹, Stariolo R², Carlini C³, Rubiolo E¹, Canavoso L¹.¹Dpto Bioq Clin, CIBICI-CONICET, Fac Cs Qcas, UNC. ²Coord Nac Control Vectores, Min Salud. ³Dpto Biofísica, UFRGS, Brasil.

In insects, lipid transport through hemolymph is accomplished by lipophorin, the main high-density lipoprotein. It is suggested that lipophorin transfers its lipid cargo to tissues by a process mediated by uncharacterized non-endocytic receptors. In this context, we have analyzed the lipid transfer between circulating lipophorin and the midgut tissue in *Panstrongylus megistus*, a Chagas' disease vector. We made special focus in the characterization of membrane proteins that interacted with lipophorin. Immunofluorescence and *in vivo* assays with fluorescently labeled lipophorin showed that this lipoprotein interacted mainly with the sub-epithelial layer of the midgut. No intracellular signal compatible with endocytosis was observed. The use of lipophorin-Bodipy-FA (a fatty acid analog) demonstrated that lipid transfer at the midgut was bidirectional, and showed no dependence with the nutritional status of the insect. By combining ligand blotting, immunoprecipitation, and mass spectrometry approaches, several midgut membrane proteins interacting with lipophorin were identified, including the β subunit of the ATP synthase complex (β -ATPase), which was pointed out in human cell lines as a lipoprotein receptor. These findings are in agreement with the function of lipophorin as a reusable shuttle and suggest for the first time a novel role for β -ATPase as a non-endocytic lipophorin receptor in the midgut of *P. megistus*.

212.

FA26 - FLUOXETINE AND VENLAFAXINE EFFECT ON RECEPTORS EXPRESSION INVOLVED IN DEPRESSIONPoretti MB¹, Sawant R², Rask-Andersen M², Chaban R², Schiöth H², Fiol de Cuneo M¹, Carlini VP¹.¹Cát Fisiol Humana, Fac Cs Médicas, UNC. ²Dep Neuroscience, Uppsala University, Sweden.

Depression is an imbalance of neuropeptides and neural hormones characterized by increased hypothalamic-pituitary-adrenal (HPA) axis activity. Only chronic treatment with antidepressants is effective in treating depression, despite their side effects. Corticotrophin releasing hormone (CRH) and vasopressin (VP) play important roles in mediating the ACTH release through their receptors CRHR1 and AVPr1b. We investigate the fluoxetine (F) and venlafaxin (V) effects on the expression of these receptors in pituitary. Adult male Albino's Swiss mice were divided in sham (SO) and olfactory bulbectomized (OB) and orally treated, during 28 days with saline, F (10 mg/Kg/day) or V (10 mg/Kg/day). The last day of treatment, the tail suspension test (TST) was applied and then, the pituitary was collected, in order to study the mRNA CRHR1 and AVPr1b expression. After F treatment CRHR1 (F=8.415, $p=0.0009$) and AVPr1b (F=3.343, $p=0.046$) expression increased in SO whereas in OB only CRHR1 (F=8.415, $p=0.0009$) expression was increased. SO and OB mice treated with F or V showed antidepressant behavior in TST (F=8.57, $p \leq 0.05$). Increased expression of CRHR1 and AVPr1b in pituitary after fluoxetine treatment could be a compensatory mechanism for decreased CRH and VP in the hypothalamus.

213.

FA28 - EFFECTS OF FOOD RESTRICTION ON THE IMMUNE SYSTEM OF ROCK PIGEON *Columba livia*

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Due to climate change, new ecological scenarios pose a mismatch between peak food abundance and its requirements by animal species. Our goal was to determine the effect of food restriction on the cellular branch of rock pigeon's immune system. On day 0, individually caged pigeons were subjected to total food deprivation. A limit of total body-mass loss was fixed at 30% (fasting -F- day). Then, birds were refed until they regained their initial body weight (refed -RF- day). Blood samples were taken on days 0, F and RF and leukocytes counts and blood smears were performed. In addition, tests for phytohaemagglutinin (PHA) swelling reaction were conducted in the bird's wings to evaluate cellular immunity. Total leukocyte counts decreased on F day and showed a recovery trend on RF day. The percentage of lymphocytes (L) decreased significantly on F day, returning to baseline on RF day. Heterophils (H) varied inversely to lymphocytes. After the fasting period, the H/L ratio rose sharply, returning to baseline on RF day. Monocytes, eosinophils and basophils showed no clear trends. In the PHA test, birds exhibited a reduced swelling on F day and no recovery on RF day. Hence, energy deficiency has substantial effects on the cellular branch of the immune system: a decrease in the total number of leukocytes, an increase in H/L ratio and a decrease in cellular immunity.

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214.

FA29 - ANTIOXIDANT DEFENSES IN AN ISCHEMIA/REPERFUSION MODEL INDUCED BY DEHYDRATION

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Pomacea canaliculata is an invasive gastropod that has been expanded to Asia, Europe and Hawaii where it is an agriculture plague and an intermediate host for the agent of meningoencephalitis. This snail could survive longer periods of desiccation when the water is scarce, changing to an anaerobic metabolism (estivation) and have a specialized tissue that accumulates uric acid and this compound act as an antioxidant during reoxygenation occurred in the arousal of estivation. We quantificate the lipid peroxidation (TBARs), reduced glutathione (GSH), uric acid (UA), allantoin and protein concentrations and the activity of superoxide dismutase (SOD) and catalase (CAT) enzymes in kidney, midgut gland and foot muscle in control, post 45 days of estivation, 20 min or 24 h after reoxygenation during arousal from estivation groups. Kidney and foot had an increase in TBARs concentration after estivation and during arousal decrease. These levels didn't change in the midgut gland. UA concentration decrease after arousal from estivation suggesting consume during their antioxidant action. All the organs count with the antioxidant action of GSH, since only in the midgut gland an increase in the CAT activity was observed. SOD activity was constant in the studied tissues of all groups. This work confirm our previous studies related to the antioxidant role of UA, observing that GSH contribute significantly to antioxidant defenses during the arousal from estivation and CAT is added to this defenses in midgut gland and kidney.

215.

FA31 - CHROMOSOME STUDIES IN CRYPTORCHID DOGS

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Cryptorchidism is the failure of the testicles to descend into the scrotum through the inguinal canal, with unilateral or bilateral phenotypic expression and variability in the location of the retained testicle(s). Cryptorchidism in dogs may have undesirable effects such as infertility, a higher risk for tumor formation and feminization, with a negative economic impact in dog breeding. Cryptorchidism is supposed to be caused by the interaction of genetic, epigenetic and environmental factors. The objective of this work was to evaluate two dogs with the clinical phenotype: A 3 year-old boxer with unilateral cryptorchidism, asymmetry of the mammary chain and long tongue; and 1.5 year-old cross breed canine with bilateral cryptorchidism. Heparinized blood was cultured for 72 h at 37C according to cytogenetic conventional methods. Twenty cells per animal were examined for karyotype determination. Both patients showed 2n= 78. The unilateral cryptorchid dog has 20% of cells with 1q+ alteration. Some studies have reported chromosomal abnormalities in cryptorchid men. The identification of chromosome alterations in patients with urological and/or genital abnormalities will permit a better localization of the putative gen/s for urogenital development.

216.

FA32 - EFFECT OF *Euphorbia serpens* ON URINARY EXCRETION OF SODIO, POTASIO AND CLORURO IN RATS

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Euphorbia serpens, known popularly as "cola de novia", "leche de paloma", is used in popular medicine as diuretic. The aim of this study was to relate the diuretic activity and the urinary excretion of Na⁺, K⁺ and Cl⁻ in Wistar rats, using the Lipschitz method (1943). Each lot was treated with: 20% infusion from *Euphorbia serpens*, furosemide (10 mg/kg) as standard drug and saline solution as negative control. Urinary volumetric excretion and urinary levels of Na⁺, K⁺ and Cl⁻ were measured in 3 hours diuresis after each treatment. All values were expressed as the mean ± SEM. Student's *t*-test was performed to evaluate the statistical differences between the control and the experimental samples. The treatment with infusion of *Euphorbia serpens* it caused an increment in the urinary excretion of Na⁺ and K⁺ (p<0.001 and p<0.05 vs. negative control, respectively). The urine samples of animals treated with *E. serpens* presented normal levels of Cl⁻. The data reported in this work indicate that the treatment with the infusion of *Euphorbia serpens* it caused an increment in the urinary excretion of Na⁺ and K⁺, while treatment of rats with the infusion had no significant effect on levels of Cl⁻. Further investigations are necessary prior to their recommendation for use as safe and effective diuretic.

217.

FA33 - EFFECT OF *Plantago major* ON DIURESIS IN RATS

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Plantago major, popularly known as “llantén”, is used in popular medicine as digestive, rubefascent, anti-inflammatory, diuretic. This study was designed to determine the diuretic activity of the *Plantago major* in Wistar rats (180-220 g), using the Lipschitz method (1943). The treated rats received methanolic extract (250 and 500 mg/kg, *p. o.*) of *Plantago major* or furosemide as standard drug (10 mg/kg). The control group received only the NaCl isotonic solution (50 ml/kg). Urinary volumetric excretion and urine chemical parameters were measured in 3 hours diuresis. 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 methanolic extract of *Plantago major* showed diuretic activity (*P. major* at a dose of 250 mg/kg: between 30 and 105 min, $p < 0.05$ vs. negative control; and at a dose of 500 mg/kg: between 45 and 180 min, $p < 0.05$ vs. negative control). The urine samples presented normal chemical parameters in all the cases. The data reported in this work indicate that the methanolic extract of *Plantago major* showed a moderate diuretic activity in comparison with furosemide, a high-ceiling diuretic agent. This fact support the use in traditional medicine of *Plantago major*.

218.

FA34 - CADMIUM ON EXPRESSION OF GENES RELATED WITH OXIDATIVE STRESS AND INFLAMMATION IN INTESTINE

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Cadmium (Cd) is an important environmental contaminant that induces oxidative damage in cells. Previous results from our laboratory showed an increase of Cd, TBAR's and metallothionein (MT) content, a decrease of glutathione reductase activity and catalase mRNA levels, and also histological alterations in the small intestine of male rats (180g) received 15 ppm of Cd in the tap water for 2 months. Now, expression of proteins related with the response of small intestine to oxidative stress and inflammation are determine in the same experimental model. Thus, MT-II, NRF-2, PPAR γ , TNF α and β -actin (as internal control) mRNA levels, were determine by RT-PCR. Tissue total RNA was isolated using TRIzol. The PCR products were analysed on 2% agarose gels containing Gel Red. Band intensities of RT-PCR products were quantified using Image J software. The expression of NRF-2 and TNF- α were increased in intestine of Cd exposed rats ($P < 0.05$), without changes in the other genes, compared to control. These and our previous results indicate that: 1- MT increase induced by oral Cd exposition did not occur by increase of MTII expression. 2- There is a small intestine response to oxidative stress since NRF-2 increases. 3- The high expression of TNF α should be associated to the histological alterations previously informed for us.

219.

FT5 - STUDY OF HEPATOPROTECTIVE ACTIVITY OF *Artemisia douglasiana* Besser

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Artemisia douglasiana Besser (*Ad*), known as “matico”, have been used in folk medicine for gastrointestinal disorders. The aim of this work was to study its possible protective effect against paracetamol and CCl₄-induced hepatic damage. Male Wistar rats (200-250 g) were divided into five groups of six animals each. Control groups received acetaminophen (640 mg/kg) or CCl₄ (0.15ml/kg); experimental groups received *Ad* lyophilized water infusion (20%) and paracetamol or *Ad* water infusion (10%) and CCl₄; blank group received vehicle. Serum aspartate and alanine aminotransferase, alkaline phosphatase, cholesterol, glutamil transferase and bilirubin were determined. The *Ad* infusion produced reduction of AST ($p < 0.05$, ANOVA- Tukey), but significant differences were not observed in ALT after administration of acetaminophen. The *Ad* infusion produced marked reduction of both, AST and ALT ($p < 0.001$, ANOVA- Tukey) after toxic dose of CCl₄ relative to the control group. There were no significantly differences in others serum parameters. These results suggest that *Ad* infusion shows hepatoprotective activity in the acute liver injury induced by acetaminophen or CCl₄. Antioxidant properties of flavonoids compounds present in *Ad* may be the origin of these effects.

220.

FT6 - CAVEOLIN-1 LINKED TO eNOS/Hsp70 INTERACTION ON ROSUVASTATIN PROTECTION AGAINST FIBROSIS IN NEONATAL OBSTRUCTIVE NEPHROPATHY

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Statins restore endothelial nitric oxide (NO) levels by up-regulating endothelial NO synthase (eNOS). Caveolin-1/eNOS interaction is essential for NO levels. Here, we evaluated whether caveolin-1 associated with eNOS/Hsp70 interaction are involved in the rosuvastatin (Ros) tubulointerstitial fibrosis protection effect during neonatal unilateral ureteral obstruction (UUO). Neonatal rats (n=5) subjected to UUO within two days of birth and controls were treated daily with vehicle or Ros (10 mg/kg/day) by oral gavage for 14 days. After UUO, morphometric evaluation of interstitial fibrosis showed increased interstitial volume with reduced NO level, increased caveolin-1, as well as downregulation eNOS and heat shock protein 70 (Hsp70) expression. Conversely, Ros treatment attenuated the fibrotic response linked to high NO availability, decreased caveolin-1, and marked upregulation of eNOS and Hsp70. Coimmunoprecipitation have shown decreased caveolin-1/eNOS as well as increased Hsp70/eNOS interaction, after Ros treatment. A dose dependent effect of rosuvastatin on decreased caveolin-1 expression was shown. In conclusion, our data of increased NO availability, involving interaction of upregulated eNOS/Hsp70 and downregulated caveolin-1 may contribute to the protection against tubulointerstitial fibrosis injury, afforded by rosuvastatin in neonatal early kidney obstruction.

221.

FT7 - ROSUVASTATIN PROMOTES THE IMPROVEMENT OF NEPHROGENIC MARKERS IN OBSTRUCTIVE NEPHROPATHY MODEL*Mazzei L, García IM, Docherty NG¹, Manucha W.**IMBECU-FCM-CONICET, UNCuyo, Mendoza, Argentina. ¹School of Medicine, Dublin, Ireland. E-mail: wmanucha@fcm.uncu.edu.ar*

Congenital obstructive nephropathy is characterized by tubular apoptosis and fibrosis. Statins prevent apoptotic and fibrotic processes in renal cells. The present study examined whether rosuvastatin (Ros) could protect against neonatal obstructive nephropathy in the rat in association with the preservation of the expression of markers of nephrogenesis. **Methods:** Neonatal rats (n=5) underwent unilateral ureteral obstruction (UUO) or sham surgery (control), were randomized to receive oral Ros (10mg/kg/day) or vehicle for 2 weeks. Renal tissue was processed for quantification of tubular dilatation, apoptosis, fibrosis, TNF- α , TGF- β 1, WT1, Snail, BMP-7 and E-cadherin protein expression. **Results:** UUO significantly increased tubular apoptosis, dilatation and tubule-interstitial fibrosis coupled to increases in TNF- α and TGF- β 1 mRNA expression. Ros-treat during UUO markedly protected against these changes. Preservation of renal WT1, Snail, BMP-7 and E-cadherin, was also associated to Ros treatment. **Conclusions:** The protective effects of Ros were sufficient to preserve a normal profile of expression for a number of markers of nephrogenesis suggesting that renal development is likely to continue despite of obstruction.

222.

FT8 - PARICALCITOL INDUCES MITOCHONDRIAL CYTO-PROTECTIVE EFFECT DURING A NEPHROPATHY MODEL*Altamirano L, García IM, Mazzei L, Ferder L¹, Manucha W.**IMBECU-CONICET, FCM, U. N. Cuyo, Mendoza, Argentina. ¹Ponce School of Medicine, Ponce, Puerto Rico. E-mail: wmanucha@fcm.uncu.edu.ar*

Background: Growing evidence proposes that vitamin D slows the progression of chronic kidney diseases. Further, activators of vitamin D receptors (VDR) have suppressant effect on the renin-angiotensin system (RAS), as well as anti-inflammatory and anti-fibrotic effects. **Aim:** This study was performed to evaluate in an obstructive nephropathy model (UUO), possible paricalcitol (Pari) cyto-protective effect at the mitochondrial level. **Methods:** Ten adult female rats were obstructed surgically and divided into two groups (control or treated). The treatment was done for 15-day duration (30ng/Kg). We evaluated: 1-parathyroid hormone, Ca²⁺ and P, 2-Fibrosis, apoptosis and mitochondrial morphology, 3-VDR, AT₁ receptor, TGF- β and Renox (NOX₄) expression and 4)-NADPH oxidase activity. **Results:** Biochemical, histological and molecular studies shows mitochondrial injury in UUO. Electronic microscopy revealed, electronically luminous nuclear material and the mitochondria were increased in size with dilated crests and spaces in their interior. Also, high AT₁ expression and NADPH activity were reverts in paricalcitol-treated animals. **Conclusions:** These results suggest, in an obstructed nephropathy model, a cyto-protected effect of the activator of Vitamin D receptors, paricalcitol, revealing for the first time a possible protective effect AT₁ receptor dependent at the mitochondrial level.

223.

FT9 - CADMIUM DETERMINATION IN CIGARETTES FILTERS BY MOLECULAR FLUORESCENCE*Talio M, Luconi M, Zambrano K, Fernández L.**Fac. Qca., Bqca y Fcia, UNSL-INQUISAL-CONICET, 5700 - San Luis. E-mail: lfernand@unsl.edu.ar*

Cigarette filters or butts are the main source of waste in the world, discarding on average 4.5 trillion cigarette butts each year. They are made of acetate, a non-biodegradable material, remaining long time in the environment and representing a potential source of pollution. Trace heavy metals are considered to be one of the main sources of pollution since they have significant effect on its ecological quality. Cadmium is very important in environmental and biological sciences; it is highly toxic even at low concentrations, causing damages to organs such as the kidneys, liver and lungs. This work proposes the quantification of cadmium in cigarette filters by molecular fluorescence. The methodology is based on the formation of a ternary complex of the metal with azo-reactive SPADNS and rhodamine B. Among the experimental parameters that influence fluorescence emission were optimized: concentrations of azo-reactive dye, concentration and nature of buffer, pH complex formation and order of addition of reagents. Regarding the stage of leaching of the metal present in cigarette filters were optimized contact time, the pH and temperature of the leaching solution. Developed methodology represents an uncommon application of luminescence to metal analysis with comparable sensitivity and accuracy to traditional atomic spectroscopies.

224.

FT11 - GLYPHOSATE OXIDATION BY UVC/H₂O₂ PROCESS: BIOLOGICAL EVALUATION IN *Rhinella arenarum**Junges CM¹, Attademo AM¹, Peltzer PM¹, Lajmanovich RC¹, Cardell L², Vidal E², Negro A², Cassano A², Zalazar C².**¹CONICET- FBCB-UNL. ²INTEC (UNL-CONICET). Sta. Fe, Arg*

Amphibians are one of the vertebrate groups most sensitive to the effects of glyphosate. Nowadays, genetically modified crops indiscriminately produced in a large area of Argentina rely on this agrochemical. The residues generated by the use of this pesticide should be correctly treated. There are remediation technologies, such as Advanced Oxidation Processes, based on the production of strongly oxidative species, such as hydroxyl radical. The combination of UVC/H₂O₂ offers some advantages, such as ease of operation. The aims of this work were to evaluate the lethal (% mortality) and sublethal effects of inhibition of acetylcholinesterase [AChE] and butyrylcholinesterase [BChE] activities on *Rhinella arenarum* tadpoles. Acute bioassays (24 and 48 h) were performed on samples containing H₂O₂ and samples from which H₂O₂ was previously removed, both treated for 120, 240 and 360 min. The commercial product processed was Eskoba® (48% a.i.) (isopropylamine salt of N-(phosphonomethyl) glycine) (50 mg/l). The effects of H₂O₂ as positive control (PC) were also evaluated. AChE and BChE activity was determined in surviving tadpoles using routine techniques. Mortality data were compared with the J_i² test and enzymatic activities with the Kruskal-Wallis method. Mortality percentages of tadpoles exposed to treated samples and to PC were significantly higher (p < 0.01) at 120 min. By removing H₂O₂, a 100% survival was achieved at only 120 min. AChE and BChE activities were significantly inhibited (p < 0.05) up to 240 min. The results confirm the efficacy of the UVC/H₂O₂ process to reduce the effects of glyphosate contamination.

225.

FT12 - EFFECT OF THE FUNGICIDE TRIFLOXYSTROBIN ON B-ESTERASE ACTIVITY IN TADPOLES OF *Scinax nasicus* (AMPHIBIA: ANURA)Attademo AM¹, Junges CM¹, Lajmanovich RC¹, Peltzer PM¹, Cabagna-Zenkhusen M¹, Lorenzatti EA².¹CONICET-FBCB-UNL, Santa Fe, Argentina. ²INTEC (UNL-CONICET), Santa Fe, Argentina.

In our country, in late 2007, new strobirulin and triazole based biocide formulations were introduced to the market. These components ensure high control of new fungal late season diseases in soybean crops. Trifloxystrobin (TFT), one of these formulations, is characterized by being very toxic to aquatic organisms, with a half-life of 8-24 hours in surface waters.

The aim of the present work was to evaluate the effect of TFT on B-esterase activity (butyrylcholinesterase; BChE and acetylcholinesterase; AChE) on tadpoles of *Scinax nasicus* (Amphibia: Anura) under controlled laboratory conditions. Acute toxicity bioassays (24 and 48 h) were performed using sublethal concentrations of TFT (0.0312, 0.0625, 0.125, and 0.250 mg/L) and a control, with the respective replicates. The commercial product used was Flynt® (50% a.i., Bayer CropScience S.A., Argentina). Seven larvae, Gosner stage 39-42, per container and per treatment were exposed under a light-dark (12:12) photoperiod at 25 ± 2°C. Muscle BChE and brain AChE activities were determined for each individual using routine techniques. The results obtained indicate an inhibition of BChE activity at all TFT concentrations assayed ($P < 0.05$) at 48 h of exposure with respect to control. A similar trend was observed for AChE, but no statistically significant differences were observed ($P > 0.05$). We preliminarily determined an inhibitory effect of TFT on BChE activity of *S. nasicus* tadpoles.

226.

FT13 - *Aristolochia argentina*: EFFECT ON GASTRIC MUCOSE AND *HELICOBACTER PYLORI* STRAINSParedes J¹, Salinas AG³, Fusco M², Sosa A², Wendel G¹, Vega AE³, Cortiñas TP³, Pelzer L¹.¹Farmacología, ²Farmacognosia, ³Microbiología. FQByF, UNSL, San Luis. E-mail: gwendel@unsl.edu.ar

Aristolochia argentina (familia Aristolochiaceae), popularly known as “charrúa”, is used in folk medicine. The aim of this study was to assess the anti-ulcerogenic effect in rats and determine the antibacterial activity on *H. pylori* susceptible and resistant strains. Wistar rats of either sex (200-250g) were employed. We examined the effect of *A. argentina* on gastric damage induced by oral administration of absolute ethanol (EtOH). The results of the macroscopic evaluation were confirmed by the data obtained by measuring the area of the lesioned mucosa by using a computer-based image analysis. *A. argentina* prevents the formation of gastric lesions (% inhibition): 250 mg/kg: 44.89% ($p < 0.01$) and 500 mg/kg: 69.45% ($p < 0.001$) vs EtOH. The minimum inhibitory concentration (MIC) of *A. argentina* was assayed by agar dilution method according to Clinical and Laboratory Standards Institute, using serial dilutions of extracts in the range of 64 at 0.032 mg/ml. *H. pylori* strains, NCTC11638 (reference strain) and five clinical isolates were assayed. The results showed that *A. argentina* had activity against *H. pylori* with MICs between 2 to 8 mg/ml for clinical isolates and 16 mg/ml for NCTC 11638 reference strain. This anti-ulcerogenic effect of *A. argentina* could be due, in part, to the presence of flavonoids in this plant. *A. argentina* may be a useful alternative treatment strategy principally in eradication of antimicrobial resistant strain.

227.

FT14 - THE LEAVES OF *Jodina rhombifolia* REDUCE ACQUISITION OF ALCOHOL DRINKING BEHAVIOR IN RATS

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Alcoholism is a serious medical, social and economic problem facing almost all human societies worldwide. Several authors cite the leaves of *Jodina rhombifolia* (Hook. & Arn.) Reissek, “peje”, “sombra de toro” (Santalaceae) in traditional medicine against the alcoholism. In the present study we investigated the ability of the infusion of the leaves to 10% in reducing voluntary ethanol intake in rats. The plant material was collected in the town of Fraga, San Luis Province. Infusion was prepared according FNA VI Ed. We used male Wistar rats of 200-250 g. Ethanol intake occurred under the two-bottle free-choice regimen between 20% (v/v) ethanol and water, 24 h/day. Rats were alcohol-naive before the start of the study. Recording of daily alcohol, water and food intake was performed throughout the 10 day of treatment. Mean daily alcohol intake in vehicle rats rose to 5-7 g/kg per day. The repeated administration of infusion (2 ml, *p.o.*, twice a day) resulted in a significant ($p < 0.001$) reduction in the acquisition of alcohol drinking. In the rat group treated with infusion the mean of the amount of alcohol consumed daily was lower than 4 g/kg per day throughout the 3 to 10-days treatment period. Food intake did not differ among rat groups. Water consumption and weight gain were significantly reduced in the group treated with the infusion ($p < 0.001$). The results obtained in this preliminary study validate the popular use of species leaves in the treatment of alcoholism.

228.

FT15 - TREATMENT OF PREVALENT HEALTH PROBLEMS OF THE MUSCULO-SKELETAL SYSTEM IN SAN LUIS

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Our objective was to identify prevalent pathologies of the musculoskeletal system (OMS) and analyze the dispensations of prescribed medicines in three pharmacies of San Luis city. We performed a descriptive, transversal and retrospective study. Data were collected of recipes during 2 months (age, sex, pathologies and prescriptions). The diagnoses and medicines were classified according to International Diseases and Anatomical Therapeutic Chemical classifications, respectively. Results (%). Diagnoses: arthrosis (A) 36, osteoporosis (O) 32, polyarthritis (PA) 15.4, back pain 5, muscle contracture 4.3. Sex and Age in A, O, PA, respectively: F (74,95,78) M (26,5,22). 14-64 (21,40,33), >64 (79,57,67). Prescriptions: A: Diclofenac 21, glucosamine 12, chondroitin+ glucosamine 12, tramadol 7, meloxicam 5, vitamins+ minerals 5, diclofenac+acetaminophen 5; O: calcium+vitamin D3 27, ibandronate 19, alendronate 11, calcium 8, vitaminD 5, PA: glucosamine 22, glucosamine+meloxicam 17, chondroitin+ glucosamine, acetaminophen, naproxen, ibuprofen, diclofenac, celecoxib, and vitamins+minerals 6. The medicines most prescribed in the A and PA were glucosamine and nonsteroidal anti-inflammatory drugs, while in O the compounds with calcium and vitamin D3 and bisphosphonates. The use of drugs whose efficacy is similar to placebo and with unfavorable risk/benefit ratio was very high. Misuse of medicines increases the health costs and the risk of apparition of adverse reactions.

229.

FT16 - DRUG UTILIZATION IN THE HOSPITAL GUARD SERVICE

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Our aim was to analyze the prevalent diagnoses, the prescriptions and the routes of administration used (VA) in the hospital guard service of San Luis city. An observational, cross-sectional and retrospective study was performed. Age, sex, diagnoses, prescriptions and VA of 283 patients were recorded. Drugs and diagnoses were classified according to ATC and ICD-10 classifications, respectively. Results (%). Diagnoses: toothache 12.5, arthralgia 10.8, recurrent obstructive bronchitis (RBO) 9, biliary colic 7.7, acute respiratory infections (AIR) 7.3, fever 7, gastroenteritis 7. Prescriptions: toothache: dexamethasone 45, diclofenac 15, ibuprofen 12.3, dipyrone 11; arthralgia: diclofenac 28, ketorolac 26, dexamethasone 20; RBO: salbutamol 30.8, dexamethasone 23; biliary colic: N-Butylscopolamine 44.2, 17.3 ketorolac, dipyrone 15.4; IRA: ibuprofen 26.8, dexamethasone, 24.4, dipyrone 14.6, fever: 66.7 dipyrone, ibuprofen 22.2; gastroenteritis metoclopramide 29.3, dipyrone 26.8, vomiting: domperidone 31.3, metoclopramide 25. VA: intramuscular 57.6, intravenous 26.5, oral 11.7, inhaled 2.1. The prevalent health problems, different types of pain, AIR, fever and gastroenteritis not justify the use of guard service, but from other specialties. Drugs that have an unfavorable risk/benefit profile (dipyrone, ketorolac, dexamethasone) are overused for different types of pain. There was a wide use of injectables, which implies a greater risk to the patient and a high cost to the health service. It is necessary to implement intervention measures to encourage better drug use.

230.

FT17 - ANTIOXIDANT, HEMOLYTIC AND CYTOTOXIC ACTIVITIES OF EXTRACTS FROM *Senecio nutans* (ASTERACEAE)Lizarraga E^{1,2}, Castro JF², Fernández F³, Perotti M⁴, Catalán C^{1,4}.¹INQUINOA-CONICET, ²Fac. Ciencias Naturales, UNT; ³Fundación Miguel Lillo, ⁴Fac. Bioq., Química y Farmacia, UNT.

Senecio nutans Sch. Bip is a perennial shrub, 20-60 cm high that grows on the South American Andes at 3500-5000 m above sea level. This herb is commonly known as "chachacoma" being widely used in Andean traditional medicine. In this work, the total phenolic, flavonoids and caffeoylquinic acids contents and radical scavenging, antioxidant, hemolytic and cytotoxic activities of aqueous (infusion and decoction) and four different organic extracts were determined.

Phenolic compounds and caffeoylquinic acids were concentrated in the aqueous extracts which also showed good antioxidant activity [β -carotene and Mo(VI) assays] and radical scavenging capacity (DPPH test). The organic extracts showed much lower antioxidant and radical scavenging activity.

The aqueous extracts exhibited low/moderate toxicity and showed no hemolytic activity. The organic extracts in turn, displayed moderate or high cytotoxicity and a relatively high hemolytic effect (dose dependent).

231.

FT18 - USE OF DRUGS IN DISEASES PREVALENT IN SAN LUIS

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The drug utilization studies are an indispensable tool to achieve better use of medicines. Our objective was to analyze diagnoses and drugs dispensed in three pharmacies San Luis city. An observational, descriptive, transversal and retrospective study was performed. The data (sex, age, disease/s and drug/s) were collected for two months. The pathologies were classified according to the International Classification of Diseases (ICD 10) and medications according to Anatomical Therapeutic-Chemical Classification (ATC) and according to their therapeutic potential value. Results (%). Diagnoses: Hypertension (HT) 20, Gastritis 7, Anxiety 4, Hypercholesterolemia 3.3, arthrosis 3.2, Osteoporosis and Glaucoma 3, Depression, Allergy, Diabetes and discomfort 2, Sinusitis 1.5, Insomnia 1. Drugs: Clonazepam 3.5, Enalapril 3.4, Carvedilol 3, Vitamins & Minerals 2.5, Alprazolam 2.3, Omeprazole 2.2, Atorvastatin and Diclofenac 1.7, Losartan and Ibuprofen 1.5, lansoprazole and Enalapril+hydrochlorothiazide (HCT) 1.4, Glucosamine, Amoxicillin+Clavulanic acid and losartan+HCT 1, fixed dose combinations (FDC): 24. Sex: F 68 M 32. Age: <15(36), 15-64(38), > 64(56). An exhaustive analysis of the diagnoses and the prescriptions marked the prevalence in the three pharmacies of chronic diseases: HTA, gastritis, anxiety, affecting mainly feminine sex. Medicines of first election and high therapeutic value were prescribed, as well as those not considered high therapeutic value were prescribed. It is necessary to deepen these studies to evaluate the medicine use in each pathology.

232.

FT20 - ESSENTIAL OIL COMPOSITION, ANTIOXIDANT AND CYTOTOXIC ACTIVITIES OF EXTRACTS FROM *Senecio viridis* var. *viridis* PHILLLizarraga E^{1,2}, Perotti M³, Catalán C^{1,3}.¹INQUINOA-CONICET, ²Fac. Ciencias Naturales, UNT; ³Fac. Bioq., Química y Farmacia, UNT.

Senecio viridis var. *viridis* Phil. (Asteraceae), commonly known as "mocora", "mocaraca" or "chachacoma del burro", is an aromatic shrub that grows abundantly in the mountainous regions of Catamarca, Salta and Jujuy (Northwestern Argentina).

In this work, the essential oil composition, the *in vitro* antioxidant activity and *in vivo* (brine shrimp test) cytotoxic effect of aqueous (infusion and decoction) and several organic extracts from aerial parts of *S. viridis* var. *viridis* were determined

The essential oil was obtained by hydrodistillation and analyzed by GC-MS. Twenty eight compounds accounting for 92.1% were identified. The sesquiterpene ketone 4 β ,5 β -eremophyl-7(11),9-dien-8-one was the main constituent (84.9%) of the essential oil.

All the extracts displayed low antioxidant and radical scavenging activity (β -carotene bleaching test, Mo(VI) reduction test and DPPH assay). Both infusion and decoction showed low cytotoxicity. The organic extracts (hexane, chloroform, ethyl acetate and n-butanol) were moderately toxic.

233.

FT22 - PRENATAL TREATMENT WITH CAPTOPRIL INDUCES EMPHYSEMA IN RAT LUNG DEVELOPMENT

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Angiotensin-converting enzyme inhibitors (ACEI) have been widely used in hypertension treatment. However, the use of ACEI during pregnancy is contraindicated for risks of fetopathy. We investigated the effect of captopril prenatal treatment in lung development. Wistar rats were treated with captopril (2,85 mg/kg/day) delivered subcutaneously with osmotic mini-pumps during the last week of pregnancy. Pup's lungs at four different postnatal ages (P0, P8, P15 and P30) were evaluated by morphological, histomorphometric and immunohistochemistry staining analysis. Captopril treatment significantly decreased body and lung weights at P0 and P8. Histological study evidenced a substantial destruction of alveolar walls in treated rats, resulting in enlargement of distal airway spaces at P8, P15 and P30, demonstrated by significant increase in the interalveolar wall distance, quantified by mean linear intercept (MLI). Cellular proliferation was evaluated using PCNA (proliferating cell nuclear antigen). The percentage of PCNA-positively stained alveolar epithelial cells in the treated group was significantly higher at P15 and P30, respect to the control group. Cell proliferation decreased with age in control animals. However, in captopril-treated lungs the relative number of PCNA immunopositive nuclei remains high during the studied ages. These findings support the hypothesis of a functional renin-angiotensin-system required for normal lung development.

234.

FT24 - PRENATAL EXPOSURE TO BISPHENOL A MODIFIES UTERINE RESPONSE TO HORMONE REPLACEMENT THERAPY

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Present evidences suggest that perinatal exposure to endocrine disruptors may affect the uterine response to ovarian hormones in adulthood. Pregnant Wistar rats were orally exposed to vehicle (control), 0.5 or 50 µg BPA/kg/day from gestation day 9 until weaning and female offspring were treated with estrogenic therapy to study uterine histo-morphology and biomarker expression. Twelve month old female offspring were ovariectomized and treated with 17 β-estradiol (E2) for 3 months. Uterine samples were obtained for histopathology, immunohistochemistry and real time RT-PCR studies. Different uterine lesions were found after hormone replacement therapy: cystic, hypertrophic and atypic glands, glands with squamous metaplasia or with daughters' glands. Animals exposed to BPA0.5+E2 showed an increase in the density of glands with daughter glands, while BPA50+E2 group had a higher density of squamous metaplasias compared with controls. P63 was expressed in several epithelial cell layers of the squamous metaplastic glands. Progesterone receptor and estrogen receptor α expression were significant decreased in the uterine subepithelial stroma of animals exposed to BPA50+E2. IGF-I and IGF-I receptor mRNA expression was significantly lower in BPA0.5+E2 group. The results suggest that the exposure to low doses of BPA reprograms the development and functional differentiation of the uterus and modifies the response to E2 therapy.

235.

FT25 - GASTRIC ANTI-ULCEROUS ACTIVITY OF METHANOLIC EXTRACT OF *Acacia visco* LEAVES AND BARK, IN RAT. EFFECT OF BLOCKING ENDOGENOUS PROSTAGLANDIN

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In previous studies we reported antiulcer effect (Biocell, 26: 3, 2002) and free radical scavenging activity (Inflammopharmacology, 18:5; 253-260) of methanolic extracts of leaves (EMehAv) and bark (EMecAv) of *Acacia visco*. This work aimed at studying the effect of blocking endogenous prostaglandin with indometacin (Ind.) on the gastric anti-ulcerous activity of EMehAv and EMecAv. The anti-ulcerous activity was evaluated according to Robert et. al. (1979). Wistar rats, both sexes were used after 24h fasted. Normal and ulcer control groups were administered with saline v.o.; Ind. + EMehAv and Ind. + EMecAv groups received Ind. 10 mg/kg, i.p. and 30 min after EMehAv 300 mg/kg or EMecAv 300 mg/kg, v.o., respectively. EMehAv and EMecAv groups received 300 mg/kg v.o. of each extract respectively. After one hour, absolute ethanol were administered v.o. to all groups (except normal group). The ulcer grade was evaluated 60 min later according to Marazzi, Uberti and Turba scale and expressed as ulcer index (UI). Statistical analysis was performed by ANOVA. Results: Control group: UI=4.89±0.20; Ind + EMehAv group: UI = 0.25±0.20 (p<0,001), EMehAv group: UI = 0.00 (p<0,001), Ind + EMecAv group: UI = 2.00±0.40 (p<0,05), EMecAv group: UI = 1.60±0.97 (p<0,01). Conclusions: The endogenous prostaglandin would not be involved in the anti-ulcerous activity of methanolic extract of leaves of *A. visco* but would be involved in the anti-ulcerous activity of methanolic extract of bark of *A. visco*.

236.

FT27 - PHYSIOLOGICAL EFFECT OF A PESTICIDE MIXTURE ON THE NEOTROPICAL FISH *Piaractus mesopotamicus*

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Among the most used compounds for pest-control in Argentinean soybeans crops are organochlorine and pyrethroids insecticides. The main goal of this study was to assess sublethal physiological effects of a pesticide mixture (endosulfan and λ-cialothrin) in *Piaractus mesopotamicus* ("pacú"). Morphological, hematological, oxidative stress and enzymatic biomarkers were analyzed. Semi-static assays (96h) were carried out in juvenile fish, which were exposed to 1.1 µg·L⁻¹ endosulfan and 0.7 µg·L⁻¹ λ-cialothrin, alone and together (in combination). Fish exposed to the pesticide mixture showed an increase in the liver-somatic index and hematological changes related to white cells (increase in the white blood cells count, changes in the leucocytes frequency), a decrease in liver transaminases activity, an increase in lipid peroxidation levels in liver, kidney and brain, and changes in the glutathione S-transferase activity in liver, intestine and gills. Endosulfan and λ-cialothrin mixture produced a strong immune response and oxidative stress in several vital organs of *P. mesopotamicus*. These results show the significance of assessing complex exposure scenarios, to advance in environmental risk assessment.

237.

FT28 - CHANGES IN THE EXPRESSION OF ANDROGEN RECEPTOR IN OVARIAN FOLLICLES OF ADULT RAT BY PERINATAL EXPOSURE TO BISPHENOL A

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We have previously found that exposure to environmentally relevant doses of BPA during the neonatal period, affect the follicle development in adulthood and the number of embryo implantation sites. The aim of this study was to evaluate whether BPA could be affecting fertility through ovarian dysfunctions. Female pregnant Wistar rats were treated orally from gestational day 9 until weaning with 50µg BPA/kg-day (BPA50), 0.5µg BPA/kg-day (BPA0.5) or vehicle (alcohol). At PND21 the pups were weaned and the ovary was removed at PND90 (N=8). In serial sections, we evaluated the expression of estrogen Receptor alpha(ERa) and beta(ERb), androgen receptor(AR) and p27 by immunohistochemistry. The expression of ERa, ERb and p27 was unchanged by BPA, whereas the expression of AR showed a decrease in primordial, preantral and antral follicles, with the dose presumed as safe (BPA50). Previous results have shown that fertility is affected by changes in AR expression. Therefore, our results suggest that perinatal exposure to BPA affects fertility through a decrease in AR expression in the ovarian follicles.

238.

FT29 - OLEUROPEIN AND HYDROXYTYROSOL FROM OLIVE OIL INHIBIT MAST CELL ACTIVATION

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The present work was designed to examine the effects of hydroxytyrosol (Ht) and oleuropein (Olp), two phenolic compounds purified from olive oil, on calcium ionophore A23187- (A23187), compound 48/80- (48/80) (non-immunologic pathway) and concanavaline A- (ConA) (immunologic pathway) induced mast cell degranulation, with the goal of testing the hypothesis that such molecules act as mast cell stabilizers.

Rat peritoneal mast cells were purified in Percoll and incubated with: 1) Buffer (control) or 2) Secretagogue or 3) Ht or Olp + Secretagogue (Secretagogues: A23187 or ConA or 48/80). β -hexosaminidase release studies by colorimetric reaction, evaluation of mast cell morphology by light and electron microscopy and mast cell viability (Trypan blue dye exclusion) were performed. Time- and dose-response curves were also carried out. Statistical analysis: ANOVA-1/Tukey-Kramer.

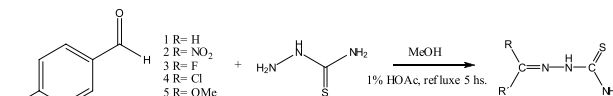
A23187, ConA and 48/80 increased β -hexosaminidase release from rat peritoneal mast cells and elicited evident morphological changes. These effects were inhibited by 100 µM Ht and 100 µM Olp.

The present study demonstrates that Ht and Olp inhibit A23187-, ConA- and 48/80-induced mast cell activation, acting thus as mast cell stabilizers in rat peritoneal mast cells.

239.

FT30 - CYTOTOXIC ACTIVITY OF THIOSEMICARBAZONES DERIVED FROM AROMATIC ALDEHYDES
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Thiosemicarbazones (TSCs) are a class of compounds that have shown great interests for their biological activities. A series of TSCs, with different electronic groups, obtained from aromatic aldehydes (**1-5**) were synthesized using equimolar quantities of thiosemicarbazide in MeOH and AcOH 1%. The reaction mixture was refluxed for 8 h. The products obtained were filtered, recrystallized and their structures were confirmed by spectral data (IR, ¹H and ¹³C RMN).



Aldehydes and thiosemicarbazones were tested *in vitro* for their capacity to inhibit the growth of human (HBL-100, HeLa, SW1573, WiDri) and murine cell lines (CT26, B16). Cells were grown at 37 °C in a humidified atmosphere containing 5% CO₂ in DMEM medium, supplemented with fetal bovine serum, l-glutamine, sodium pyruvate, penicillin and streptomycin. Cytotoxicity activity was evaluated by MTT method. Absorbance of the solution was measured at 550 nm. Study of cell death was performed by flow cytometry. Thiosemicarbazones showed a potent growth inhibitory effect toward humans and murine cell lines. Halogenated TSCs were the most active compounds.

240.

FT31 - CADMIUM EXPOSITION ON REDOX BALANCE IN AORTA. EFFECT OF SOY BEAN AS PROTEIN SOURCE IN THE DIET

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Cadmium (Cd) is one of the most toxic pollutants. Its toxicity has been associated to cardiovascular alterations. Here, we study the oxidative effects of Cd in aorta and the antioxidant activity of soybeans, as protein source in the diet, as a preventive agent of redox alterations caused by Cd. 6 lots of adult male Wistar rats were conformed to 6 rats each. 3 lots received casein and 3 lots soybeans as protein dietary source, respectively. Within each protein group, 3 lots were conformed: one received water without Cd (control group) and the other two received 15 and 100 ppm of Cd (as Cl₂Cd) in the drinking water, respectively, for 60 days. In thoracic aorta the content of thiobarbituric acid reactive substance (TBARS), as indicator of lipoperoxidation, and the activities and protein levels (Western blot) of enzymes involved in the redox status were determined. Aorta of rats exposed to 100 ppm Cd and dietary casein showed an increase of NAD(P)H oxidase (NOX-2) protein, activity and protein expression of catalase (p<0.001) and glutathione peroxidase (p<0.01), and activity of superoxide dismutase. They were not modified with 15 ppm Cd, compared to control. TBARS, which were increased after 15 ppm Cd (p<0.01) returned to control values after 100 ppm Cd. Most of these changes did not occur when casein was replaced by soybeans in the diet.

241.

FT32 - SELF-MEDICATION IN STUDENTS OF NURSING AND NUTRITION IN SAN LUIS

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Self-medication is taking medication without medical intervention. Often the drug is used as a solution to any problem. Our goal was to analyze the use of nonprescription drugs, health problems for which they were consumed and the place of purchase. 139 interviews were conducted (May - June 2011), including: the existence or not of prescription, age, sex, diagnoses, medications, place of purchase. Drugs were classified according to ATC and potential therapeutic value, and the diagnoses according to ICD-10 classification. Results (%): self-medication 64.8. Sex: F 88.9, M 11.1. Age: <26 (78.9), 26-40 (11.1), >40 (10). Diagnoses: headache 16.7, menstrual cramps 11.3, fatigue 10, sore throat, 8.7, anxiety 8, stomach pain 7.7, cough 6.2, cold 5.9, sleep problems 3.9, decreased physical and intellectual performance 3.9. ATC Group: M 45.5, N 30.7, A 15.5, A 5.1, J 2.3. Drug: ibuprofen alone and combined (A,C) 39.2, paracetamol A,C 13.1, aspirin A,C 11.9, N-butylhioscina 5.1, propinox+clonixinate lysine 4, diclofenac 3.4. Fixed Dose Combinations (FDC) 23.9. Place of purchase: pharmacy 80.1, drugstore 12.5. Different types of pain, respiratory and digestive problems were prevalent. Non-steroidal, anti-inflammatory and analgesic antipyretic drugs were the most used then some medicines for the digestive and respiratory system. High self-medication, sale in drugstore, and the use of drugs in CDF of not high therapeutic value, as are the most cold medicines, lead us to conclude that it is necessary to develop strategies to avoid negative the negative connotations for the health which this misuse implies.

242.

FT33 - THE rs12255372(G/T) AND rs7903146(C/T) POLYMORPHISMS OF THE TCF7L2 GENE ARE ASSOCIATED WITH TYPE 2 DIABETES MELLITUS

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The Transcription Factor 7 like 2 (TCF7L2) is involved in the etiology of T2DM. The rs7903146 and rs12255372 polymorphisms have been associated with an increased risk of T2DM in multiple populations. The aim was study the frequencies of the polymorphisms rs12255372 (G / T) and rs7903146 (C/T) in the TCF7L2 gene in T2DM and control subjects. The polymorphisms were determined by the technique of Tetra Primer ARMS-PCR. The genotype frequencies of rs12255372 were 47.8% for GG, 47.8% for GT and 4.6% for TT in controls, in diabetics were 19.2% GG, 69.2% and 11.5% GT % TT. To rs 7903146 were 56.5% CC, 39.1% CT and 4.4% TT in controls, in diabetics were 26.9% CC, 50% CT and 23.1% TT. The frequency of the "T" allele of both rs12255372 (G/T) and rs7903146 (C/T) polymorphisms was significantly higher in diabetic subjects (46% and 48%) compared with control subjects (28% and 24%) (p<0,001 and p<0.0001, respectively). The presence of "T" allele of the rs12255372 (G/T) and rs7903146 (C/T) polymorphisms of TCF7L2 gene confer susceptibility to T2DM.

243.

FT34 - HOSPITALARY PHARMACOVIGILANCE IN SAN MARTIN (MENDOZA)

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Adverse drug reactions (ADRs) are a major public health problem and often occur during hospitalization or motivate it. Our objective was to study at a hospital in San Martín, Mendoza, the frequency of ADRs that occurred during hospitalization, analyze risk factors, and determine its severity, avoidance and causality. A retrospective observational study was carried out. Adverse events (AE), lack of efficacy (LE) and medication errors (ME) were recorded from medical records of 181 patients admitted in Medical Clinic, and 15 from other services, from 6/1/2009 to 5/31/2010, 8-10 days/month. The ADRs were classified by severity, causality, avoidance and causality. The possible association with sex, age, polypharmacy and comorbidity was analyzed. Results (%). AE 81.8, LE 16.4, ME 1.8. avoidable/ potentially avoidable 76.4. Proven 3.6, probable 38.2, possible 32.7. Severe 34.6, moderate 23.6, mild 41.8. Sex: M 59.7, F 40.3. The average age was 47.9 years old. Diagnoses associated with ADRs: general disorders 30.6, psychiatric 19.4, central and peripheral nervous system, gastrointestinal and cardiovascular 9.7, skin 6.5. Specific diagnoses: LE 16.4, psycho-motor excitation, pruritus and drowsiness 7.3. Medicines suspects: nervous system 27.4, cardiovascular 24.2, anti-infectives 22.6, systemic hormones 6.5; Individual drugs: clonazepam 9.1, dexamethasone 7.3, isosorbide mononitrate and ranitidine 5.5. Comorbidity was found as the only predictor of onset of RAM. Most were preventable. The high percentage of patients who had some ADRs is a major cause of morbidity due to the high number of severe reactions. This study highlights the need for closer monitoring of drug therapies and the importance of pharmacovigilance hospital as a tool for public health.

244.

FT37 - ANTIBIOTIC PRESCRIPTION IN HOSPITALS AND SOCIAL SECURITY OF SAN LUIS CITY

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The objective was to analyze the prescription of antibiotics (ATB) in two hospitals (A, B) and a social security (SS) of the city of San Luis. Prescriptions were collected for 1 month. Age, sex, diagnoses and prescriptions were recorded. Drugs and diagnoses were classified according to ATC (Anatomical Therapeutic Chemical) and ICD-10 (International Classification of Diseases) respectively. Results (%) for A, B and SS: age ≤15: 53.7, 48.8, 24, >15: 46.3, 51.2, 76. Sex: F 63, 58.6, 64.8, M 37, 41.4, 35.2 Prescription ATB: 20.2; 16.3, 8.4. Mono-drugs 100, 98.8, 72.4. Fixed-dose combinations (FDC) 0, 1.2, 27.6. ATB prevalent and indications for ICD-10 groups for A, B and SS respectively: amoxicillin 58.7, 50, 13 for respiratory (J) 76.5, 84.2, 51, for abnormal symptoms and signs (R) 7.5, 5.3, 1.9, alimentary tract (K) 6, 1.9, 29.4, eye and ear (H) 2.5, 7.2, 5.8; genitourinary (N) 2.5, 0.4, 7.8. Cephalexin 24.9, 21.3, 10.7; for N 16.5, 23.6, 16.6; skin and subcutaneous tissue (L) 20, 20.2, 33.3; injury, poisoning and other consequences of external causes (S and T) 27, 15.7, 14.3; J 8.2, 12.4, 2.4. Penicillin G 4.1, 12.2, 5.6 for J 85.7, 88.2, 63.6. Ciprofloxacin 8.8, 3.8, 5.8 for N 86.7, 56.2, 60.9. Amoxicillin+clavulanic acid 14.3 (SS) to J 55.4, K 16, H 14.3. In A, more children were treated, in B and SS adults, and women prevailed. In SS ATB in FCD were prescribed. In A and B amoxicillin was prevalent followed by cephalexin, in SS amoxicillin+clavulanate was followed by amoxicillin. The ATB were prescribed for respiratory diseases, followed by genitourinary in A and B, and digestive in SS.

245.

FT40 - HYPERTENSION, CO-MORBIDITY AND USE OF ANTIHYPERTENSIVE DRUGS IN A HEALTH CENTER OF GODOY CRUZ (MZA.)Palomo V¹, Manucha W², Calderón C³.¹Min. Salud Mendoza, ²Fac. Medicina-UNCuyo, ³FQBF-UNSL. E-mail: ccal@unsl.edu.ar

The Health Care District of Godoy Cruz consists of 16 health centers, in one of them, the Health Center 149, a cardiologist works. Our goal was to analyze the prevalence of hypertension (HTA), the associated pathologies and drug consumption in this center. For this reason we analyzed the medical records of hypertensive patients who attended the Health Center from April to June 2011. Age, sex, principal diagnosis, secondary diagnoses and prescription medications were collected. The guide of the area, the International Classification of Diseases (ICD-10) and the anatomical-therapeutic-chemical classification (ATC) were used. Results: Patients with hypertension 90%. Sex: F 71% M 29%. Age: 55-79 years (45%). Secondary diagnoses: 40% had 2 and 22% 3. 54% of hypertensive patients had hypercholesterolemia, 27% anxiety, 23% overweight, and 17% hypothyroidism. 67% of patients with HTA taken from 1 to 4 drugs and 33% of 5 to 10. Enalapril (E) were prescribed in the 70%, and among them, 50% associated with 1 or 2 diuretics (hydrochlorothiazide, spironolactone, furosemide), and 45% to another antihypertensive (atenolol 78%, amlodipine 22%). E+hypocholesterolemic was prescribed 30%, E+anxiolytic 25%, E+levothyroxine 20%. It is of major concern to us the high percentage of associated pathologies and polypharmacy in patients with hypertension, the same as factors that predispose to adverse reactions.

246.

FT41 - CHANGES IN AROMATASE EXPRESSION AFTER EXPOSURE TO 17 β - ESTRADIOL AT ENVIRONMENTAL LEVELSGuyón NF^{1,3}, Amé MV^{2,3}, Roggio MA^{1,3}, Wunderlin DA^{2,3}, Bistoni MA¹.¹Cát Div Anim II, FCEFYn; ²Dto Bioq Clín, FCQ, UNC; ³CONICET. E-mail: nguyon@efn.uncor.edu

There is growing evidence that environmental estrogens can reach high enough levels to exert adverse reproductive effects on wild fish populations. Some chemical compounds can affect the enzyme cytochrome P450 aromatase, which catalyze the conversion of androgens to estrogens. The main goal of this work was to assess the effect of chronic exposure to environmentally relevant concentrations of 17 β -estradiol (E₂) on *cyp19a1a* (gonadal aromatase) and *cyp19a1b* (brain aromatase) gene expressions. Eighty adult males of *Jenynsia multidentata* (Anablepidae, Cyprinodontiformes) were exposed to 0, 50, 100 and 250 ng/L E₂ during 28 d. Brain and gonads were excised, total RNA was extracted and gene expressions were measure by real time PCR. Our findings demonstrated that E₂ exposure resulted in a very clear increase in brain aromatase transcript abundance at all assayed concentrations in comparison to control. Although there were not significant differences among treatments, a trend toward increased *cyp19a1b* transcript abundance in a concentration-dependent manner was observed. However, no effects on gonadal aromatase expression were observed. The remarkable changes observed in *cyp19a1b* gene expression suggest this parameter as a sensitive and potentially useful biomarker for exposure to environmentally relevant estrogenic compounds.

247.

FT42 - IMPAIRMENTS OF MALE FISH SEXUAL BEHAVIOR AS BIOMARKER OF XENOESTROGEN EXPOSURE

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The reproductive biology of *Jenynsia multidentata* (Anablepidae: Cyprinodontiformes) provides a useful model to study the effects of xenobiotic compounds. It is viviparous fish species and presents sexual dimorphism. Mating behavior is coercive. Our main goal was to evaluate alterations in reproductive behavior of *J. multidentata* using behavior biomarkers to xenoestrogen exposition. Adult males ($n=15$) were exposed for 28 days at 17 β -estradiol (50, 100 and 250 ng/L E₂), 17 α -etinilestradiol (10, 75 and 250 ng/L EE) and 4*n*-nonilfenol (1 and 10 μ g/L 4*n*-NP). For each treatment there was a control group. At the end of the experiment, sexual activity was registered for 20 min and different behavioral parameters such as number of persecutions, copulatory attempts and number of copulations, were estimated. Relations among these variables were calculated in order to evaluate the effectiveness of male behavior. At the lowest concentration of E₂ it was registered an exacerbation of sexual behavior. The 4*n*-NP exposure caused a decrease in some parameters, such as the number of copulations while in EE exposure all the variables significantly decreased respect to the control group. According to these results, we concluded that the xenoestrogen affect the reproductive behavior of *J. multidentata*. The behavior parameters analyzed could be considered as useful exposition biomarkers to study the effects of xenoestrogens on male sexual activity.

248.

FT43 - ASSESSMENT OF GENOTOXIC POTENTIAL OF INFUSIONS OF *Jungia polita* USING *Allium cepa* AS ASSAY SYSTEM

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One way of analyzing the genotoxicity is to examine the apical meristem of *Allium cepa*. This is an efficient experimental system to study the presence of genotoxic and mutagenic agents in different environments. *Jungia polita* Griseb. (Asteraceae) n.v. "zarzaparrilla" is used as a depurative and anti-sclerotic in popular medicine. The objective of this work is to analyze the genotoxicity of infusions of *J. polita* by the experimental system *A. cepa*. To that end, infusions of *J. polita* at 10%, 20% and 30% were prepared. Bulbs of *A. cepa* were placed in distilled water as control group and in three experimental groups with the infusion at 10%, 20% and 30%, respectively for 24 hours. Then, the roots were fixed with Carnoy and dyed with carmine. Approximately 3000 cells were analyzed by treatment. The Mitotic Index (MI) and the deterioration produced to the genetic material were calculated. The optic microscopy revealed absence of chromosomal aberrations in the negative control and in the three experimental groups. The cytogenetic analysis showed the presence of nuclear fragmentation and condensation in the cell populations exposed to the three concentrations of infusion. The MI of control was 9% and 0% in the three concentrations studied. These results suggest that the infusions of *J. polita* tested may activate intracellular molecular signals which may lead to the detention of growth and to cell death by apoptosis or necrosis.

249.

FT44 - STUDY OF GENOTOXIC EFFECTS OF INFUSIONS OF *Prosopis torquata* IN MERISTEMATIC CELLS OF *Allium cepa*

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FQByF. UNSL. IMIBIO. CONICET. E-mail: macang@unsl.edu.ar

In order to detect possible genetic damage is needed to develop complementary genotoxic and pharmacological experiments. The *Allium cepa* test system allows to evaluate DNA damage and level of disturbance in the mitotic cycle. The genus *Prosopis* has fruits that are high in protein and carbohydrates that vary according to each species studied and are used both for human and animal pharmaceutical purposes. The objective of this study was to assess the genotoxic effect of infusions of the fruit of *P. torquata* in meristematic cells of *A. cepa* strain, to determine their subsequent use in food and pharmacy. The infusions were prepared from the fruit of *P. torquata* to 10, 20 and 30%. *A. cepa* bulbs were placed in distilled water as a control group and in three experimental groups with the infusion at 10%, 20% and 30% respectively for 24 hours. Then, the roots were fixed with Carnoy and dyed with carmine. Approximately 4000 cells were analyzed per each treatment. The Mitotic Index (IM) and the impairment of genetic material was calculated. The microscopic analysis revealed DNA damage in proportion to the concentration in the infusion. It was observed that dissolution at 30% produce up to 100% of cells with signs of apoptosis. The IM control was 11% and 0% in the three concentrations studied. These results suggest that infusions of *P. torquata* produce a cytostatic effect rather than a clastogenic effect in the concentrations studied as they completely inhibit cell division and produce a marked genetic damage in the cell population treated with the infusion of 30%.

250.

FT45 - BIOMONITORING OF GENETIC DAMAGE IN HUMANS EXPOSED TO AGROCHEMICALSPeralta L³, Mañas F³, Gentile N¹, Bosch B¹, Gorla N², Aiassa D¹.¹Depto Ciencias Naturales, FCEyN, UNRC; ²CONICET; ³Depto. Clínica, FAV, UNRC.

Using tests of genotoxicity in peripheral blood of people exposed to pesticides is possible to determine the effects of such exposure on integrity of genetic material. The quantification of genotoxicity was carried out in two groups: one for exposed to agrochemicals in different locations in Cordoba, and a control group not exposed. Were carried out tests for Chromosomal aberrations (CA), Micronuclei (MN) and Comet. The CA test showed 2.36 ± 1.74 and 4.68 ± 3.55 aberrations, the MN test 7.25 ± 1.48 , and 10.81 ± 5.21 and Comet assay 115.1 ± 71.11 and 3037 ± 3731 arbitrary units in the control and exposed groups respectively in each case (the results are expressed as mean \pm standard deviation). We found a statistically significant increase in the value of genetic damage in the group exposed to agrochemicals in relation to the control group in all three trials. These results suggest that exposure to chemicals causes an increase in genetic damage, which can be detected by tests for genotoxicity. Genotoxicity assays allow us to detect and quantify the damage even when it is still reversible and thus can prevent or reduce the exposure time.

251.

FT46 - DETERMINATION OF ZINC TRACES IN WATER SAMPLES FROM GUANACACHE LAKE BY SOLID PHASE LUMINESCENCEVega M¹, Augusto M¹, Talio M², Fernández L².¹Inst. Cs. Básicas, FFHA, UNSJ- San Juan; ²Fac. Qca., Bqca y Fcia, UNSL-INQUISAL-CONICET, 5700 - San Luis. E-mail: lfernand@unsl.edu.ar

Zinc plays an important role in several biochemical processes; however, if it is in excess, this metal can also produce damage in the human body, including arrhythmias/dysrhythmias, increase susceptibility to autoimmune reactions, between others.

Chemofiltration on nylon membranes pre-treated with hexadecyltrimethylammonium bromide (HTAB) and eosin dye (eo) is proposed for zinc traces quantification by solid surplus spectrofluorimetry ($\lambda_{exc} = 532$ nm; $\lambda_{em} = 548$ nm). At optimal experimental conditions, quantitative recovery was reached with a detection limit of 0.662 $\mu\text{g L}^{-1}$ and quantification limit of 2.20 $\mu\text{g L}^{-1}$. The calibration sensitivity was of 1.22 $\text{L } \mu\text{g}^{-1}$ for the new methodology with a linear range of 2.20 $\mu\text{g L}^{-1}$ to 779 $\mu\text{g L}^{-1}$ Zn(II). The tolerance levels of potential interfering ions were studied with good results. The methodology was validated by standard addition method and satisfactorily applied to Zn(II) contents determinations of water samples arising from Guanacache Lake (San Juan, Argentine) without previous treatment. The performing obtained in sensitivity and selectivity thanks to chemofiltration step, converts the proposed methodology in an adequate alternative to conventional techniques for Zn(II) traces determination.

252.

FT47 - SALURETIC EFFECT OF THE INFUSION OF *Cuphea glutinosa* CHAM ET SCHLTDL (LYTHRACEAE) IN RATS

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Cuphea glutinosa (Lythraceae), popularly known as "Siete sangrias or Sanguinaria", has wide use in the folk-medicine practice, especially as diuretic and hypotensor. The study evaluates the saluretic activity of different concentrations infusions of this. Three infusions doses (2.5%, 5% y 10%) of *Cuphea glutinosa* (5 mL/ kg body weight) and Furosemide (10 mg/ kg body weight) were orally administered to rats. The electrolytes levels, pH, density, and other parameters were measured in the urine of saline-loaded rats. The oral administration of 2.5%, 5% and 10% of infusion increased significantly the K^+ excretion to compare it with that induced by control, but it was less than the one induced by Furosemide. However the treatments did not increase significantly the Na^+ excretion levels. Other parameters were unaffected. The results suggest that the infusion of *Cuphea glutinosa* induce kaliuretic response and it's not effective to induce natriuretic response.

253.

FT48 – ANTIALLERGIC HERBS (*LATO SENSU*) FROM WEST-CENTRAL ARGENTINEAN ETHNOMEDICINE

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Many plants or their active ingredients are known for their anti-genic effects, but many more are used as a broadly antiallergic. Countered allergies and inflammatory conditions, and improve performance of the immune system, eventually turn to body balance and harmony's characteristics of the health state. These plants show antihistamines, antiseptics, emollients, balsamic, expectorant, anti-inflammatory effects and even contribute to the healing of skin and mucous membranes, relieving dermatitis, respiratory allergies, rhinitis, blepharitis, hayfever, sneezing, rashes, itching, etc. We collected and analyzed both folk and validated data obtained through semi-structured surveys of connoisseurs, healers and regional markets, and ethnomedical references. We found 64 species cited as anti-allergic (*lato sensu*) belonging to 35 families, highlighting Asteraceae (22%), Lamiaceae & Plantaginaceae (10%), and Ephedraceae, Fabaceae, Magnoliaceae, Polygonaceae, Rosaceae & Verbenaceae (3%). 26 families bring one species each. Four species (6%) are excluded for phytotherapies (ANMAT 1788/00) and another 4 are in the allowed list (ANMAT 1637/01). 41% are official drugs in 8 Pharmacopoeias and 19% take part of remedies in Argentinean vademecum, of which 8% are not official drugs. 49% are sold in health food stores and pharmacies in the west-central Argentina and only 1 of those making up the restrictive list was found in trade. 17% of all species are native to west-central Argentina.

254.

FT49 - EFFECT OF *Amaranthus. hypochondriacus* ON CHOLESTEROL METABOLISM IN RATS INTOXICATED WITH ETHANOL

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The aim of this study was to evaluate in the liver of male *Wistar* rat treated with ethanol, the potential beneficial role of the seed of *Amaranthus hypochondriacus* (*Ah*) on the expression of genes involved in cholesterol metabolism: 3-hydroxy-3-methyl-glutaryl-CoA reductase (HMGCoAR), LDL receptor (LDLr), sterol regulatory element binding protein 2 (SREBP-2) and cholesterol 7 α -hydroxylase (CYP7A1). The animals were divided into four groups of six subjects each, two of those groups were fed diet AIN-93M containing casein as protein source, and the other two with AIN-93M containing *Ah*. One of each protein group received 20% ethanol in the drinking water, being: *AhC* (*Ah* control) and CC (casein control), *AhE* (*Ah* ethanol) and CE (casein ethanol). The experiment was performed for 4 weeks. The mRNA levels of the molecules were estimated by RT-PCR. The *AhE* group showed a decrease in the expression of HMGCoAR and an increase in that of LDLr compared with CE (P < 0.001) and *AhC* (P < 0.01). HMGCoAR mRNA levels increased in CE compared with CC (P < 0.001). The *AhE* presented a trend of decrease of SREBP-2 respect to CE. CYP7A1 was not significantly different among groups. We conclude that in rats under the influence of ethanol, *Amaranthus hypochondriacus* would contribute to improve cholesterol metabolism.

255.

FT50 - EFFECT OF SOYBEAN MEAL ON THE EXPRESSION OF PPAR α AND CPT-I IN RATS FED HIGH-CALORIE DIET

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PPAR α (peroxisome proliferator-activated receptor) and CPT-I (carnitine palmitoyltransferase I) have a key role in the catabolism of fatty acids. The aim of this study was to evaluate the gene expressions of PPAR α and CPT-I in the liver of rats fed high-calorie diets made with different protein sources such as soybean and casein. The animals were divided into two groups: control, fed AIN-93 diet, and experimental, fed AIN-93 diet high-calorie (34.15% sucrose, 42% of calories from fat) for 9 weeks. After that, each group was divided into two: one using casein and the other one using soybean as protein source, respectively. Animals were fed for 45 days and then sacrificed. Thus, the groups being: CC (control casein), CS (control soybean), HC (high-calorie casein) and HS (high-calorie soybean). PPAR α and CPT-I mRNA expressions were estimated in the liver tissue by RT-PCR. In CS group, PPAR α and CPT-I expressions were increased in relation to CC (P < 0.01) while in HS group both transcripts were decreased compared with HC (P < 0.01). Both genes showed a lower expression with the high-calorie soy diet than their controls (P < 0.001). Soy bean, as protein source in the normal-calorie diet, could increase the fatty acids transport into the mitochondria inducing their catabolism. This favorable behavior of soy is not observed with a high-calorie diet.

256.

FT51 - MINERAL CONTENT FRUITS'S SEEDS FROM *Ramorinoa girolae* Speg. (FABACEAE)Luna LC¹, Monferran M², Feresin GE¹, Wunderlin DA², Tapia A¹.¹IBT. ICB-UNSJ, San Juan, ²ISIDSA, UNC, Córdoba, Argentina. E-

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The determination of minerals and trace elements in food is an important part of nutritional and toxicological analyses. *Ramorinoa girolae* Speg. (Fabaceae) vernacular name "chica" is argentinian monotypic genus of Fabaceae. Following with our study on nutritional properties and chemical composition of this specie, twenty nine elements were quantified in seeds of *R. girolae* samples: Li, Be, B, Na, Mg, Al, K, Ca, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, Ga, As, Se, Rb, Sr, Mo, Ag, Cd, Te, Ba, Tl, Pb and Bi. Results have shown that K, Mg, and Ca were major minerals of these seeds. Na, Fe, Zn, B, Cu, Mn, Al, Sr, Rb, Mo and Be contents of seeds were found to be very low. Potassium was quantitatively the most important metal, accounting for 59% of the metal contents, having a concentration of 8174.76 mg Kg⁻¹. Magnesium and calcium were present in moderate amounts, accounting for 22% and 17% of total minerals, respectively. Fifteen of twenty nine minerals were below LOD ($\mu\text{g g}^{-1}$): Li, V, Cr, Co, Ni, Ga, As, Se, Cd, Ag, Te, Ba, Tl, Pb and Bi was upper detection limit. In addition, these elements play an important role in human metabolism, and interest in these elements is increasing together with reports of relationships between trace element status and oxidative diseases. This is the first report of mineral content and beneficial effects of the seeds of *R. girolae*.

We are grateful to UNSJ, ANPCYT (PICT 2008-0554) and CONICET. (LL, GEF and DAW)

257.

FT52 - PHENOLICS, FLAVONOIDS AND ANTIOXIDANT ACTIVITY OF *Tagetes* spp. FROM TUCUMÁNVidal EA¹, Luna L¹, Tereschuk ML², Slanis A³, González M², Feresin GE¹, Tapia A¹.¹IBT. ICB-UNSJ, San Juan ²FACET-UNT, ³FCNeIML-UNT y FML, Tucumán. E-mail: lluna@unsj.edu.ar

In recent years, a significant revival in the search for compounds from natural sources with antioxidant activity. The genus *Tagetes* includes 12 species that grow wild in Argentina. The aim of this study was determine the content of total phenolics, flavonoids and antioxidant activity. Dichloromethane (EDCM) and methanol (EM) extracts of four *Tagetes* spp growing from Tucumán Province: *T. minuta* L., *T. rupestris* Cabrera, *T. pusilla* Kunth and *T. campanulata* Griseb. were assessed through the scavenging effects on radical DPPH, FRAP (ferric reducing-antioxidant power) in order to evaluate the antioxidant capacity. EM of *T. rupestris* and *T. minuta* showed the highest percentages of phenolics (>21% and >20% GAE/100g extract, respectively); and flavonoids (>10% and >7% QE/100g extract, respectively). In addition, this species showed the highest antioxidant activity (>89% and >90% of DPPH decoloration at 10 µg/mL) and stronger FRAP a 200 µg/mL (0,419 y 0,446, respectively), compared with standard gallic acid at 10 µg/mL. These species studied so far are an excellent source of antioxidant compounds. We are grateful to UNSJ, ANPCYT (PICT2008-0554), UNT. CIUNT (26E/401) and CONICET. (L.L. and G.E.F.)

258.

FT53 - POLYPHENOLS AND ANTIOXIDANT ACTIVITIES OF *Pistacia vera* var Kerman ARGENTINEAN CULTIVARSFabani MP¹, Luna L¹, Ighani M², Tapia A¹, Wunderlin DA³, Feresin GE¹.¹Instituto de Biotecnología-UNSJ. ²Empresa Pisté – Pistachos Argentinos. ³UNC-ISIDSA-CIBICI.

Today, nuts consumption is being promoted due they have an important role in preventing several diseases associated with oxidative stress. The aim of this study was to evaluate polyphenolic content, total anthocyanins and antioxidant activity of *Pistacia vera* cv Kerman from San Juan (Argentina). Methanol acidic extracts defatted were obtained (MeOH-H⁺E) from cultivars of five, nine and eleven years. Total phenolics (TP), flavonoids (FL) and total anthocyanins (TA) were quantified. Antioxidant capacity was determined by DPPH and FRAP assay. TP mean value ranged from 292.72 to 528.88 mg GAE/100 g DW whereas FL oscillated between 9.76 and 20.64 mg QE/100 g DW. AT mean concentration varied from 0.66 to 1.15 µg cyanidin-3-glucoside/100 g DW and significant differences among cultivars were observed ($p < 0.05$). Antioxidant activity of MeOH-H⁺E was high and presented a positive and significant correlation ($r = 0.79$) between TP and antioxidant activity (DPPH). Argentinean pistachios are a good source of polyphenolic compounds with high antioxidant activity which contributes to maintaining health, preventing the deterioration that occurs from free radicals. This is the first report of chemical profile and antioxidant activity of Argentine cultivars of *Pistacia vera*. Acknowledgements: UNSJ and ANPCYT (PICT 2008-0554). FMP and LL the fellowships CONICET. FG and WD are researchers of CONICET, Argentina.

259.

FT54 - MINERAL NUTRIENTS AND HEAVY METALS OF *Pistacia vera* var Kerman ARGENTINEAN CULTIVARSFabani MP¹, Monferran M², Ighani M³, Tapia A¹, Wunderlin DA², Feresin GE¹.¹Instituto de Biotecnología-UNSJ. ²UNC-ISIDSA-CIBICI. ³Empresa Pisté – Pistachos Argentinos.

Nuts (walnuts, raisins, almonds) are an excellent source of essential trace elements in the diet. The aim was to evaluate the content of mineral nutrients and heavy metals of *Pistacia vera* var Kerman from San Juan (Argentina). Unroasted pistachios of five, nine and eleven cultivars years old were used to quantify twenty-nine elements by Q-ICP-MS. K was the most abundant, followed by Ca and Mg. The average content of minor elements decreased as follows: Na>Fe>Zn>Cu>Mn. B, Ba, Be, Cr, Li, Mo, and Se were not detected. Since metal contamination could take place during handling and processing of pistachio nuts, the presence of Ag, Al, As, Bi, Cd, Co Ga, Ni, Pb, Te, Tl and V were analyzed. Only aluminium was detected above the detection limit. Compared with other nuts, pistachios contain minerals in appreciable amounts beneficial to maintain the physiological conditions; in addition Na intake is very low, which is very important for health, especially hypertensive individuals. This work is the first report of mineral nutrients content and heavy metals in pistachio seeds cv Kerman from province of San Juan (Argentina).

Acknowledgements: UNSJ and ANPCYT (PICT 2008-0554). FMP the fellowship CONICET. FG, MM and WD are researchers of CONICET, Argentina.

260.

FT55 - ANTIMICROBIAL AND ANTIOXIDANT ACTIVITIES OF HONEY FROM TUDCUM, SAN JUAN PROVINCE, ARGENTINA

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Honey is a sweet and flavorful natural product which is consumed in San Juan province, Argentine for its high nutritive value and to treat diseases related to the presence of human pathogenic bacteria, as cold, cough and flu states. Honeys from Tudcum district, (Iglesia, San Juan, Argentina) were assessed against a panel of standardized and clinical isolates of pathogenic bacteria. The antioxidant capacity was assessed through the scavenging effects on radical DPPH and the FRAP (ferric reducing-antioxidant power) assay. The MIC values were determined using the microbroth dilution method according to protocols of the NCCLS. Results showed that methicillin-resistant *Staphylococcus aureus* ATCC 43300 *Escherichia coli* ATCC 25922, *Escherichia coli* LM1, *Escherichia coli* LM2, *Pseudomonas aeruginosa* ATCC 27853, *Yersinia enterocolitica* PI, *Salmonella enteritidis* MI and *Salmonella sp.* LM strains were inhibited with MICs values between 1500 and 2000 µg/ml. A moderated DPPH activity was found for honey (>20% at 20 mg/ml). The good antibacterial activity of honey against clinical bacterial isolates indicates the usefulness of honey in traditional medicine against bacterial infections.

We are grateful to UNSJ, ANPCYT (PICT 2008-0554); and CONICET (BL, LL and GEF)

261.

FT56 - MORTALITY EFFECTS OF SEMI-SYNTHETIC DITERPENES ON *Tenebrio molitor*

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Natural products (NP) have extensive structural and functional diversity and provide research topics for chemists, biologists and doctors. The use of synthetic pesticides for controlling insects has caused resistance, emergence of new pests, pollution and other damage resulting from accidental misuse and application. The NP of plants may be an option for an integrated management program and emerge as an alternative for controlling stored grain pests.

Previously we report the insect growth regulatory effects of natural labdanes from *Grindelia pulchella* Dunal var. pulchella, in this study we obtained 12 semi-synthetic derivatives of the natural metabolites and we show the effects they produced on adults of *Tenebrio molitor*. The mortality of larvae of *T. molitor* was studied by toxicity tests by topical application of the compounds and they were monitored every 24 hours until day 5, showing mortality values between 45 and 100%. The best results against insects were obtained with side chain oxygenated products.

Acknowledgements: This work was supported by CONICET (PIP0628), ANPCyT (PICT-UNSL 2008-0022) UNSL (PROICO-7301) and AECI (PCIA / 025750/09).

262.

FT57 - BIOACTIVITY AND PRELIMINAR PHYTOCHEMICAL STUDIES OF *Nepeta cataria* L.

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The genus *Nepeta* (Lamiaceae) is comprised of approximately 250 annual and perennial species which are native to temperate Europe, Asia, North Africa, and the mountainous regions of tropical Africa. Some of these species are well known for their medicinal properties and are widely used in folk medicine. The pharmacological properties and various biological activities are usually ascribed to nepetalactone compounds primarily found in the essential oils of the *Nepeta* species.

As part of a systematic search of potential biopesticides, repellency and toxicity properties of a mixture of two nepetalactone diastereomeric and extracts of *Nepeta cataria* L. were evaluated using *Tribolium castaneum* Herbst (Coleoptera) Tenebrionidae. The results of repellency in the bioassay show that the methanol extract presented a significant repellency.

In the case, of the diastereomeric mixture nepetalactone, insects had a high mortality rate, although the rate of repellency of these products was not significant. Toxicity was evaluated by exposure to the contact surface and found that the mortality rate was 100% at 48 hours after starting the assay.

This work was supported by CONICET (PIP0628), ANPCyT (PICT-UNSL 2008-0022) UNSL (PROICO-7301) and AECI (PCIA / 025750/09).

263.

IM3 - DIFFERENT PROFILE OF RESPONSE TO Th1/Th2 STIMULUS BY CLARA CELL

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Clara cells (CC) contribute to lung Th1/Th2 homeostasis by secreting innate immunity molecules (ii) such as SP-D and CC16. Asthma is a Th2-type chronic inflammatory disease; its current therapeutic uses Th1 stimulation with ligands of TLRs to restore the balance. Our aim was address the response of CC to Th1/Th2 stimulus using LPS from *E. coli* and an OVA asthma model respectively. In OVA group, females BALB/c (n=30) were inoculated i.p with 0.1ml of OVA/alum (1mg/ml) at day 0 and 14 and then challenged with 50µl of OVA (1mg/ml) or vehicle i.n on days 24 to 34; LPS group received 50µl of LPS(0.2mg/ml) or vehicle i.n and sacrificed at 4, 6, 8 or 24 h. Morphological changes were evaluated by light and electron microscopy, CC16, SP-D, TNFα and Epidermal Growth Factor receptor (EGFR) content by immunostaining, and mucin by AB-PAS. Bronchoalveolar lavage was used to analyze the profile of cells and TNFα secretion by ELISA. OVA showed mucous metaplasia of CC (p <0.001), SP-D and CC16 decreased, and apical EGFR increased. Besides, it exhibited infiltration of eosinophils (p<0.001) and hypertrophy of muscular layer strongly marked with TNFα. While LPS showed changes at 6h as CC hypertrophy, increased of CC16, SP-D, TLR4 and TNFα, infiltration of neutrophils (p <0.001) and peak in TNFα (p<0.001), changes were reversed at 24 h. We conclude that CC are capable of responding specifically to Th1/Th2 stimulus, and Th1 signaling enhance ii components of CC which could regulate the allergic inflammatory response.

264.

IM6 - IDENTIFICATION OF *Clostridium chauvoei* IMMUNOREACTIVE PROTEINS BY MASS SPECTROMETRY

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Clostridium chauvoei is an anaerobic bacillus, causative agent of blackleg, a fatal disease that affects mainly cattle and sheep. The immunity of *C. chauvoei* is considered anticellular, being the somatic and flagellar antigens the most studied. The aim of this work was identified by mass spectrometry and partial protein sequencing immunogenic proteins of *C. chauvoei*. We used the strain *C. chauvoei* ATCC 10092 The cellular soluble proteins were separated by gel electrophoresis (1D SDS-PAGE) The protein bands were cut, faded, reduced, alkylated and digested with trypsin prior to being injected on a LC-ESI-MSMS. The following proteins were identified: pyruvate flavodoxin oxidoreductase (130 kDa); heat shock proteins HSP-70 (66-kDa) protein flagellin FLIACS (34 - kDa); spore germination protein (42 - kDa) fractions of lower PM flagellin (23 k-Da). It was possible to identify the main immunoreactive proteins involved in the immune response of *C. chauvoei*. The results confirmed the importance of flagellar antigens in *C. chauvoei* vaccine formulation. Furthermore, this work is the first to identify *C. chauvoei* immunoreactive proteins that are considered vaccines candidates in other clostridium species.

265. MI3 – EFFECTIVE BINARY AND TERNARY ANTIMICROBIAL FLAVONOIDS COMBINATIONS WITH NALIDIXIC ACID AGAINST *Escherichia coli* ATCC 25 922

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E. coli is an important pathogen associated with a large number of infections in humans. The present work aims to determine the synergistic effect of dihydroxychalcones-nalidixic acid (NA) and dihydroxychalcones- NA- rutin (Ru) combinations against this Gram (-) bacterium. Using a kinetic turbidimetric method developed previously, the antimicrobial activity of 2',3-(OH)₂-chalcone, 2',4-(OH)₂-chalcone and 2',4'-(OH)₂-chalcone, its combinations with NA (constant concentration: 2 µg/mL) and its combinations with NA (constant concentration: 2 µg/mL) - Ru (constant concentration: 20 µg/mL) were assayed. The application of an action mechanism allowed minimal inhibitory concentrations (MICs) evaluation. In the Table MICs values (µg/mL) are informed for chalcones alone and its binary and ternary combinations.

2',3-(OH) ₂ -chalc			2',4-(OH) ₂ -chalc			2',4'-(OH) ₂ -chalc		
0	NA	NARu	0	NA	NARu	0	NA	NARu
121.5	76.5	52.5	75.8	44.2	28.1	74.8	57.5	40.0

All combinations assayed showed synergism. Although 2',3-(OH)₂-chalcone-Ru-NA combination shows bigger synergic effect, 2',4-(OH)₂-chalcone-Ru-NA combination was more efficient against *E.coli* ATCC 25 922 (MIC: 28.1 µg/mL).

266. MI4 - 2',4-DIACETYLCHALCONE PREPARATION AND ANTIBACTERIAL ACTIVITY AGAINST *Escherichia coli* ATCC 25 922

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In the present study, 2',4-dihydroxychalcone, a compound with higher antibacterial activity against *E. coli*, was selected for to obtain 2',4-diacetylchalcone. For dihydroxylated chalcone treatment with pyridine, dimethylaminopyridine and acetic anhydride yielded a product that was identified by NMR. Using a kinetic turbidimetric method developed previously, the antimicrobial activity of 2',4-diacetylchalcone and its combinations with nalidixic acid (NA) (constant concentration: 2 µg/mL) and NA (constant concentration: 2 µg/mL)-Rutin (Ru) (constant concentration: 20 µg/mL) were assayed. The minimal inhibitory concentrations (MICs in µg/mL) were evaluated and informed for both compounds alone and its binary and ternary combinations.

2',4-dihydroxychalcone			2',4-diacetylchalcone		
0	NA	NA-Ru	0	NA	NA-Ru
75.8	44.2	28.1	151.0	91.4	139

The derivative obtained showed less antimicrobial effect against *E. coli* (MIC: 151 µg/mL) and only 2',4-diacetylchalcone-NA combination showed synergism (MIC: 91.4 µg/mL). Further research will attempt to incorporate other functionalizations on the hydroxyl groups such as trimethylsilyl and diphenyltertbutylsilyl to enhance the bacteriostatic activity of hydroxylated flavonoids.

267. MI6 - PREVALENCE OF *Streptococcus agalactiae* COLONIZATION IN PREGNANT WOMEN IN THE CITY OF SAN LUIS. SENSITIVITY PROFILE AND RESISTANCE TO MACROLIDES

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Streptococcus agalactiae is part of the microbiota of the gastrointestinal tract, where colonizing the genital tract, which is important in pregnant women by the possibility of transmission to the newborn and subsequent risk of developing severe infections in newborns. The aim of this study was identifying the frequency of *S. agalactiae* maternal colonization in pregnant women in San Luis and perform a surveillance study of resistance to macrolides accompanied by the characterization of the phenotypes of resistance involved. We analyzed 324 samples of pregnant women. Samples were taken between 35-37 weeks of gestation. Vaginal and perianal swabs were taken from both the vaginal introitus and the anorectus and were inoculated into selective Todd Hewitt broth 24 h at 37°C, and the subculture in sheep blood agar 24 h at 37°C with CO₂(5%). Suspicious colonies were identified by conventional biochemical tests. Of the 324 pregnant women included in the study in 21 of them was isolate *S. agalactiae*. Corresponding to a total prevalence of 6.5%. To perform the susceptibility of the isolates by the disk diffusion test was found that they were sensitive to penicillin and vancomycin and only one strain showed resistance to clindamycin and erythromycin expressing a constitutive MLS_B resistance phenotype. Determine the prevalence of carriage of *S. agalactiae* in a given region and the sensitivity of this organism to antibiotics, is essential to establish antibiotic prophylaxis and thereby decrease the rate of transmission to the newborn as well as reduce the incidence of severe infections.

268. MI7 - PREVALENCE OF *Streptococcus agalactiae* COLONIZATION IN PREGNANT WOMEN IN THE CITY OF SAN LUIS ACCORDING TO AGE AND UNDERLYING DISEASES

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Streptococcus agalactiae are involved in various infectious processes in susceptible adults. Is now known mainly as the causative agent of invasive disease in neonates and infants under three months, causing sepsis, pneumonia and meningitis. The aim of this study was known the prevalence of *S. agalactiae* colonization in pregnant women according to age and underlying diseases. We analyzed 324 samples of pregnant women, 230 of the Clinic and Maternity CERHU and 94 of Hanna Abdallah Health Center. Samples were taken between 35-37 weeks of gestation. Vaginal and perianal swabs were taken from both the vaginal introitus and were inoculated into selective Todd Hewitt, and the subculture was performed in sheep blood agar 24 h at 37°C with CO₂(5%). Suspicious colonies were identified by conventional biochemical tests. Patients were divided into 5 age groups as follows: 16 to 20 years, from 21 to 25, from 26 to 30, from 31 to 35 and 36 to 41 years. From 21 pregnant women was isolate *S. agalactiae*, corresponding to a total prevalence of 6.5%. The *S. agalactiae* carrier prevalence in CERHU was 6.5% against 6.4% of Hanna Abdallah. Porting analyzing the prevalence according to age of the patients showed no significant differences between groups analyzed (p < 0.11), although a trend higher in patient older than 35 years. To relate porting versus underlying diseases, it was found that only 2 (7,7%) of the 26 patients that presented underlying diseases were colonized by *S. agalactiae*, likewise 19 (6,4%) of the 298 patients without underlying diseases were colonized by this organism. No significant difference was observed according to presence of underlying disease (p < 0.79).

269.

MI8 - IDENTIFICATION OF *Pseudobutyribivrio ruminis* ISOLATED FROM RUMEN OF CREOLE GOATSGrilli D^{1,3}, Cerón M⁴, Schnittger L⁴, Paez S^{1,2}, Egea V^{1,2}, Cobos E¹, Allegretti L^{1,3}, Arenas N³.¹UMaza. ²IADIZA-CCCT-Mendoza. ³U.N.Cuyo. ⁴INTA Castelar.

In the region of Lavalle, Mendoza; goats compose their diet with a high proportion of shrub species, which constitute a major supply of fiber. The high efficiency in the utilization of the fiber may be due, among other factors, to characteristics of ruminal fibrolytic bacteria. Therefore, the study of these bacteria assumes great importance in goat production systems in our country. The aim of this study was to identify and characterize through genetic and biochemical tests, the cellulolytic bacteria isolated from rumen of Creole goats. Sequences of the gene (DNA) encoding subunit 16S ribosomal RNA (16S rDNA) are a powerful tool for taxonomic classification of rumen bacteria and to demonstrate the vast microbial diversity present in the rumen of these animals. We isolated 13 strains of strictly anaerobic bacteria from rumen of Creole goats grazing on rangelands of NE Mendoza. One of them was genetically identified using 16S rDNA sequencing, morphology and physiological characteristics and fermentation end products. The results support the designation of this strain as belonging to the genus *Pseudobutyribivrio ruminis*, which is closely related to *Butyribivrio fibrisolvens*. These biotypes are predominant in rumen of animals adapted to rigorous conditions feeding and diets of poor nutritional quality. It is the first report the isolation and identification of a bacterial strain from rumen contents of Creole goats.

270.

MI9 - GROWTH AND HEMOLYSIN PRODUCTION BY *Listeria monocytogenes* UNDER LOW OXYGEN CONDITIONS

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Listeria monocytogenes is a Gram positive bacterium, causative agent of listeriosis. The organism enters the human body frequently through ingestion of contaminated foods. The aim of this study was to evaluate the growth and listeriolysin O (LLO) production in oxygen limited environments. Microorganism: *L. monocytogenes* CLIP 74902. Culture Medium (CM) (g/l): proteose peptone 30, yeast extract 5, trypticase 5, cysteine chlorhydrate 0.5, pH 7.6, supplemented with glucose (g/l): 1, 2, 5. Cultures of *L. monocytogenes* were performed in microfermenters containing 700 ml CM and incubated at 37°C for 100 h, under static conditions. Redox potential (Eh) was continually monitored. Biomass was estimated by dry weight (DW) determinations at the end of cultures. In the supernatants, remaining glucose was quantified and hemolytic activity (HA) was determined spectrophotometrically at 540 nm using sheep erythrocytes. The results were expressed as percentage of hemolysis. The DW (g/l) values obtained with glucose (g/l): 1, 2, 5; were as follow: 0.28, 0.30, 0.36 and HA (%): 55, 62, 5. Remainder glucose (3.12 g/l) was only detected in the culture with the highest concentration initial (5 g/l). In all cases, the Eh was equal to -175 mv. Results showed that: i) glucose had a repressive effect on LLO synthesis and ii) the ability of *L. monocytogenes* to grow and produce LLO in low oxygen conditions such as vacuum-packaged foods or gastrointestinal tract microenvironment.

271.

MI12 - GROWTH AND PROTEASE PRODUCTION BY NON-O1/NON-O139 *Vibrio cholerae* USING FAST AND SLOW ASSIMILATION CARBON SOURCES

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Non-O1/non-O139 *Vibrio cholerae* strain is the etiologic agent of cholera-like syndrome. Hemagglutinin protease (Hap) is one of the major secreted proteins, which is an important colonization factor of the small intestine. The growth and Hap production were analyzed using fast and slow assimilation carbon sources.

Microorganism: non-O1/non-O139 *V. cholerae*. Culture medium (CM) g/l: proteose peptone 30, yeast extract 5, trypticase 5, pH 7.6. Overnight cultures grown in CM with agitation were diluted 1:1,000 in two 500-ml flasks containing 100 ml of fresh CM supplemented with sugars (g/l): glucose 11 and dextrin 15, respectively. Cultures were incubated at 37°C with shaking (120 rpm), for a total period of 20 h. Growth was monitored by periodic determinations of dry weight (DW) and the specific growth rate (μ) was calculated. From supernatants, obtained at the end of the culture, were estimated glucose remaining and protease activity (PA) using azocasein 0.5 %. DW (g/l), μ (h⁻¹) and PA (U/l) obtained with glucose were: 4.59, 0.82, 380. With dextrin, respective values were as follows: 4.84, 0.74, 516. No residual glucose was detected. Differences observed between both culture of non-O1/non-O139 *V. cholerae*, showed that slowly utilized carbon sources enhance the growth and Hap production, effect has been observed for other bacterial secondary metabolites as toxins and antibiotics.

272.

MI13 - *Lactobacillus* ADHESION TO EPITHELIAL CELLS FROM BOVINE TEAT CANAL

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Bovine mastitis is the principal economic loss in the dairy herd. In the last decades, due to indiscriminate use of antibiotics, new strategies are being developed for the prevention of this disease. The aim of this work was to determine the capacity of lactic acid bacteria (LAB) to co-aggregate bovine mastitis pathogens (BMP) and to adhere to bovine teat canal cells (BTCC). Co-aggregation between LAB (n=6) and the BMP (n=20) was assayed by co-incubation of 10⁹ ufc/ml of LAB and BMP, Gram-stained and microscopic observation. For adhesion assays, 10⁵ cells/ml obtained by scrapping the bovine teat canal wall was incubated with LAB suspensions, Gram stained and optical microscopy observations. Among LAB strains analyzed, *Enterococcus hirae* 7-3, *E. hirae* CRL1834, *Weissella cibaria* CRL1840 and *Pediococcus pentosaceus* CRL1831 showed 100 % co-aggregation, while *P. pentosaceus* CRL1832 and *W. cibaria* CRL1833 showed no co-aggregation to *Staphylococcus chromogenes*. In addition, *W. cibaria* CRL1833 not co-aggregated to *S. capitis* and *Pseudomonas* spp. *E. hirae* 7-3 and *P. pentosaceus* CRL1831 showed 95% of adhesion. and *E. hirae* CRL1834, *P. pentosaceus* CRL1832, *W. cibaria* CRL1833 and *W. cibaria* CRL1840 had 75%-90% of adhesion. These preliminary results are the basis on selecting strains to future design a probiotic to be applied on veterinary to prevent bovine mastitis.

273.

MI14 - *IN VITRO* INHIBITION OF BOVINE MASTITIS PATHOGENS BY LACTIC ACID BACTERIA FROM BOVINE MILK

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The application of probiotics bacteria is an alternative to the use of antibiotics in bovine mastitis. In this work we evaluated the ability of lactic acid bacteria (BAL) isolated from bovine milk to inhibit bovine mastitis pathogens (BMP). Inhibition of bovine teat canal microbiota (TCM) and BAL antibiotic resistance profiles were also evaluated. Inhibition tests were performed by placing a streak line of each LAB on MRS agar plates and antibiotic resistance profiles determined by standardized technique (NCCLS, 2002). All BAL (n=6) inhibited at least one of BMP (n=20). *Enterococcus hirae* 7-3 had the highest percentage of inhibition against BMP (50%) and TCM (73%). *Pediococcus pentosaceus* CRL1831, *P. pentosaceus* CRL1832 and *E. hirae* CRL1834, *Weisellia cibaria* CRL1833 and *W. cibaria* CRL1840 showed a low inhibition against BMP (5-20%), but also showed a high inhibitory capacity against TCM (40-80%). For all BAL, a high inhibition against *Staphylococcus aureus*, the main BMP in Argentina was observed. All BAL were resistant to streptomycin. None of the strains were resistant to erythromycin, penicillin, rifampicin, gentamicin and ampicillin-sulbactam. The high inhibition of BMP observed together with the sensibility observed to antibiotic makes adhesive *E. hirae* 7-3 a potential strain to further studies to be included in a probiotic product for veterinary use.

274.

MI16 - TETRADECYLTRIMETHYLAMMONIUM DEGRADATION BY *P. Putida* AND RELATION TO GLOBAL REGULATORY SYSTEMS

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Pseudomonas putida ATCC 12633 is able to use the detergent tetradecyltrimethylammonium bromide (TTAB) as sole C and N source. TTAB degradation involves C-N bond cleavage by TTAB-monooxygenase activity, leading to products tetradecanal and trimethylamine (TMA). In order to determine two-component system CbrAB role in regulating metabolism of TTAB and TMA in *P. putida* A ATCC 12633, an insertional mutant in the gene homologous to *pp4695* (possible histidine kinase sensor) was obtained and physiologically characterized. In rich medium, mutant strain *P. putida* A ATCC 12633 *hk::Gm* growth was similar to that obtained with wild strain, but in saline supplemented with carbon preferential sources, the mutant strain has, with respect to the parental strain, lag phase and generation time increased and growth was lower. These results suggest that CbrAB system affect the expression of genes belonging to pathways of preferential sources utilization. On the other hand, we observed that mutant strain, was unable to grow with TTAB as carbon, nitrogen or carbon and nitrogen source, but TMA was used as N source. Possibly, inactivation of histidine kinase sensor avoids what this strain is capable of sensing the presence of detergent and, therefore, not induce genes required for use. Failure to detect growth when TMA is used as a C source would CbrAB system involvement in their metabolism.

275.

MI18 - PULSED FIELD GEL ELECTROPHORESIS (PFGE) IN *Yersinia enterocolitica* STRAINS ISOLATED FROM FOODS IN SAN LUIS, ARGENTINA

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Pulsed field gel electrophoresis (PFGE) allows the comparison of genomic restriction profiles of strains belonging to the same bacterial species. In the present work, this technique was used to establish clonal relationships between eight *Y. enterocolitica* B1A strains isolated from different foods in San Luis, Argentina. The preparation of chromosomal DNA and its restriction with *Xba*I, were performed according to the PFGE protocol standardized by PulseNet (USA). Electrophoresis was carried out using a CHEF-DR III system (BioRad) at 6 V/cm at 14°C for 20 h with the following pulse times: initial time 1.8 s and final time 20.0 s. *Salmonella* Braenderup H9812 and *Y. enterocolitica* W1024 B2/O:9 were used as DNA size standard and as reference strain, respectively. The strains were grouped into two clusters A and B, producing five genomic types (GTs). Cluster A included four genomic types (GTs) where all *Y. enterocolitica* B1A strains with different serotypes were included. Cluster B included only the reference strain. Clusters A and B showed 68% similarity. PFGE demonstrated a high power of discrimination between strains of the same bioserotype which could contribute to the knowledge of the epidemiology of this bacterium in our region.

276.

MI19 - EVALUATION OF GENOMIC RESTRICTION PROFILES OF *Salmonella* STRAINS ISOLATED IN SAN LUIS, ARGENTINA

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Pulsed field gel electrophoresis (PFGE) is a valuable technique for comparing, grouping and differentiating DNA restriction patterns among isolates of a serotype specific and thus to establish epidemiological links between strains. The aim of this study was to establish clonal relationships between strains of *Salmonella* strains isolated in San Luis. Thirteen *Salmonella* strains belonging to the following serovars: S. Newport (6) and S. Gaminara (5) isolated from pig tongue and tonsil, respectively, and S. Typhimurium (1) isolated from human feces were tested. PFGE was performed using the restriction enzyme *Xba*I (Fermentas) according to the PulseNet standard protocol (USA). Electrophoresis was carried out using a CHEF-DR III system (BioRad) at 6 V/cm at 14°C for 20 h with the following pulse times: initial time 2.2 s and final time 63.8 s. Restriction pattern analysis and the cluster definition were performed using Statistica 7.0 software. *Salmonella* Braenderup H9812 was used as DNA size standard. Three genomic types (GT), with 70% similarity, based on the serovar of each strain and the common origin of the samples were obtained. These results contribute to a database that could allow the comparison with other electrophoretic patterns of strains of the same species isolated in our region.

277.

MI20 - HIGH SUSCEPTIBILITY OF *Salmonella* STRAINS TO DIFFERENT GROUPS OF ANTIBIOTICS

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Increases in the bacterial resistance against commonly used antibiotics such as: beta-lactam antibiotics (penicillin and cephalosporins), tetracyclines and fluoroquinolones are frequently reported. Our aim was to determine the antimicrobial susceptibility of twelve *Salmonella* strains belonging to the following species: *Salmonella* Newport (7) and *S. Gaminara* (4) both of animal origin (wild boar), and *S. Typhimurium* (1) of human origin, isolated from feces of a symptomatic patient, in San Luis Argentina. The susceptibility was tested against fourteen antimicrobial agents belonging to groups with different mechanisms of action: ampicillin, cefotaxime, ceftriaxone, nalidixic acid, ciprofloxacin, trimethoprim - sulfamethoxazole, fosfomicin, chloramphenicol, gentamicin, colistin, furazolidone, aztreonam, erythromycin, tetraciclina. The disk diffusion method according to Kirby-Bauer was used and bacterial suspensions at concentrations corresponding to 0.5 MacFarland were seeded on Mueller Hinton agar. Diameters of inhibiting zones were read after incubation at 37°C for 24 h. The results of all measurements expressed values above the cutoff point. These results revealed a high susceptibility of local *Salmonella* strains against all antibiotics including the most frequently used in the human and veterinary clinical therapy.

278.

MI21 - BIOFILM FORMING ABILITY OF *Yersinia enterocolitica* STRAINS

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The objectives of the present work were: a) to study the capacity of different *Yersinia enterocolitica* (*Ye*) strains to form biofilms onto polystyrene, and b) to know the media conditions that inhibit the biofilm formation. Nineteen strains of *Ye* were studied with crystal violet technique. The culture medium was trypticase soy broth (TSB) added with 0.25% glucose. Furthermore, two strains were studied under different concentrations of K₂HPO₄ (0; 1.8; 9; 18 or 27 mM), NaCl, glucose or sucrose (0; 0.5; 2; 5 or 10 %). These strains were: *Ye* B1A/O:7,8-8-1,9 (C1, strong biofilm) and *Ye* B2/O:9 (3G, weak biofilm). Fourteen strains were able to form a strong biofilm; four strains formed a moderate biofilm, and one strain formed a weak biofilm. On the other hand, *Ye* 3G was able to form a moderate biofilm with 5% glucose and sucrose, and with 1.8 mM K₂HPO₄. The highest ability to form biofilm of *Ye* C1 strain was observed with null glucose or sucrose, 2% NaCl and 27 mM K₂HPO₄. On contrary, the ability to form biofilm of C1 strain was significantly inhibited with concentrations higher or equal than 0.5% sucrose, 2% glucose or 5% de NaCl. All *Ye* strains were able to form biofilm onto polystyrene on different amount, being *Ye* B1A (C1) the strongest biofilm-forming strain and *Ye* B2/O:9 (3G) the weakest biofilm-forming strain. Culture medium components influence on the biofilm formation. High osmolarity inhibits both bacterial planktonic and sessile growth.

279.

MI22 - ANTIBACTERIAL ACTIVITY OF EXTRACTS OBTAINED FROM *Azorella trifurcata* (Gaertn.) Pers.

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Azorella trifurcata (Gaertn.) Pers is a native plant known by the vernacular names "Yareta". It was collected in Mendoza, Argentina. This species is used in folk medicine as antitussive and expectorant, as well as antiseptic, antiparasitic, antirheumatic and hypoglycaemic. Different extracts were prepared using mixtures of ethyl acetate and *n*-hexane and mixtures of ethyl acetate and methanol on flash chromatography. The aim of this study was to evaluate the antibacterial activity of *A. trifurcata* against methicillin resistant *Staphylococcus aureus* ATCC 43300, *Pseudomonas aeruginosa* ATCC 27853, *Listeria monocytogenes* ATCC 74910, and *E.coli*. The antibacterial activity was assayed *in vitro* using **microwell dilution** assay **method**. Suspensions of 10⁶UFC/ml strains were used. Organic extracts were dissolved in DMSO and tested in a concentration ranging from 8 to 1 mg/ml. TTC was used as visual indicator of bacterial growth. After 24-hour incubation at 37°C, the antibacterial activity of the extracts was defined as absence of red in the wells. Extract *A.trifurcata* 100% *n*-hexane showed activities against *S. aureus*, *Paeruginosa* y *E.coli* at doses of 8 mg/ml and against *L.monocytogenes* at doses of 4 mg/ml. Extract *A. trifurcata* 70% ethyl acetate/*n*-hexane showed inhibitory activity against all strains at a concentration of 8 mg/ml and extract *A. trifurcata* 2% ethyl methanol/ethyl acetate showed inhibitory activity against *S. aureus* and *L. monocytogenes* at doses of 4 mg / ml and against *P. aeruginosa* and *E. coli* 8 mg/ ml. The discovery of plant extracts with antibacterial properties could contribute to the fight against bacterial infections.

280.

MI23 - EVALUATION OF ANTIBACTERIAL ACTIVITY OF *Baccharis sagittalis* (ASTERACEAE)

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The aim of this work was investigate the antibacterial activity of *Baccharis sagittalis* against *Staphylococcus aureus* ATCC 43300, *Listeria monocytogenes* CLIP 74910, *Pseudomonas aeruginosa* ATCC 27853 and *Escherichia coli*.

B. sagittalis "carqueja" is used in Argentine in folk medicine as hepatic, anti-inflammatory and eupeptic. Three crude organic extracts (acetone (A), chloroform (B), and methanol (C)) were tested. The antibacterial activity was assayed by micro-well dilution method in broth supplemented with 0.01% (w/v) of tetrazolium red. Organic extracts were dissolved in DMSO and tested in a concentration ranging from 8 mg /mL to 1mg/mL. After 24h incubation at 37°C the antibacterial activity was defined as absence of red colour. The wells that showed no bacterial growth were confirmed by agar plating and determined the minimum bactericidal concentration (MBC). Against *S. aureus* and *L. monocytogenes*, acetone extract showed significant activities at doses of 4 mg/mL (CIM coincided with the CBM), while against *P. aeruginosa* only chloroform extract resulted bioactive. *E. coli* was resistant to the action of the 3 extracts. On the other hand, methanolic extract exhibited the same antibacterial effect that the control. Future studies will be needed to assess antioxidant activity and selective toxicity of *B. sagittalis*.

281.

MI24 - GENERAL PROTEIN PALMITOYLATION INHIBITOR 2-BROMOPALMITATE ALTERS GLIDING AND INVASION IN *Toxoplasma gondii*Alonso AM¹, Cóceres VM, Angel SO¹, Corvi MM¹.¹Instituto de Investigaciones Biotecnológicas-Instituto Tecnológico de Chascomús (IIB-INTECH). Chascomús. Argentina.

Protein palmitoylation refers to the reversible covalent attachment of palmitic acid to proteins. This post-translational modification has been shown to play a part in diverse processes such as signal transduction, cellular localization and regulation of protein activity. Although many aspects of protein palmitoylation have been identified in mammalian and yeast cells, little is known of this modification in protozoan parasites and more precisely in *Toxoplasma gondii*. The use of a general inhibitor can shed light of the functional significance of this modification in this parasite. Incubation of fresh extracellular *T. gondii* parasites with 2-bromopalmitate altered the parasite's gliding motility and reduced the invasion to the host cells in a dose-dependent manner, with 50% inhibitory concentrations between 50 and 100 mM. At the concentrations tested no effect was observed on the parasite's morphology, basic metabolism or replication. These results suggest that palmitoylation could be modifying proteins that are key players in gliding and invasion in *T. gondii*.

282.

MI26 - FOOD BOTULISM IN MENDOZA, ARGENTINA (1977- 2011)

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Food botulism (FB) is currently the least common form of human botulism. In Argentina, the first outbreak happened in Mendoza in 1922 for consumption of home-canned asparagus. Between 1977 and 1991, there were 10 outbreaks (27 cases). After 18 years without records, two outbreaks were produced, one in 2009 (only 1 case), by ingestion of pickled viscacha (*Lagostimus maximus*), and another in 2011 (2 cases), by pickled eggplant. The two foods were home-canned. In both outbreaks the diagnosis was confirmed by: (1) detection of botulinum toxin (BoT) in food debris, intestinal contents and serum of patients, and (2) by detection of spores of *C. botulinum* (Cb) in all materials except serum. BoT detection and typing were performed by mouse bioassay. In all materials and isolates were identified serotype A. In the remnants of food consumed, the titles of BoT were: 13.632 and 1.810 DL₅₀/ml, in viscacha and eggplant respectively. Neither BoT was detected nor Cb was isolated in the other 7 bottles of viscacha and 5 of eggplant. In the 2009 outbreak, despite the high toxicity of food and the delay in the administration of antitoxin, the case was severe but favorable. In 2011, despite of early antitoxin treatment one of the patients died. The BA is preventable through health education, by means of teaching the community in the implementation of best practices in home canning and heating prior to consumption to destroy the toxin.

283.

MI27 - STUDY OF CRYOPROTECTANTS AND TEMPERATURES IN FREEZE-DRYING OF THE BACTERIUM *Rahnella aquatilis*

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The bacterium *R. aquatilis* is an effective Biological Control Agent against to phytopathogen mould. Commercialization of ACB requires developing a formulation that conserves the viability and a high concentration of eligible cells. The objective of this work was (a) evaluating different temperatures and mixtures of cryoprotectants to conserve the viability of *R. aquatilis* by means of freeze-drying (b) the application of scanning electron microscope (SEM) to detect bacteria on the matrix of bacterial formulations. *R. aquatilis* BNM 0523 isolated and identified in our laboratory. Samples were freezing stored at -20°C or -70°C, prior to freeze-drying, using as cryoprotectants the following mixtures: (1) SMYG (Skim Milk 10%; Yeast Extract 0.5%; Glucose 1%); (2) SMYG + Glycerol 1% and (3) Skim Milk 10% (SM10)+Trehalose 10%. Viability of *R. aquatilis* cells after treatments was expressed as a percentage of surviving cells compared with the initial number of cells (CFU/ml) by duplicate. The freeze-dried formulations were observed by SEM. The viability of *R. aquatilis* quickly freezing (-70°C) before the freeze-drying with SMYG was of 98% ($p < 0.05$); while when they were frozen slowly (-20°C) the viability decreased to 63% ($p < 0.05$). SEM micrographs of SMYG mixture revealed a freeze-dried material with uniform and porous structure that made rehydration easy. In the case of SMYG+Glycerol and SMYG+Trehalose the bacterial cells were detected on the aggregates surface, it was coinciding with smaller viability. The best results for surviving of *R. aquatilis* cells were obtained by using mixture SMYG as the protecting and rehydrating medium, and a quickly freezing (-70°C) before the freeze-drying.

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284.

MI29 - ISOLATION AND FERMENTATIVE STUDIES ON WINE YEAST

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Several yeast species are capable of forming ethanol by fermentation of monosaccharides and oligosaccharides. The quantitative expression of this property is, however, quite variable from less than 1-2% (w/v) to 14% and higher in some strains of the genus *Saccharomyces*. Tolerance to concentrations of ethyl alcohol higher than 12-13% (w/v) is a technological character highly appreciated in the case of yeasts utilized as fermentation starters in the wine industry. The aim of this work was the isolation of yeasts from grape macerate and to compare yours fermentative features with commercial starters. Was make a macerated to variety Syrah and Muscatel grape from San Luis and Mendoza and later was isolated yeasts on YEPD medium, at 2,4,7,9 and 12 days. Identifications were realized with API 20 C BioMeriux system. Subsequently were selected some genus *Saccharomyces* yeasts for to study fermentative kinetic and fermenting power, in comparison with commercial starters. The test was carried out on grape must to 23° Brix, at 28°C in Erlenmeyer flasks plugged with a special glass device (Müller valve), for 16 days. Results showed that two strains of *Saccharomyces cereviceae* had fermentative kinetic and fermenting power similar to commercial strains. Whereas two strains of *Saccharomyces exiguus* had a performance many poor, managing to produce only 5-6 % of alcohol. These preliminary results do that the selected yeasts and that presented good fermentative characteristics are candidate possible to be commercial wine starters.

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285.

MI34 - GENETIC CHARACTERIZATION OF *Helicobacter pylori* CULTURABLE COCCOID FORMS

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Morphological conversion from spiral to coccoid forms has been described for *Helicobacter pylori* under several conditions such as aerobiosis alkaline pH, prolonged incubation and treatment with a proton pump inhibitor or antibiotics. Coccoid form has been considered as viable but non-culturable form of the bacterium. The aim of this study was to establish the culturability of coccoid forms and to characterize DNA and ribosomal RNA (rRNA). *H. pylori* NCTC 11638 reference strain and two clinical isolates were used to obtain coccoid forms under aerobic conditions. These forms were subcultured using Mueller Hinton agar supplemented with 7% horse blood. DNA and RNA from *H. pylori* bacillary and coccoid forms were isolated by rapid boiling and Trizol methods respectively. The results demonstrate that coccoid forms obtained from the three strains can be subcultured *in vitro* under adequate conditions. The coccoid forms showed distinct highly specific fragmentation patterns in both DNA and rRNA molecules. The cleavage pattern presumably reflects a predetermined physiological process in response to environmental conditions. From our data, the non-random fragmentation of DNA and rRNA in *H. pylori* does not induce loss of viability and coccoid forms can maintain transcriptional and translational processes.

286.

MI35 - DIAGNOSIS OF *Helicobacter pylori* IN SALIVA. DETECTION OF CLARITHROMYCIN RESISTANCE

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Helicobacter pylori colonize the gastric mucosa and leads to the development of chronic gastritis, peptic ulcer and gastric cancer. The presence in the oral cavity is considered as a source of infection and re-infection after antimicrobial therapy, as well as intra-family transmission. The aim of the study was to determine the sensitivity of PCR for diagnosis of *H. pylori* from saliva samples and detection of clarithromycin resistance (CLA-R) genotypes. PCR sensitivity was determined using serial dilutions of a saliva sample from *H. pylori* negative patient artificially contaminated with 1.5×10^6 ufc/ml of NCTC 11638 *H. pylori* reference strain. For diagnosis of infection and detection of CLA-R genotypes, 2-3 mL of saliva samples from each of four family members: two parents symptomatic and diagnosed *H. pylori* positive and two children non symptomatic. The specie-specific antigen was used for diagnosis of infection. Genotypes of CLA-R were determined by RFLP-PCR method using *BsaI* and *MboII* enzymes. Limit detection of *H. pylori* from saliva sample was 10^2 ufc/ml. The detection in saliva confirmed the presence of bacteria into oral cavity of parents, and one of the boys. Mother and son showed A2143G genotype, while father was CLA-S. The results showed that the saliva may serve as an effective and valuable noninvasive sample to diagnose of infection and monitor the CLA resistant strains.

287.

MI36 - FERMENTATION KINETIC AND ANTIMICROBIAL ACTIVITY OF *Bacillus* sp SL-6

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Bacillus species produce a large number a bioactives compounds against bacteria, fungi, protozoa and viruses. This report describes the detection of antimicrobial activity (AA) and sporulation stage of *Bacillus* sp SL-6 in batch cultures. It was cultured on Synthetic Mineral Broth with orbital shaking at 200 rpm for 36 h at 30°C. The optical density, pH and glucosa concentration were evaluated at several sampling times. Spores were obtained by thermal treatment and enumerated as CFU/ml. The samples were centrifuged and filtrated to obtain cell-free supernatants (CFS) The AA of CFS was tested against *Staphyococcus aureus* ATCC 29213, *Yersinia enterocolitica*. W1024 and *Candida albicans* ATCC 36801 by the agar well diffusion method and quantified as activity units per millilitre (AU/ml). Activity against *C. albicans* and *S. aureus* showed a similar pattern in exponential phase (1200 and 800 AU/ml, respectively), with scarce stability at stationary phase. The A.A. against *Y. enterocolitica* occurred after reaching the stationary phase (800 AU/ml), it was stable and showed a good correlation with the sporulation stage of the producer bacteria. In conclusion, the AA of *Bacillus* sp. SL-6 metabolites was strongly dependant of growth phases, with only typical behaviour as secondary metabolites against *Y. enterocolitica*.

288.

MI37 - KINETIC STUDY AND PHOTOSYNTHETIC PIGMENTS PRODUCTION OF *Nostoc minutum* IN THE PRESENCE OF AS(V)

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Cyanobacteria are photosynthetic microorganisms able to grow in contaminated heavy metal aquatic environments with potential toxic mobilization through the food chain. Arsenic is widespread in the environment due to both natural sources and anthropogenic activities. The aim of this work was to study the influence of As(V) on growth and photosynthetic pigments content of the filamentous diazotrophic cyanobacteria *N. minutum*. Cultures were grown for 16 days in Watanabe medium added of As(V) 1000 mg/l with continuous illumination of 9,84 Klux at 30°C. Biomass concentration was estimated by OD at 580nm and dry weight measurements. *N. minutum* specific growth rate increased in the presence of As(V) with a final biomass concentration of 1.6 g/l. Photosynthetic pigments pattern was similar with the highest increment for chlorophyll *a* 23,12mg/g vs 8,25 mg/g. Total carotenoids and aliphycocianin showed values of de 0,84 and 60mg/g in the presence of As(V) while control culture was 0,60 and 48,47mg/g. Finally phycocianin showed no variation in both conditions. Cell shape also changed considerably in the presence of As(V) they became thinner with longer filaments and less strenght of the exopolisaccharide sheath. This results showed that *N. minutum* growth and photosynthetic pigments were stimulated in the presence of high As(V) concentrations, characteristic with potential risk in toxic mobilization through the food chain.

289.

MI38 - USE OF A COMPLEX MIXTURE OF CRYOPROTECTANTS ON THE VIABILITY AND BIOCONTROL YEAST *Cryptococcus laurentii*

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The development of formulations that maintain the viability and antagonistic activity of microorganisms that are Biological Control Agents (BCA) is one of the most important steps to implement this method of control of postharvest rots. Freeze drying is a method of choice for long-term preservation of microorganisms. *C. laurentii* (BCA), is an effective against *Botrytis cinerea*, was isolated and identified in our laboratory. The aim of this study was to preserve by lyophilization *C. laurentii*, in a stable and active state, using a complex mixture of cryoprotectants: SMYG (Skim Milk 10%, Yeast Extract 0.5%, Glucose 1%). The yeast was frozen at -70°C overnight and lyophilized for a period of 10 hours. The survival percentage was calculated by plate count before and after treatment in duplicate by serial dilutions on Yeast Glucose Medium (YGM). The antagonistic capacity of freeze-dried *C. laurentii* cells against *B. cinerea* was tested on Red Delicious apples. It was evaluated as Severity Reduction (SR %) and Disease Incidence (DI %). Results showed that the viability of the yeast after 24 hours was 96%, at 90 days 94.9% ($p=0.05$). RS% was 79.3% (24 hours) and 73.4% at 90 days ($p=0.05$). DI% was 51.4% and 69.2% respectively ($p=0.05$). Taking into account the high percentage of viability and functional activity, the complex mixture (SMYG) was good cryoprotectant. This treatment could be useful in order to achieve an adequate commercial formulation of this biocontrol agent.

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290.

MI39 - STUDY OF ATTENUATION DEGREE OF A *Yersinia enterocolitica* YOPH DEFICIENT MUTANT STRAIN

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Yersinia enterocolitica (*Ye*) is an enteropathogenic bacterium which causes gastrointestinal disorders. Pathogenic *Ye* strains carry a plasmid (pYV) that encodes *Yersinia* outer proteins (Yops) which interfere with immune mechanisms in the host. Yop H prevents phagocytosis of *Ye* by macrophages and blocks the lymphocyte activation. The aim of the present work was to investigate the degree of attenuation of *Ye* deficient in Yop H ($\Delta yopH$) after oral infection in C57BL/6 mice. The profile of Yops by SDS-PAGE confirmed the absence of YopH in *Ye* $\Delta yopH$ strain. C57BL/6 mice were orally infected with *Ye* wild-type (WA-314) or $\Delta yopH$ (1.5×10^8 CFU). Percent survival was higher (80%) in *Ye* $\Delta yopH$ infected mice compared with *Ye* WA-314 infected mice (50%). Accordingly, at days 7 after infection, bacterial counts were significantly lower in feces ($p<0.001$), spleen, mesenteric lymph node and Peyer's patches of *Ye* $\Delta yopH$ infected mice ($p<0,0001$). The mutant strain was completely clarified from these organs 21 days after infection. We conclude that *Ye* $\Delta yopH$ was markedly attenuated after oral infection. The results suggest the potential use of *Ye* $\Delta yopH$ as vaccine carrier for oral immunizations

291.

MI40 - LAND USE AND BACTERIA ANTIBIOTIC RESISTANCE IN GROUNDWATER. RÍO CUARTO. CÓRDOBAGambero L¹, Bettera S, Blarasin M, Frigerio C.Fac Cs Exactas Fco Qca y Nautarales. UNRC. ¹CONICET.

Groundwater in Rio Cuarto and the surrounding environment is used in various activities and exposed to different contamination sources (septic tanks and livestock-agricultural activities). The water table varies between 2 and 28 m, influencing the aquifer pollution vulnerability. We examined the bacteriological quality of groundwater samples (n=25), and the relation between bacterial indicators and land uses. Total viable mesophilic aerobic bacteria (RT), total and faecal coliforms (CT and CF), *E. coli* and *Ps. aeruginosa* were determined by standard methodology Antimicrobial susceptibility testing of *E. coli* was performed by a disc diffusion method. The highest value of RT was 6×10^3 cfu/ml and CT was of 1100 MPN/100ml. *Ps. aeruginosa* was found in six wells. The highest count of CF was 75 MPN/100ml whereas *E. coli* was isolated in six wells. The wells located adjacent to potentially contaminant sources showed higher counts of CT and CF, as well as the presence of *E. coli*. The 86% of strain showed resistance to erythromycin and penicillin, 29% to ampicillin. None was to tetracycline, ciprofloxacin, amoxicillin+clavulanic acid, chloramphenicol and cephalothin. The antibiotic resistance pattern showed high percentage of *E. coli* resistant to common antimicrobials in veterinary medicine revealing mainly the impact of livestock in groundwater. The spread of resistant bacteria in aquatic environments could pass through the food chain to humans.

292.

MI48 - CHARACTERIZATION OF CANDIDA SPECIES FOUND IN VAGINAL SWABS OF SYMPTOMATIC WOMEN

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Yeasts of the *Candida*'s genus are a widely distributed group of microorganisms that can cause pathologies. In recent years it has been observed an increase in the percentage of non-albicans' *Candida*, with decreased resistance towards antifungal agents, especially fluconazole. Our objectives were to characterize the different species of *Candida* found in vaginal swabs, and evaluate the antifungal susceptibility of *Candida* non-albicans species found. We studied 46 samples from patients with symptoms of vulvovaginitis, and for its identification Sabouraud dextrose agar (Britain), CHROMagar *Candida*, Nickerson Agar (Merck), Api *Candida* (BioMerieux), urease broth (Britania), Pope agar (Britania) Neo-Sensitab (25ug Fluconazole (Flu), Voriconazole 1UG (Vo), amphotericin 10ug (A), 5ug caspofungin (Cas)) were used. Of 46 samples studied, the *Candida*'s species isolated in percentage were: *albicans* 70%, *glabrata* 15%, *guilliermondii* 11%, *krusei* 4%. Resistance to fluconazole was of a 100% in *C. krusei*. *C. glabrata* and *C. guilliermondii* showed intermediate sensitivity by 14% and 60% of the isolation to fluconazole and caspofungin, respectively. A 100% of the isolation was sensitive to Vo and A. The filamentation was positive in the 100% of the *albicans*' species and 87% *albicans* formed chlamydospores. From our results we conclude that is relevant to the determination of the different *Candida*'s species that cause disorders to avoid treatment failures to deliver the antifungal treatment, due to increased resistance to azoles

293.

MI49 - QUANTIFICATION AND IDENTIFICATION OF SALMONELLA SPP AND SHIGELLA IN WATER SAMPLES IN AREAS ADJACENT TO STREAM PLUGSPastor A¹, Durando P², Varela P³,¹FCEFYN-UNSJ; ²FCA-UNC; ³IBT-FI.-UNSJ.

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In this research we determined the presence of bacteria *Shigella* spp. and *Salmonella* spp. in San Juan River water from 500 m above the mouth of Arroyo Plugs (Site 1) and downstream of the confluence of the river itself (site 2). To do this, at each site were taken every 45 days, two samples of water each time (between December 2008 and November 2009). The quantification and identification of bacterial species were performed according to protocols of the American Public Health Association (APHA, 1998). The results have established the presence of these bacteria in both sites. The population densities of *Shigella* and *Salmonella* measures at Site 1 were significantly lower ($p < 0.0358$ $p < 0.0045$ respectively) measures in the area of the stream with the San Juan River (Site 2). Moreover, the samples analyzed for each gender, 56.52% were identified as *Shigella flexneri* suspected ($n = 13$), 51.02% as *Salmonella enteritidis* ($n = 25$) and 30.61% ($n = 15$) as *Salmonella typhimurium*.

These results signal the marked deterioration of the bacteriological quality of water in the river San Juan.

294.

MI50 - C. botulinum TOXIN PRODUCTION INTERFERENCE BY LACTIC ACID BACTERIA

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It is widely known the beneficial effect of ingestion of probiotics for human health. Its use is widespread, especially in pharmaceutical and food industries (functional foods). Our aim is to identify probiotics that inhibit colonization and/or neurotoxin production of *C. botulinum* (*Cb*), to develop formulations to prevent infant botulism (*IB*) (intestinal toxemia), the most common form of botulism in the world. Strains of lactic acid bacteria (*Lab*) were isolated, one of each of two pharmaceutical formulations (*Lab-Bfl* and *Lab-Tfl*), and one of a dietary supplement (*Lab-Act*). *Lab-Tfl* was identified as *Lactobacillus paracasei* subsp *paracasei* (*Lpp*) (CERELA, Tucumán). The three strains were tested *in vitro* by co-culture with *Cb* strains isolated from *IB* cases of Mendoza (*BL-Mz116* and *BL-Mz125*), for interference testing of development (*DI*) and neurotoxin production (*NPI*). The *DI* was performed on MRS agar by culture of *Cb* "in masse" in a Petri dish, which once solidified was drilled (3 or 4 holes), filled with culture of *Lab* in broth. The results were verified by the presence of inhibition zones around the perforations. For the *NPI* was used co-cultures in liquid medium (MRS broth), varying proportions of *Cb* and *Lab*, and subsequent determination levels of toxin. The two *Cb* were inhibited by the three *Lab* in solid media, and toxin production was interfered by *Lpp* in co-culture with *Cb*.

295.

MI51 - PREVENTION OF HEMOLYTIC UREMIC SYNDROME: TRACING OF CONTAMINATION AND GENETIC DIVERSITY OF Escherichia coli BY STEC (SHIGA TOXIN) IN BOVINE SAMPLES

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Shiga toxin producing *Escherichia coli* (STEC) constitutes an important cause of diarrhea of infectious origin and diarrhea associated with hemolytic uremic syndrome. Cattle are considered the main reservoir and origin of STEC infection to humans.

It has been assumed that *E. coli* in the feces of cattle are spread to meat during slaughter and processing. This study was designed to trace STEC contamination in an abattoir. The overall objective was to evaluate the transmission of STEC from the gut of cattle to the minced meat during slaughter and further processing. One of the specific objectives were to investigate generic *E. coli* as an indicator of general contamination during the process and to study the genetic variability of generic *E. coli* strains obtained to estimate the degree of transmission of bacteria during the process. Screening was performed by PCR in 97 samples from 24 animals. Seven strains of *Escherichia coli* O157: H7 were isolated. ERIC-PCR was performed in 104 *E. coli* strains. The similarities between the DNA patterns of isolates of generic *E. coli* were determined and a dendrogram reflecting the similarities was constructed. The major genetic relation was observed among strains from the same areas of each animal. The highest percentage was 50% similarity between two strains of rectal swabs. The lowest similarity was 6% and was between unrelated strains. These results indicate the persistence of the flora during the slaughtering process and could be used to warn of the possibility of contamination of ground beef.

296.

MI52 - SCREENING OF YEASTS IN VITRO TO BIOCONTROL Aspergillus carbonarius ISOLATED FROM NATURAL OLIVESPesce VM¹, Guerra GB¹, Nally MC¹, Toro ME¹, Castellanos de Figueroa LP, Vázquez F¹.¹IBT. FI. UNSJ. ²PROIMI. Tucumán.

San Juan is one of the main olives producers in Argentina. *Olea europaea* (L.) is affected by *Aspergillus* sp. Dominance and antibiosis of yeasts against *A. carbonarius* (*A.c.*) were evaluated. Microorganisms: 59 grape pathogenic fungi biocontrol yeasts and 2 isolates of *A. carbonarius* from natural olives. Dominance Index: yeasts and fungi were inoculated in dual cultures (Malt Extract Agar: MEA). Interaction was evaluated by numerical scores: 1 (mutual intermingling); 2 (mutual inhibition on contact); 3 (mutual inhibition at distance); 4 (dominance on contact); 5 (dominance at distance). Antibiosis: 10^8 cells.ml⁻¹ yeasts (10µl) were inoculated in equidistant wells of MEA. Fungi mycelium was seeded in the centre of the plate. Inhibitory effect of yeasts on fungi growth was observed (halos). Most yeast showed Index 2: 63% against *A.c.*I and 42% against *A.c.*II. Index 5 was observed in four yeasts against *A.c.*I and only one against *A.c.*II. *Torulasporea delbrueckii* Td129 reduce 64.5% growth of *A.c.*I and *Pichia membranifaciens* Bpm6 reduce 52.7% growth of *A.c.*II. No yeasts showed antibiosis against *A. carbonarius* I and II. Biocontrol yeasts of pathogenic fungi of grapes could be potential antagonists of *A. carbonarius* in olives. Dominance of these yeasts against *A. carbonarius* was determined.

297.

MI56 - *IN VIVO* CURATIVE ACTIVITY OF ENOLOGICAL YEASTS AGAINST *Botrytis cinerea* IN GRAPE

Muñoz MA, Nally MC, Pesce VM, Radicetti DS, Toro ME, Vázquez F. IBT-FI-UNSJ. E-mail: andru_85@hotmail.com

Yeasts have been studied because they possess many features which make them suitable as biocontrol agents in fruits. These microorganisms show a protective effect that diminishes with fruit ripening and senescence (preventive effect), and there are few reports about their curative activity (eradicant effect). The aim of the present work was to evaluate the *in vivo* curative activity of enological yeasts against *B. cinerea* in Superior Grape. The curative activity assay was performed using 40 wine yeasts, which had antagonistic activity *in vitro* and *in vivo* preventive effect assays; against *B. cinerea* B14. Spores suspension (20µL) of *B. cinerea* (10⁴spore/ml) was inoculated into a wound. After 2 and 24h 20µL of yeasts (10⁶cfu/ml) were inoculated. Fruits were stored at 20°C for 7 days. Incidence was determined. After 2h of yeasts inoculation, 17 isolates *B. cinerea* inhibition (8 *S. cerevisiae*, 1 *S. kluyveri*, 1 *S. bayanus*, 4 *T. delbrueckii*, 1 *C. sake*, 1 *D. vanrijiae*, 1 *D. hansenii*). After 24h of yeasts inoculation, 28 isolates showed antagonistic activity (17 *S. cerevisiae*, 5 *T. delbrueckii*, 1 *C. sake*, 1 *S. bayanus*, 1 *S. chevalieri*, 1 *S. kluyveri*, 1 *H. uvarum*, 1 *D. vanrijiae*). These enological yeasts have curative activity against *B. cinerea*. This work represents an initial step for further research in other pathosystems in which *B. cinerea* is a serious pathogen.

298.

MI57 - BIOCONTROL *IN VIVO* OF DIFFERENT STRAINS OF *Botrytis cinerea* USING ENOLOGICAL YEASTS IN GRAPE

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Some yeasts have been reported and patented as postharvest biocontrol agents. Direct screening on fruit is the most efficient methodology to obtain antagonistic microorganisms. The aim of the present work was to evaluate the antagonistic capacity *in vivo* of enological yeasts against 4 strains of *B. cinerea* (B11, B14, B15, B25). These yeasts were selected as antagonistic in previous *in vitro* and *in vivo* tests, 67 isolates against B11, 82 against B14, 125 against B15 and 151 against B24. This *in vivo* antagonism test was: 20µL of a yeast suspension (10⁶cfu/mL) was inoculated in wounds. A suspension (20µL) of a *B. cinerea* spores (10⁴esp/mL) was inoculated after 2h. Fruit were stored at 20°C for 7 days and the incidence was determined. Results showed that 7% of the yeasts tested against B11 inhibited their development, 23% inhibited B14, 32% inhibited B15 and 39% inhibited B24. These antagonistic yeasts belong to different species: 55 *S. cerevisiae*, 2 *S. chevalieri*, 2 *S. kluyveri*, 2 *S. bayanus*, 19 *T. delbrueckii*, 4 *C. sake*, 1 *C. milleri*, 3 *C. parapsilosis*, 2 *C. famata*, 1 *C. catenulata*, 3 *D. hansenii*, 2 *D. vanrijiae*, 1 *K. marxianus*, 1 *H. uvarum*, 1 *C. albidus*, 1 *Z. bailli*, 1 *S. roseus* and 1 *P. guilliermondii*. *S. cerevisiae* BSc243 inhibited 3 strains of *B. cinerea* (B11, B14, B15). Enological yeasts could be used effectively, at a feasible concentration for commercial use, against different strains of *B. cinerea* under shelf-life conditions.

299.

MI58 - EPIDEMIOLOGICAL CHARACTERIZATION OF HUMAN METAPNEUMOVIRUS, RESPIRATORY INFECTIOUS AGENT IN CHILD POPULATION OF CÓRDOBA CITY

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In 2001 van den Hoogen discovered Human Metapneumovirus (hMPV, RNA virus, single negative chain) in Hollanda. Located in Paramixoviridae Family, Pneumovirinae Subfamily, and Metapneumovirus Genus, it produces respiratory infections in all ages, specifically children. The purposes of this study are to develop, standardize and implement diagnostic methods for the study of this viral strain circulating and establish the prevalence and local profile of hMPV in children of the Province of Cordoba. We took samples of nasopharyngeal swabs and aspirates of children 0-5 years of age hospitalized in Hospital Santísima Trinidad. The viral antigen are detected by direct immunofluorescence. The result consistent with literature consulted regarding the prevalence of respiratory viruses in children, occupied first by Respiratory Syncytial Virus. Seasonality, in the samples analyzed until July 15 a case was found positive for hMPV in July, in child of one year old.

300.

MI59 - VEGETATION DISTRIBUTION INFLUENCE IN BACTERIA DIVERSITY OF MONTE DESERT, SAN JUAN

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Microbial diversity encompasses genetic variability within taxons (species), their relative abundance and functional groups of different communities. Bacteria and filamentous fungi are the most important microorganism groups related to energy flow and transfer of nutrients in the soil ecosystem. The aim of this work was to determine bacterial diversity associated to soils from patches of *Bulnesia retama*, *Larrea divaricata* and interspaces in Médanos Grandes San Juan, Argentina. Two samples in one year were taken at random from patches and interspaces (microsites, 2007 and 2008). Molecular identification was carried out using PCR 16S rDNA amplification. In all microsites, 64 isolations were done using colony topography. Five genera were molecularly identified: *Arthrobacter sp.*; *Bacillus sp.*, *Microbacterium sp.*, *Providencia sp.*, y *Stenotrophomonas sp.* *B. subtilis* was widely distributed (46.9%) in all microsites; *B. cereus* 7.8%. The other species were *B. licheniformis* (15.6%); *B. megaterium* (10.9%); *Providencia sp.* (10.9%); *Bacillus sp.* (6.25%); *Microbacterium sp.* (3.1%); and *Stenotrophomonas sp.* (1.6%). *Bacillus* was the most conspicuous genus and was associated to vegetation patches.

301.

MI60 - MICROORGANISM ABUNDANCE IN SOILS OF INTERDUNES ENVIRONMENT, MEDANOS GRANDES, SAN JUAN

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In arid environments, the lack of water limited biological activity. This habitat presents patches of vegetation and interspaces. In the Médanos Grandes, interdunes environment provides more vegetation cover and consequently greater water retention. The abundance of microorganisms in soil varies spatially as well as temporarily. The aim of this work was "to determine the abundance of bacteria, yeasts and filamentous fungi from *Bulnesia retama* and *Larrea divaricata* patches and interspaces, from interdunes of Monte Central Desert. Samples were taken at random from soil of patches and interspaces in April (high precipitations) and August (low precipitations) of 2009. Microorganism abundance was determined by plating count method (CFU/g soil⁻¹). Results showed that in *L. divaricata* and *B. retama* soil patches, bacteria abundance was significantly different respect yeasts and filamentous fungi in April ($p=0.0136$). Whereas filamentous fungi abundance was significantly greater in soils of both patches in August ($p=0.035$). Our data indicate that bacteria and filamentous fungi abundance showed a heterogeneous spatial pattern in associated soils of patches of *L. divaricata* and *B. retama* from the Monte Central Desert.

302.

MI62 - SPACIAL VARIATION OF SOIL INDICATORS QUALITY FROM NATIVE MONTE AND A CULTIVATED AREA

Medina E¹, Vega Avila AD¹, Loto F², Toro ME¹, Paroldi HE¹, Baigori M¹, Babelis G³, Vázquez F¹.

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The enzymatic activities of microorganisms and chemical parameters can be used as indicators of quality of soils. The aim of this work was to compare chemical parameters and enzymatic activities of soil between native Monte and a *Vitis vinifera* L cultivated area. Samples were taken from soil of patches and interspaces in the monte; and rows and interfiles of vine soils, in April of 2010. Conductivity, pH, moisture, nitrogen, phosphorus, potassium, organic carbon and organic matter were measured. Cellulases, amylases and xylanases were quantified. ANOVA and principal components analysis (PCA) were used for results evaluation. This analysis showed that component 1 formed groups in rows, interfiles, and patches of *B. retama*. The influenced parameters were conductivity, cellulases, xylanases, amylases, nitrogen, organic matter, organic carbon, moisture and pH. Axis 2 formed groups at patches of *L. divaricata* and interspaces. Variables influencing groups were potassium and phosphorus. Results showed a framed spatial heterogeneity on microsites formed by patches of vegetation and vines.

303.

MI64 - PREVALENCE OF *Candida* spp IN PATIENTS WITH DENTURE STOMATITIS

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Denture stomatitis is an inflammatory process of the oral mucosa. *Mucous Atrophic* (with erythematous lesions) and *hyperplasic lesions* are the main forms of denture stomatitis. It is more frequently found in women, essentially in the upper maxillary palate surface in contact with a dental prosthesis. There are many factors involved in the etiology of this lesion and it has been associated with the presence of *Candida* spp. and the dental prosthesis. The objective of this study was to determine the prevalence of *Candida* spp in the mucosa of the oral cavity of denture wearers. The study population consisted of patients with denture stomatitis treated at the Prosthodontic Clinics, School of Dentistry, Universidad Nacional de Cuyo, Mendoza, Argentina. We performed sampling by swabbing the oral mucosa inflamed, which were planted on Sabouraud Dextrose Agar medium and incubated at 37°C for 48-72 hours. The identification of fungal species was performed using *Candida* CHROM agar and germ tube. Of the total samples tested gave positive cultures to *C. albicans*: 37.5% *C. glabrata*: 37.5%; *C. parapsilosis*, 12.5%; *C. lipolytica*: 12.5%.

304.

MI66 - EFFECT OF SANITIZER ON GROWTH AND BIOFILM FORMATION OF *Clostridium perfringens*

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Clostridium perfringens is an anaerobic gram-positive pathogen, causative agent of numerous diseases Recently it has been informed to be capable of forming biofilm, increasing its resistance to chemical agents. The aim of this study was to determine the inhibitory effect of the sanitizer sodium tripolyphosphate (TSPP) in biofilm formation and bacterial growth of *C. perfringens*. An enterotoxigenic strain of *C. perfringens*, isolated from food in San Luis, was used. TSPP was added to a final concentration of 0.8% in culture medium. The concentration of planktonic cells was determined by optical density measurements, while the viability of planktonic and biofilm cells (sessile) was established by plate count and the most probable number (MPN) at 24, 48 and 72h of treatment. The biofilm obtained was observed by optical microscopy (OM) and scanning electron microscopy (SEM). After 48 h of treatment with TSPP 2- \log_{10} and 1- \log_{10} units reduction in viability was observed for planktonic and biofilm cells respectively After 72 h a total loss of viability was observed in both cells. The OM and SEM revealed marked morphological changes of the capsule of cells treated with TSPP. The results indicate the need to set sanitizers inhibitory concentrations taking into account the stage of biofilm of *C. perfringens*.

305.

MI68 - MOLECULAR SCREENING OF PATHOGENIC *E. coli* AND ROTAVIRUS STRAINS IN DAIRY NEONATAL CALVES

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The objective of this study was to carry out a molecular screening on pathogenic bovine strains of *E. coli* and Rotavirus. Individual rectal swabs (n=620) were collected from dairy calves and cultured on Mc Conkey agar. Lac (+) colonies compatible with *E. coli* were biochemically characterized and DNA was extracted by heating. Intestinal virulence factors (LT, STb, STa, F17, eae, F5, F41 and VTg) were identified by PCR. Fecal samples (n=96) were collected, and extracted RNA was analyzed by SDS-PAGE with silver stain. The isolates were compatible with: *E. coli* (58,55%), *Proteus spp.* (32,90%), no growth (4,19%), *Enterobacter spp.* (3,23%) and *Citrobacter spp.* (1,13%). From *E. coli* isolates characterized by PCR (n=156), a 69,87% were non-pathogenic *E. coli* and 30,13% were pathogenic strains compatible with *ETEC* (22,44%), *VTEC* (4,48%) and *EPEC* (3,21%). In fresh fecal samples, 2.1% of Rotavirus and 7.3% of Picobirnavirus were found. Non-pathogenic *E. coli* was the most prevalent microorganism in rectal swabs. However, there was a high prevalence of *ETEC*. Picobirnavirus cannot be considered a causal pathogen of diarrhea because the model of viral infection is unknown. A low prevalence of Rotavirus in fecal samples was found.

306.

MI69 - FIRST DETECTION OF HUMAN METAPNEUMOVIRUS-(MPVh) IN CÓRDOBA IN 2011

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The Respiratory Virus Laboratory of the Institute of Virology UNC aims to investigate in 2011 the circulation of MPVh in populations of Córdoba. In 2001, Van den Hoogen discovered MPVh in Holland. Located in the Family Paramyxoviridae, subfamily Pneumovirinae and Genus Metapneumovirus. In Argentina in the study by Maffey *et al.* (2008) a total of 119 patients, 102 cases were positive for VRS and 10 MPVh. The detection of the agent is done by direct methods such as direct DFA technique to be very sensitive, as it provides the sample of a few infected cells giving a typical picture, which allows a rapid diagnosis with certainty. The samples are HNF obtained from infant hospitalized for respiratory diseases in the Children's Hospital. The result was the detection of the first clinical case of MPVh in an one month old infant, showed bronchiolitis. When doing research also faces other respiratory viruses resulting negative for all. In conclusion, although infectious respiratory diseases are multi-causal, since the focus of our research virological where respiratory viral agents face 8, the positive outcome is attributed to MPVh as a causative agent of this infection circulating in Córdoba.

307.

MI70 - IN *Pseudomonas putida* A ATCC 12633 PHOSPHATIDYLCHOLINE ACT AS A TEMPORARY RESERVOIR OF Al^{3+}

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In this work we have cloned and sequenced a gene in *P. putida* A ATCC 12633 that encodes a phosphatidylcholine synthase (PCS) and characterized a *pcs*-deficient mutant. Sequence analysis of *P. putida* KT 2440 revealed the ORF PP0731, which has 26 % sequence identity to a PCS protein in *Sinorhizobium meliloti*. The *orf pp0731* was cloned and expressed in *Escherichia coli* BL21, a phosphatidylcholine-deficient organism. The *P. putida* A ATCC 12633 *pcs* homologue was disrupted to generate a corresponding mutant. In the *pcs*-deficient mutant, detectable levels of phosphatidylcholine were not found, and the mutant was much more sensitive than the wild-type strain when challenged with Al^{3+} . Only 50% of the mutant cells managed to grow in the presence of the ion (the amount of viable cells decreased from 10^{12} ufc ml^{-1} to 10^6 ufc ml^{-1} 15 min after Al^{3+} addition). Also, Al^{3+} in the membrane of wild-type and mutant strains was visualized using the fluorochrome 2',3',4',5,7-pentahydroxyflavone (morin reagent). In the mutant strain, the distribution of green fluorescence was quite similar to that of wild-type cells, but they had a lower level of fluorescent intensity in the cell membrane, consistent with a reduced presence of Al^{3+} . These results support that phosphatidylcholine is involved in the response of *P. putida* to Al^{3+} and acts as a temporary reservoir of available ions.

308.

MI71 - BIOCHEMICAL CHARACTERIZATION OF *E. coli* β -GALACTOSIDASE FUSED TO A HISTIDINE HEXAPEPTIDE

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The aim of this study was to compare the activity of β -Gal_{wt} with a recombinant beta-Galactosidase (β -Gal_{His6}) against lactose. β -Gal_{His6} was overexpressed in *E. coli* and contained 6 histidine residues fused to the carboxyl terminal, which facilitated its purification by metal-ion affinity chromatography, (IMAC). The hydrolysis of lactose was quantified spectrophotometrically at varying conditions of temperature, pH, concentration of Mg^{2+} (activator) and lactose. Both enzymes showed similar affinity for Mg^{2+} ($K_d \cong 2 \mu M$) (activator) as for lactose ($K_d \cong 4.2 mM$). The recombinant enzyme showed a slight resistance to inactivation by temperature and pH. The specific activity of β -Gal_{His6} was lower with respect to β -Gal_{wt}, suggesting that the presence of histidine could intervene in the reaction mechanism. Possible conformational changes can not be discarded.

309.

M172 - RESPONSE TO BROMIDE-TETRADECYL-TRIMETHYLAMMONIUM IN A *P. putida* CARDIOLIPIN SYNTHASE DEFICIENT*López GA, Heredia R, Boeris PS, Lucchesi GI.**Dpto. Biología Molecular. FCEFQyN. UNRC. 5800-Río Cuarto. Córdoba. Argentina. E-mail: glopez@exa.unrc.edu.ar*

Pseudomonas putida A ATCC 12633 responds to tetradecyl-trimethylammonium (TTAB) through quantitative changes in phospholipids with specific variations in the content of phosphatidic acid, phosphatidylglycerol and cardiolipin. Sequence analysis of *P. putida* KT 2440 revealed a gene *pp5364* as cardiolipin synthase (*cls*). The *P. putida* A ATCC 12633 *cls* homologue was disrupted to generate the corresponding mutant, *P. putida* A ATCC 12633 Δ *cls*. In the mutant strain, the cardiolipin levels and distribution into the membranes, visualized using the fluorescent dye 10-*N*-Nonyl acridine orange, was similar to detected in the wild-type strain, indicating that cardiolipin synthesis occurs by other gene in this organism. The strain was grown on a basal salt liquid medium with glucose and NH_4Cl and exposed to 50 mg l^{-1} of TTAB. The cardiolipin level was similar to the one observed in TTAB absence but the presence of TTAB resulted in an increase in phosphatidylglycerol and phosphatidic acid levels (2 and 10 fold, respectively) with respect to the levels in cells grown without the surfactant, indicating that phosphatidic acid is the primary membrane-associated factor for the response to TTAB.

310.

NQ1 - SYNTHETIC STEROIDS WITH OXYGEN BRIDGES'S ACTIVITY ON GABA_A RECEPTOR*Rey M¹, Veleiro AS², Ghini A², Burton G², Coirini H^{1,3}.**Lab. Neurobiología IBYME-CONICET; ²Dept Química. Orgánica. UMYNFOR-FCEN-UBA; ³Dept. Bioquímica Humana-FMED-UBA. E-mail: rey@dna.uba.ar*

Previously we have described the binding behavior of synthetic esterooids (SE) structural analogues to Allopreganonole (Allo) and Pregnanolone (Pregna), using specific agonists of GABA_A receptor. In this work, we describe the effects of these steroids on their specific binding site, capable of modulate the channel opening. We employ two Allo's analogues, SB11-1 and Ns4, and two Pregna's analogues, Ns1 and Ns2. These ES have an oxygen bridge that limits the flexibility of the structure and protect them from a possible enzymatic degradation, resulting in greater stability for use *in vivo*. We evaluate changes in the binding of ³⁵S-TBPS in synaptosomes obtained from rat cortex and cerebellum. Incubations were performed at 25°C for 90 min in the presence of GABA (500 μM) using a 5-1000 nM range for each steroid. Nonspecific binding was determined using picrotoxin (1 mM). On the one hand, Allo; SB11-1; Ns4 and Pregna; Ns1; Ns2 produced an inhibition according to their structural feature (IC_{50} = 86,29; 89,12; 92,89 / 133,65; 141,25 y 489,7 nM respectively), although the Ns2 was the least effective. While the values are similar to their analogs, previous results indicate that the SE stimulate the binding of ligands muscimol (except SB11-1) or flunitrazepam (except Ns2). Referring to the latter two ligands, Ns1 has a behavior similar to Alo. Future trials are necessary for validation and consideration of any of these SE as therapeutic tools. Based on all observed, arguably the NS1, despite its structural analogy Pregna type, seems to be most similar in its action to Allo.

PICT-0727/06,- UBA-M012, PIP860.

311.

NQ3 - POTENTIAL IMPLICATIONS OF Ki-67 EXPRESSION IN SURGICALLY RESECTED PITUITARY ADENOMAS*Serrano L¹, Sereno C², Condomi Alcorta S², Katz D², Selvlever G², Gargiulo PA¹, Cervio A².**¹Laboratory of Neurosciences and Experimental Psychology, FCM, UNCuyo, Mendoza, Argentina. ²Neurosurgery Department, FLENI, Buenos Aires, Argentina.*

Pituitary adenomas constitute the most frequent sellar neoplasms and account for about 15% of intracranial tumors. In spite of being benign, these tumors can grow sufficiently to compromise main encephalic and vascular structures within the skull base. In this work we characterized Ki67 activity in resected adenomas with respect to clinical and histopathological features. Though the quantification of this proliferation marker we aimed to assess the existence of factors which could be associated with a higher or lower proliferative rate. After revising a number of 79 cases we observed that the overall mean expression of Ki-67 was $2.3 \pm 0.22\%$. Our analysis did not demonstrate any difference in Ki-67 activity between sex, age or histopathological subtype. Nevertheless, there was a significantly higher Ki-67 expression in tumors which recurred $F(1,77)=7,483; p<0,01$. Additionally, the likelihood of presenting an increased number of recurrences correlated also with higher Ki-67 initial levels $F(3,75)=3,04; p<0,05$. These preliminary data suggest that Ki-67 activity could be useful to predict tumor recurrence, having this fact important implications for patient follow-up.

312.

NQ5 - SYNTHETIC STEROIDS ACTION ON 3 β HSD ENZYME ACTIVITY IN HIPPOCAMPUS AND CEREBELLUM OF RAT*Rey M¹, Veleiro AS², Ghini A², Burton G², Coirini H^{1,3}.**Lab. Neurobiología IBYME-CONICET; ²Dept Química. Orgánica. UMYNFOR-FCEN-UBA; ³Dept. Bioquímica Humana-FMED-UBA. E-mail: rey@dna.uba.ar*

The 3 beta-hydroxysteroiddehydrogenase (3 β HSD) is present in different tissues, it catalyzes Pregnenolone(P5) into Progesterone(P4). Allopreganolone (Allo) and Pregnanolone (Pregna) derivate from P4 and they are able to modify this enzyme activity. Previously we have described the action of several synthetyc with structural similarities to Allo and Pregna on various biological systems in order to consider them as potential therapeutic drugs. In this study we evaluated the effect of two such drugs (SB11-1 and NS1) on the activity of the 3 β -HSD on nervous system tissues. Homogenates of hippocampus (HC) or cerebellum (Cb) from rat, were incubated at 37°C by 5 min with Glicine-BSA-NAD⁺ Buffer, then P5 was added, the different concentrations of steroids in study (10 μM , 50 μM y 100 μM) were incorporated, and measured the absorbance at 340nm was done by 5 min. SB11-1 produced a greater decrease in enzyme activity at lower concentration than its natural analogue (Allo), while Ns1 had a significant effect only at higher concentrations than its analogue Pregna. This result would indicate that Ns1 could be used systemically at low doses without affecting the enzyme activity related to the synthesis of natural steroid precursor, that acts on the same receptor.

PICT-0727/06, UBA-M012, PIP860.

313.

NQ10 - KETAMINE INDUCES CHANGES IN THE EXPRESSION OF COAT PROTEINS (AP-2) IN NUCLEUS ACCUMBENS OF RATSRuiz AM¹, Carvelli L¹, Capella P¹, Sartor T¹, Gil Lorenzo AF², Domínguez S³, Sosa MA¹.¹IHEM. ²IMBECU. CONICET. FCM. UNCuyo. ³FQBF UNSL.

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Clathrin-mediated endocytosis is important in maintenance of membrane homeostasis at nerve terminals. Conditions of nerve injury, accompanied of changes in levels of dopamine and glutamate, affect the expression and localization of these proteins. Ketamine in the CNS could involve blocking or changing levels of surface receptors to NMDA. In turn, this could be associated with alterations in the endocytic machinery at nerve terminals. Explore possible changes in the expression of the AP-2 coat protein as an index of changes in the endocytic activity in different areas of rat brain treated with ketamine. Adult male rats were treated with effective subanesthetic dose of ketamine (10 mg/kg) or saline, and sacrificed at 15 min, or 24 hours after. Nucleus Accumbens and Prefrontal Cortex right and left (Accd, Acci, Cxd, Cxi) homogenates were analyzed by Western blot. The expression of AP-2 in Accd treated with ketamine significantly increased over control at 15 min and 24 h 100 ± 13.96 vs $178.12 \pm 15.84^*$ and 100 ± 15.30 vs $191.16 \pm 11.30^*$ respectively. Acci of treated rats shows a significant increase 100 ± 14.00 vs $179.15 \pm 21.54^*$ only after 24 hours. In contrast, in the Cxd and Cxi were not observed significant changes in AP-2 expression. The selective increase in the AP-2 expression in Nucleus Accumbens treated with ketamine suggests that the blocking effect of the drug on NMDA receptors alters the endocytic pathway via AP-2, an effect not observed at the level of the prefrontal cortex.

314.

NQ12 - GENETIC MANIPULATION IN THE RAT PINEAL GLAND: IN UTERO INTRAVENTRICULAR INJECTION AND ELECTROPORATION

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In the post-genomic era, identifying gene products and revealing their roles represent an actual challenge. The pineal gland, through its hormone melatonin, is a key effector and regulator of the circadian timing system. The differentiation factor NeuroD1 is thought to be a pineal phenotype determinant. Two NeuroD1 KO mice have been developed. Although new avenues have emerged, these models have disadvantages. Whereas the total KO mice die shortly after birth, the conditional KO mice lack NeuroD1 in both retina and pineal gland. Our group initiated controlled genetic manipulation in the rat pineal gland by optimizing the *in utero* intraventricular injection and electroporation. The use of vectors carrying reporter genes driven by promiscuous promoters has revealed that the number of positive cells was highly dependent on the pineal mitotic activity and time of injection. Highly skilled operators were needed to increase embryo survival rate. It is expected that pineal-selective gene targeting will help us to clarify unknown aspects of the pineal biology and its role in the general physiology. This procedure may also represent a cost-effective approach compared to the maintenance of transgenic mouse lines.

315.

NQ13 - PRECONDITIONING PROMOTES GABA- AND GLUTAMATERGIC NEURONS IN NEONATAL HYPOXIC-ISCHEMIC BRAIN INJURY

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Hypoxic preconditioning (HC) protects from ischemic brain lesions *via* neurogenesis. GABAergic neurons are recognized by GAD1/GAD67. The differentiation factor NeuroD1/BETA2 acts as a hinge element that favors differentiation of glutamatergic over GABAergic neurons. HC-induced neurogenesis may be mediated by both molecular determinants. We analyzed three groups: control (C: sham-operated non-asphyxiated P8 pups), lesioned (L: P8 pups with permanent ligation of the right carotid artery followed by hypoxia), and preconditioned and lesioned (PCL: P7 pups in auto-hypoxic environment before injury). Samples were collected at P15. The hippocampal and cerebellar expression of both markers was evaluated by IHQ and WB. The PCL group showed an increase in GABAergic neurons near the granular layer of the dentate gyrus and in the cerebellar Purkinje cell layer. In the L group, NeuroD1-positive cells decreased in the hippocampal dentate gyrus, cerebellar external germinal and internal granular layers. Migrating immunoreactive cells decreased in the same brain areas. WB confirmed these results. These observations indicate that both cellular phenotypes are induced by HC.

316.

NQ14 - ANGIOGENESIS AND GLIOSIS IN ROP. HYPOTHERMIA AS A THERAPEUTIC TOOLRey-Funes M¹, Dorfman VB², Ibarra ME¹, Peña E¹, Loidl CF¹.

Retinopathy of Prematurity (ROP) is the main cause of blindness in children. We have described that animals exposed to perinatal asphyxia develop retinal lesions consistent with ROP. Our aim was to study the impact of hypoxia-ischemia on retinal neovascularization and astroglial response in animals subjected to an experimental model of ROP, and to evaluate hypothermia as a possible protective treatment. We used retinas of male rats (n=5/group), of 7, 15, 21 and 30 days old (d) exposed to perinatal asphyxia (20 min, 37°C, PA). We evaluated neovascularization studying the expression of adrenomedullin (AM) by immunohistochemistry (IHC) and Western Blot (WB) and the number of vessels by tomato lectin; astroglial response was studied by GFAP expression by IHC and WB. Hypothermia was tested in animals subjected to asphyxia at 15°C (20 min, HYP). Born to term animals were used as controls (CTL). AM was immunolocalized in the internal processes of Müller cells and in the inner retina. Increased AM expression was observed by WB from 15 d ahead in CTL. AP showed significant AM expression increment from 7 d ahead respect to CTL ($83 \pm 3\%$ 7d, $38 \pm 6\%$ 15d, $33 \pm 4\%$ 21d, $35 \pm 6\%$ 30d, $p < 0.05$). HYP group showed similar AM expression pattern than CTL. Immunoreactivity of GFAP was positive in the inner limitant and perivascular glia of 21 d PA and extending into the inner processes of Müller cells at 60 d. This expression was not evident in CTL or HYP groups. A significant increased expression of GFAP was observed by WB in PA vs CTL from day 7 ahead, with a peak at 15 d ($235 \pm 8\%$, $p < 0.05$). The number of the inner retinal vessels showed a significant increase in PA vs CTL at every age (PA: 10.5 ± 1.3 , CTL: 4.6 ± 1.1 vessels/field, $p < 0.01$), while HYP was similar than CTL. In conclusion, AP results in astroglial and angiogenesis in the inner retina and hypothermia showed to be protective to avoid the damage.

317.

NQ15 - SEX DIFFERENCES IN THE EXPRESSION OF LIVER X RECEPTORS IN THE BRAIN

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Glucose and lipid metabolism alterations are common to diabetes and Alzheimer's disease. The oxysteroids play an important regulatory effect. Here we evaluate the presence of specific receptors in the hypothalamus (HT) and cerebellum (Cb) from male and females 9 months old rats, born of diabetic (OD) and controls (OC) mothers. We used a gestational diabetes model to study the expression of these proteins by western blot. Animals were subjected to a glucose load before sacrifice. OD male progeny, showed a tendency to be intolerant to glucose. In the case of OD female, the average response was similar to OC, only one animal showed a clear delay in the response. LXR- α expression showed no differences between OD and OC either male or female in Cb, and it could not be detected in the HT. LXR- β showed a significant increase in Cb in both OD sexes, whereas in HT there was an increase in OD females, and a decrease in OD males. The results indicate that gestational diabetes affects the offspring's neural circuitry, differently in males than in females. In 9 months old male rat, causes an altered expression of LXR- β in the HT, which could be related to the ability to regulate properly, a glucose load.

06CM09-UCCuyo, PIP-860-CONICET, UBACYT-M012.
 in the OD. PIP-860-CONICET, 06CM09-UCCuyo.

318.

NQ16 - ONTOGENY OF THE EXPRESSION OF LIVER X RECEPTORS IN RAT BRAIN

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Gestational diabetes (GD) been related to a diabetogenic effect in the progeny. Liver X receptors (LXR) are potential drug targets for the regulation, treatment or prevention of diabetes. Here we evaluate early and late changes of LXR in the hippocampus (HC) and hypothalamus (HT) of the male offspring of diabetic mothers (OD). Streptozotocin-induced GD model was used to assess LXR- α and LXR- β expression by western blot. The tissues were obtained from OD and controls (OC) animals at 18 days of gestation (E18), postnatal day 1 (P1), 10 and 35 days, and at 7 and 11 months of age. In the HC: LXR- α showed a significant increase in OD animals vs. OC ($p=0.03$) as well as differences among ages ($p < 0.001$). OC animals showed high levels at P10 while in the OD were found at E18-P1-P10. LXR- β was increased by GD. The OC showed a peak at P10, while high OD values were found in 35 days old tissue. In the HT: LXR- α showed no effect neither by DG not by age, with a maximum at P10. OC showed a peak at 35 days of age while for OD it was observed at P1. A significant effect on LXR- β was observed by GD ($p < 0.05$) but not by age. This indicates that altered fetal milieu cause regional changes of LXR expression and may be the cause for impairment in the regulation of lipid and glucose metabolism in the OD.

PIP-860-CONICET, 06CM09-UCCuyo.

319.

NQ17 - KETAMINE INDUCES NEUROLOGICAL DEFICITS ON WORKING MEMORY

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Ketamine constitutes an NMDA-antagonist with an important psychotogenic potential and its administration in healthy subjects can lead to similar symptoms to those observed in schizophrenic patients. Because of this fact, it is nowadays suggested that NMDA dysfunction within the central nervous system could be involved in schizophrenia pathophysiology and specially linked with its negative and cognitive symptoms. In this context, we assessed the working memory effects produced by ketamine in animals, taking into account that this function is commonly affected in schizophrenic patients. To this aim we used an experimental cohort of male rats which were evaluated on a hole board task after a treatment with parenteral administration of saline (controls, $n=20$), ketamine 5 mg/Kg ($n=20$), ketamine 10 mg/Kg ($n=20$), ketamine 15 mg/Kg ($n=24$) and ketamine 20 mg/Kg ($n=19$). The results evidenced a working memory disruption in groups treated with high dosis of ketamine vs controls ($p < 0.05$) and we conclude that NMDA antagonism could produce cognitive and executive deficits similar to this main clinical feature shown by schizophrenic patients.

320.

NQ18 - KETAMINE DISRUPTS WORKING MEMORY IN A HOLE BOARD TASK

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Ketamine, a N-methyl-D-Aspartic-acid (NMDA) receptor antagonist, is an anesthetic that has psychotogenic effects in special conditions. Since working memory deficits have been described in schizophrenia, we studied here the effects of Ketamine on this function, using a hole board test to search for a disruption in this task. Male rats derived from a Holtzman colony were used weighing 240-270 g, divided in 5 groups (saline control, Ketamine 5, 10, 15 and 20 mg/kg, $n=19-24$). Locomotor activity (ambulatory, non ambulatory and vertical movements) and number of entrances to the holes were measured. A significant modification on locomotion and holes exploration parameters was observed in the test. We conclude that Ketamine disrupts working memory within present conditions.

321.
NQ19 - BRAIN RENIN-ANGIOTENSIN SYSTEM (RAS) IS INVOLVED IN THE LONG LASTING NEURONAL ACTIVATION INDUCED BY AMPHETAMINE

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The enhanced response to psychostimulants relies on time-dependent neuroadaptation involving long-lasting altered behavioral and neurochemical responses. A single exposure to psychostimulants or morphine is sufficient to induce persistent locomotor sensitization, as well as neurochemical and electrophysiological changes in rodents. It provides a unique model to study the bases of long-term behavioral plasticity. Our study tested the hypothesis that Angiotensin II AT₁ receptors are involved in the neuroadaptive changes induced by a single exposure to amphetamine (AMPH) and that such changes are related to the development of neuroplasticity. Wistar male rats (250-300 g) were pretreated with an AT₁ blocker, candesartan (3 mg/kg po) for five days and after that injected once with AMPH (5mg/kg ip). The Fos immunoreactive neurons (Fos-ir) in response to AMPH (0.5 mg/kg) were determined 3 weeks later. Our results showed an increase in Fos-ir neurons in AMPH pretreated rats in Hippocampus dentate gyrus and Prefrontal cortex and this response was prevented by the AT₁ receptor blockade. The brain RAS should be considered for a better understanding of the mechanisms involved in psychostimulants-induced neuroadaptive changes.

322.
NQ20 - SPINAL LUMBAR RHIZOTOMY AND THE STUDY OF GROWTH INHIBITORY MOLECULES IN REGENERATION

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Sensory neurons of the dorsal root ganglia are a popular model for investigating the consequences of injury due to an unusual characteristic – they send one pseudounipolar axon into both the peripheral and central nervous systems. While the former is capable of regenerating after injury (e.g. in the popular sciatic nerve crush model), regeneration of the same axon in the central root fails when it encounters the astrocytes of the spinal dorsal root entry zone. While the peripheral injury is a simple procedure, the central injury is far more invasive and difficult to perform. Here, we describe the anatomical guides and functional prerequisites for successful application of this model in Sprague-Dawley rats. Of particular importance is knowledge of the functional anatomy within the vertebral column, in order to ensure an accurate and limited lesion to the relevant spinal root. We describe the performance of lumbar spinal laminectomy on xylazine/ketamine anesthetized adult rats, the identification of the spinal roots corresponding to the peripheral sciatic nerve, and their axonotmesis. Issues of animal wellbeing are considered. This procedure allows the evaluation of factors contributing to failed regeneration at the peripheral/central boundary, the testing of potential interventions to overcome this barrier, and the morphological and functional assessment of correct recovery at the spinal level.

323.
OC3 - NOTCH SYSTEM INVOLVEMENT IN STEROIDOGENESIS, VIABILITY AND MIGRATION OF HUMAN OVARIAN TUMORAL GRANULOSA CELLS (KGN)

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 IBYME-CONICET.*

Introduction: Ovarian cancer is a solid tumor that has a temporally response to treatments. The Notch system is involved in tumoral angiogenesis and it also participates in the survival of tumoral cells. Particularly, Notch 3 and Jagged 2 (members of the Notch system) are increased in ovarian carcinomas. However, there are no studies about the role of this system in tumoral granulosa cells (TGC).

Objectives: To study the function of Notch system in steroidogenesis, apoptosis and migration of human ovarian tumoral granulosa cells (KGN: cell line established from an ovarian granulosa cell carcinoma) and some intracellular pathways. Methodology: In KGN cells in culture in the presence or absence of a Notch system inhibitor (DAPT), we determined: a. progesterone and estradiol levels by RIA, b. PARP cleavage by western blot, c. cellular migration by wound assay, and d. Phosphorylated-AKT levels by western blot

Results: The inhibition of Notch system decreased the synthesis of progesterone and estradiol ($p < 0,05$; $p < 0,0001$, respectively). We also observed an increase in PARP cleavage levels ($p < 0,05$), a decrease in cellular migration ($p < 0,05$), and a decrease in the levels of phospho-AKT ($p < 0,05$).

Conclusion: The Notch system is involved in fundamental cellular parameters of TGC, in part through activation of PI3K/AKT pathway. We conclude that the Notch system is implicated in the processes studied in human ovarian tumoral granulosa cells.

324.
OC5 - SURVIVAL DETERMINATION IN WISTAR-LEWIS RATS WITH COLON CANCER INDUCED BY 1, 2 DIMETHYLHIDRAZINE

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Colon cancer is one of the most common causes of death in the world. Many animal models were developed in which induced chemical carcinogenesis is obtained and is comparable to that in human beings. 1, 2 dimethylhydrazine (DMH) is a potent alkylating agent which produces a reliable cancer model in rodents. Total survival was determined in Wistar-Lewis rats with induced colon cancer. 23 rats were taken and divided into 2 groups: 5 animals were the control group and received no treatment while the rest were administered 20 mg/kg DMH once a week for 8 weeks. Each animal was weighed weekly and the survival data was recorded daily. The small and large intestines were analyzed pathological and histologically by routine techniques. Sample data was processed using SPSS and Kaplan-Meier curves were obtained. This type of rats show a life span of approximately 1095 days and the rats with induced colon cancer in this experiment, extend their survival up to 246 days.

325.

OC8 - LEPTIN INFLUENCES MAMMARY CARCINOGENESIS IN HYPOTHYROID RATS

López Fontana C, Maselli ME, Sasso CV, Semino S, Zubiría MG*, Jahn GA, Carón RW

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We aimed to determine the influence of the hormonal background induced by hypothyroidism on mammary carcinogenesis. Female Sprague-Dawley rats were treated *per os* with a single dose of DMBA (15mg/rat) at 55 days of age and divided into two groups: euthyroid (EUT, n=17) and hypothyroid rats (HYPO, 0.01% PTU in drinking water, n=16). All the animals were weighted weekly and observed until the appearance of the first palpable tumor. The latency, incidence and progression of tumors were determined in the two groups. Whole blood samples and a piece of normal mammary gland and tumor were taken for hormone determinations and histological analyses. Statistical analysis was performed by Student T test and Chi square (IC>95%). HYPO showed significantly retarded growth and reduced levels of circulating GH. The latency of onset of tumors was longer, and the incidence and tumor growth rate were lower in HYPO than in EUT. The histopathology was similar in the two groups. No statistical differences were observed in prolactin levels. However, circulating leptin was significantly lower in HYPO than EUT even though body fat mass was similar in both groups. To note, leptin levels were higher in HYPO rats that developed mammary tumors than in non tumoral HYPO. In conclusion, low serum leptin concentrations may be responsible of the decreased carcinogenesis observed in HYPO rats.

326.

OC9 - BLADDER CANCER: TUMOR PROGRESSION IS ASSOCIATED WITH CADHERIN SWITCH AND BETA-CATENIN NUCLEAR RELOCALIZATION

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In bladder tumors, loss of epithelial cadherin (cadE), presence of neural (cadN) and placental (cadP) cadherin ("cadherin switch") and aberrant β -catenin localization is observed. β -catenin is phosphorylated in Ser 33 for degradation but, in abnormal conditions, it accumulates, translocates to the nucleus and activates genes related to invasion/metastasis. We have reported a decrease in cadE in a murine bladder cancer model, namely MB49/MB49I cells, being the latest more invasive. The aims of the research were: 1) To study the "cadherin switch", by evaluating expression of cadE, cadN and cadP and 2) To characterize expression of β -catenin. Studies were done in MB49I and MB49 cell cultures and orthotopic tumors using Western Immunoblotting, Immunocyto/histochemistry. 1) Low levels of mature cadE (120 kDa) were found in MB49 and no signal was detected in MB49I, 2) CadN was detected in both cell lines and cadP only in MB49I, 3) Total levels of β -catenin were higher in MB49I 4) β -catenin nuclear accumulation was found in MB49I cultures and tumors 5) A Ser33/fosfo- β -catenin lower signal was detected in MB49I compared to MB49 tumors. In conclusion, the study first describes changes in β -catenin phosphorylation during progression of bladder cancer using a murine experimental model.

327.

OC10 - HISTOPATHOLOGICAL DIAGNOSIS FREQUENCY OF NEOPLASMS IN COMPANION ANIMALS

Redondo E, Schleaf N, Alcoba A, Bagnis, G, Maffrand C. UNRC.

The aim of this work was characterize the type and biological behavior of different tumors in companion animals by histopathology. An record of retrospective and prospective major malignancies affecting small animals, was performed using frequency tables. Samples were taken from the cases that were submitted to diagnostic lab of the Animal Pathology Department of Universidad Nacional de Río Cuarto. Samples were fixed in 10% buffered formalin solution (4% formaldehyde). The histological paraffin embedded sections were stained with hematoxylin-eosin (H-E). Special stainings were used in some cases. A total of 553 specimens was submitted to pathological diagnosis between years 2009 to 2011. Over the total, 55 (10%) corresponding to oncology cases, of which 3 were cats and the rest were dogs. The average age of the animals with tumors was between 6 and 8 year old, although a large proportion of the processed chips lacked such information. The frequency of females was higher (n = 26) than males, although there were 14 entries without specifying gender. With respect to the breed, the Boxers presented more neoplastic cases, however, 15 records did not specify breed. The cross-breed had the highest rate of tumors presentation. Skin tumors accounted for the largest proportion according to the affected organ being nearly 50% (n = 27) of the total samples processed. Over all cases, 20% (11/55) were squamous cell carcinomas (SCC), correlated with high frequency of skin tumors, that represented the majority of tumors in small animals. The SCC is the most frequent neoplasia. The lack of data is the most common mistake in casuistry recording of the Department of Animal Pathology.

328.

OC13 - APE1 EXPRESSION IN BIOPSIES FROM BREAST CANCER PATIENTS TREATED WITH ANTHRACYCLINES NEOADJUVANT MONOCHEMOTHERAPY

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Doxorubicin (DOX) produces reactive oxygen species which damage DNA. This damage may be repaired by the Base Excision Repair system (BER). APE1 is a key enzyme in BER. Objectives: 1) to determine the APE1 expression before and after chemotherapy with DOX or epirubicin (EPI) and 2) to correlate APE1 with drug response and patient survival. Thirty-three patients received 75 mg/m² of DOX or 120 mg/m² of EPI, during 4 cycles before surgery, and 6 cycles with CMF after surgery. Nuclear expression of APE1 (evaluated by immunohistochemistry) increased after drug administration (p=0.03). No correlation was found between nuclear/cytoplasmic APE1 expressions with clinical/ pathological response. APE1 expression did not correlate with patient survival. Our results suggest that APE1 expression is not a useful predictive/prognosis marker in breast cancer. cancer.djuvant DOX/ EPI monochemotherapy.

329.
PV3 - PRODUCTIVITY OF *Menodora decemfida* (OLEACEAE) UNDER DIFFERENT LEVELS OF IRRIGATION

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CONICET- IADIZA.

This species is a pre-Andean endemism of forage value that inhabits the mountains of the region of Mendoza, San Juan, La Rioja and Catamarca. In Mendoza, it is distributed in the foothills, on slopes of southern exposure between 1200 m and 1400 m; at higher altitudes (1,600 m) it occurs on sunny sites and at lower elevations (900 m) on shady areas. The objective was to determine productivity levels of *Menodora decemfida* with supplementary irrigation for forage production purposes. Work was conducted over two growing seasons in the experimental field of CCT-Mendoza. Average annual rainfall (1901-1950) is 194 mm, with an average temperature of 16.1°C. The experimental design was a Latin square with 5 treatments (irrigation levels and cutting intensity) and 10 replications (plants). The treatments were: 1 - Control without irrigation and cutting at the end of 2 years, 2 - Systematic irrigation every 30 days using 1 liter/ plant (200 m³/ha/year) and periodic cuts at 10 cm height, 3 - Systematic irrigation every 15 days with 1 liter/plant (400 m³ year) and cuts at 10 cm height, 4 - Systematic irrigation every 7 days with 1 liter/plant (800 m³ year) and cuts at 10 cm height, and 5 - Systematic irrigation every 7 days with 1 liter/plant (800 m³/ha/year) and periodic cuts at 5 cm height. The design consisted in setting plots of 1.2 m x 0.5 m, totaling 16,660 plants/ha. Analysis of variance was favorable to treatment 4, with a productivity of 800 kg/ha. This treatment received 800 m³ of water/ha and intense periodic cutting at 10 cm height. Results show that, water intake being equal, cutting intensity hinders productivity. This species responds to irrigation and is easily replanted, making it possible to increase plant density and productivity in the field.

330.
PV4 - PHENOTYPIC CHARACTERIZATION OF PANICUM COLORATUM VAR COLORATUM FOR SALT TOLERANCE

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Lack of reliable phenotyping protocols has hindered progress in molecular and genomic studies for the increase in stress tolerance in forage grasses. The aim of this study was to design a protocol for the identification of *P. coloratum* var *coloratum* materials adapted to saline conditions at the establishment stage. The cultivars of this species are stabilized populations, thus intracultivar variability is to be expected. The trials were conducted in a greenhouse, plants were grown on sand and perlite irrigated with nutrient solution. Trial duration was 600 °D, simulating the initial stage of vegetative growth. Three salinity levels were tested: 0, 200 and 400 mM NaCl. Leaf appearance and tillering rates, relative growth rate, leaf protein, chlorophyll, and Na⁺ and K⁺ concentrations were assessed. Two indexes were calculated with this information, IP integrated growth variables in the absence of stress, and IT reflects the relative alteration of those variables under stress conditions. None of the materials combined high growth in the absence of stress and salt tolerance. Some materials combined high growth under stress with a high degree of tolerance while others exhibited low values for both variables. Variability for growth under stress in *P. coloratum* var. *coloratum* could be identified with this protocol.

331.
PV6 - OPTIMUM PERIOD OF COOL SEASON GRASSES WINTER OVERSEEDING ON *Cynodon dactylon* IN THE SOUTH-CENTER CÓRDOBA, ARGENTINE

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South-center Córdoba turfgrasses are based on Bermudagrass (*Cynodon dactylon*) that go dormant in fall, so overseeding with cool season species is needed to keep turf green all year. There is no information about optimum overseeding date (OD). Temperatures that cause Bermudagrass growth rate decrease and those that allow the rapid establishment of the species for overseeding, must be considered. The aim was to determine the optimum overseeding period of *Lolium perenne*, *L. perenne* Excel I, *Lolium multiflorum* and *L. multiflorum* Axcella in the south center of Córdoba. Four species were overseeded in 5 OD every 15 days. Ground cover, visual quality and biomass were measured and air and soil temperatures were recorded. The first 4 OD had higher values of coverage (90%) and visual quality (4.5) than the 5th OD, due to Bermudagrass produced less competition. In 2011, average soil temperatures (AST) and minimum air (MAT) during the trial period were below regional normal values. The 1st OD, done at thermal values above the optimal range, performed well, due to a decrease in MAT and AST after seeding. Although, 4th OD showed a good performance, Bermudagrass growth rate had declined, causing a low quality turf. All genotypes tested are suitable for overseeding, when TMS and AST ranging between 18.4 and 19.2°C and 9.5 and 10.4°C, respectively. In years of normal temperatures, forward or delay the OD, would result in low quality overseeding.

332.
PV7 - EFFECT OF SOIL WITH MANURES BIRD IN THE SAFETY AND QUALITY LETTUCE

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Lettuce is the main leaf vegetable produced in the Río Cuarto, where crop producers use soil-applied manure aviaries. The purpose was to evaluate two poultry manures incorporated into soil on quality and safety of lettuce and in the microbiological status of the soil. The study was carried out at the UNRC, with two cultivars: Waldmann's Green and Rapidmor, planted on 23/09/09, 22/04 and 09/09/10, and harvested after 90-120 days. The chicken manure was incorporated in two doses (2.5 and 5 kg.m²) and a urea to the control (12 and 24 g.m²). The design was RCB with 6 treatments, 4 repetitions and a sample of 10 plants each. The plots were 2.5 m² with 112 lettuce plants each. Was recording: green weight (BW), dry weight (DW) and length (L) of plants and compared to ANOVA. Microbiological counts were performed on manure, on harvested manure-soil and lettuce. The plants from plots with manure showed higher percentages of BW, DW and L than the plants in the plots with urea, the values being 43, 36 and 33% respectively, with no differences between manures. The plants with urea showed the lowest water content, 8.5% of DW compared to 6.7 and 7.1% in plants with hen and chicken manure. Only in the first planting time there were differences between doses, the highest dose showing the highest values of BW, DW and L. The maximum counts of *E. coli* were 9.9x10⁴ in chicken manure, at harvest the counts in soil and lettuce were below the 1000 ufc.gr. *Salmonella* was not found. The manure had a positive effect on quality of lettuce and did not show any risk of microbiological contamination.

333.

PV8 - MANURE EFFECT INCORPORATED ON THE GROUND OVER NITRATE CONCENTRATION IN LETTUCE LEAVES

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Nitrate toxicity is considered in lettuce when it overcomes the barrier of maximum permitted levels, set at 2500 mpp for summer productions and 4000 mpp for winter productions. This study evaluated the levels of nitrates in lettuce grown in soils with addition of manure. This work was made with Waldmann's Green Rapidmor cultivars planted on three dates: 23/09/09, 22/04 and 09/09/10. Prior to each planting, hen manure (HM), chicken manure (CM) and urea (U) were added in the control plots. For each fertilizer were used 2 doses (MAX and MIN). The design was CAB with 6 treatments, 4 replicates and a sample unit of 10 plants. We analyzed the concentration of nitrate in soil in the early crop stage and the leaves of lettuce at harvest. Comparisons were performed using ANOVA. The concentration of nitrate in soil presented a difference in the HM:MAX. Analyzing the dose and the fertilizers independently, there is a higher concentration on the HM fertilizer and on the MAX dose. In lettuce leaves, nitrate concentrations also showed a difference in the HM:MAX. The concentrations of nitrate do not exceed the reference value. These results show that the quantity and type of manure used by the local horticultural farmers in the lettuce crop, generate nitrate concentration in leaves close to the limit acceptance for human consumption.

334.

PV9 - PEANUT POD PRODUCTION IN GENOTYPES WITH DIFFERENT GROWTH HABIT AND BRANCHING PATTERN

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Virginia and Spanish peanuts market types are differentiated by their growth habit, branching degree and reproductive buds allocation that form patterns of pod distribution and contributions from the branches to crop yield. The objective of this study was describing the dynamics of pod number and weight per branch category in different cultivars. Every 10-13 days were sampled pod, leaves and stems of each branch into two genotypes grown under non-limiting conditions at three sowing dates (2009/10) and growth curves were built (start, end, duration and rate). The most productive branches were cotyledonary, other n+1 and n+2 cotyledonary in both genotypes, with 93 and 89% of the total yield for Virginia and Spanish cultivar, respectively. These branches had an earlier onset, the fruit appearance and growth rate higher and longer duration. The partition factor was differential between them and is determined probably by sink size; that is higher in branches which first define yield components that give them a comparative advantage over those of later development.

335.

PV11 - THE EFFECT OF NANOINSECTICIDES ON THE FEEDING ACTIVITY AND NUTRITIONAL PHYSIOLOGY BY OF STORED PRODUCT INSECT PESTSStefanazzi N^{1,3}, Stadler T², Buteler M², Ferrero AA².¹Dto. de Biol., Bioq. y Farmacia, UNS. B. Blanca, ²Lab. Tox. Ambiental (IMBECU), CONICET-Mendoza; ³CONICET.

Antifeeding activity and the alteration in nutritional physiology of *Sitophilus oryzae* and *Tribolium castaneum* adults were evaluated using disks of wheat flour with nanostructured alumina (NSA) or diatomaceous earth (DE) as positive control at concentrations of 0, 0.1, 0.05, 0.025, 0.0125, 0.00625 and 0.00312% (w/w). Flour disks were prepared according to the method of Huang *et al.* (2000) with some modifications. The nutritional indices calculated were: relative growth rate (RGR); relative consumption rate (RCR) and efficiency of conversion of ingested food (ECI). The antifeeding effect (AE) was also calculated. Positive values represented a feeding deterrent effect and negative values a feeding stimulant effect. In adults of *S. oryzae*, NSA significantly ($p < 0.05$) reduced the RCR at the concentration of 0.05%. Mortality observed was 78 and 80% at the concentrations of 0.05 and 0.1% respectively and feeding deterrent action was observed at the highest concentration (0.1%). In adults of *T. castaneum*, NSA reduced the RCR at the concentrations of 0.1, 0.05, 0.0125 and 0.00625% ($p < 0.01$) but no feeding deterrent action or mortality were observed. Our results showed that NSA had post-ingestive toxicity in *S. oryzae* and decreased food consumption in *T. castaneum*.

336.

PV12 - FUMIGANT ACTIVITY OF TWO SPECIES OF ALOYSIA AGAINST *Tribolium confusum* (COLEOPTERA: TENEBRIONIDAE)Benzi VS^{1,2}, Stefanazzi N^{1,2}, Murray AP^{1,3}, Ferrero AA².¹CONICET, ²Departamento de Biología, Bioquímica y Farmacia. UNS., ³INQUISUR. UNS. E-mail: veronicabenzi@conicet.gov.ar

Continuing with the research of the bioactivity of the essential oils of native plants, the objective of this work was to evaluate the fumigant activity of the essential oils of *Aloysia polystachia* (Ap) y *Aloysia citriodora* (Ac) against *Tribolium confusum*. The essential oils were extracted from fresh leaves using a Clevenger-type apparatus. *T. confusum* is a susceptible strain that was reared on wheat at 30° C y 65-75% h.r. To determine the fumigant toxicity of essential oils, filter papers were impregnated with 40µg of ethanol solutions at different concentrations. Each filter paper was attached to a glass vial, and each vial was introduced inside a glass flask of 40 mL with a top. Ten adults of *T. confusum* were placed inside the flask. Five independent replicates were conducted. Mortality was evaluated at 72 h. LC50 was calculated by Micro Probit 3.0. Treatments do not differ significantly. (CL50 Ap: 5.92 mg/L air, CI 95%: 5.29-6.58, X²: 2.75) (CL50 Ac: 5.53 mg/L air, CI 95%: 2.64-6.81, X²: 0.12).

337.

PV13 - THERMAL REQUIREMENTS FOR THE GERMINATION OF COOL SEASON SPECIES USED FOR OVERSEEDING TURFGRASS

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Fall overseeding on *Cynodon dactylon* is done with cool season grasses. The aim of this work was to determine the thermal requirements for germination of *Poa trivialis*, *Lolium multiflorum*, *L. perenne* y *L. perenne* x *L. multiflorum*. Constant temperatures (8, 12, 16, 20, 24 and 28 °C) and variable temperatures (30/20; 28/16; 24/14 and 20/10 °C, 8/16 hrs) were evaluated to estimate the germination power (PG), the base temperature for germination (Tb), and the thermal time (Tt), lag time (Tlag), average time (TMG) and total time (TFG) of germination for each species. Data were studied with ANOVA and regression analysis. The Tb levels found for all species were =3°C. The Tt required to reach 50-75% of germination was between 100 and 370 °Cd for the analyzed species. The Tlag increased with decreasing temperature, from 3 to 7 days at 28°C and 17 to 26 days at 8°C. The TMG increased with decreasing temperature and varied with the species. The TFG and PG were highly influenced by temperature in each species, being generally 11 to 22 days at 28°C and 33 to 44 days at 8°C. Overall, PG reduction was due to constant extreme temperatures -high and low, and to high variable temperatures ($p < 0,001$), increasing the proportion of dormant seeds with low constant temperature and dead seeds with high constant and variable temperatures. Levels depended on the species.

338.

PV15 - EFFECT OF SALINITY ON THE GROWTH AND IONIC LEVELS IN OLIVEOlivieri G¹, Turchetti J², Ruiz M^{1,3}, Vita F³.¹FCFyN. ²Unidad Integrada INTA-UNSJ. ³EAA San Juan INTA. E-mail: moruiz@sanjuan.inta.gov.ar

The salinity tolerance of olive is associated with the crop's ability to restrict the transport to leaves of Na⁺ and Cl⁻ and maintain high cellular levels of K⁺. This study aimed to determine the effects of salinity on growth and distribution of ions in the plant in two cultivars of *Olea europea* L., cv. Barnea and Arbequina. The same was carried out in the Agricultural Experimental Station (INTA San Juan), Argentina, Plants (240), were maintained in greenhouse conditions for two seasons (2009-2010). The treatments applied were saline solutions (NaCl) of 4 dS/m, 6 dS/m and 8 dS/m, and a control of 2dS/m, were determined in each morphometric parameters (stem growth and number of leaves) and chemical (ion content in root, stem and leaf). The growth of both cv decreased as salinity increased, being Barnea the most affected. With respect to the distribution of ions are significant differences between the cv in the leaves levels of Na⁺ and Cl⁻. The results indicate that high concentrations of Cl⁻ were most harmful leaf growth than Na⁺ concentrations being most affected Barnea.

339.

PV16 - DAMAGE CELLULAR FOR SALINITY AND COLD IN OLIVESDias I¹, Olivieri G¹, Ruiz M², Vita F².¹FCFyN. ²EAA San Juan INTA. E-mail: moruiz@sanjuan.inta.gov.ar

The station freezes and soil salinity represents the main obstacle for the cultivation of the olive tree in San Juan. At which this study aimed to evaluate the combined effects of salinity and cold in seedlings of *Olea europea* L. cv. Barnea and Arbequina. The same was carried out in the facilities of the Agricultural Experimental Station (INTA San Juan), Argentina. Plants (240), were maintained in greenhouse conditions, The treatments applied were saline solutions (NaCl) of 4 dS/m, 6 dS/m and 8 dS/m, and a control of 2dS/m, after a year of application of these treatments, they underwent cold (0°C, -5°C and -10°C) in individual leaves and in complete plants. The cellular damage was quantified by means of the test of relative electric conductivity (CER) in leaf. Applied salt treatments induced changes in response to cold in both leaves and plants. There were higher values ??of (CER) in both leaf and in plants subjected to -10°C (80% CER) in those samples came from the treatments saline (6 and 8dS/m) for 25% of CER in control. This implies that as the salinity of the medium increases the level of cell damage caused by the cold increases. This can be attributed to Na⁺ and Cl⁻ has adverse effects on cell membranes. Similar values were obtained CER in leaves and whole plants after subjecting them to low temperatures, indicating that the test to evaluate chilling is equally effective in leaf that in complete plant.

340.

PV18 - PHENOTYPIC, GENOTIPIC AND SYMBIOTIC CHARACTERIZATION OF AN ALFALFA (*Medicago sativa* L.) NODULATING STRAIN

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Alfalfa is the most important forage legume in Argentina and its productivity could be increased if it would be associated to high efficient rhizobia in biological nitrogen fixation. The objective of this study was the phenotypic and genotypic characterization of a *Sinorhizobium meliloti* 3DOh13 strain. Plant growth-promoting properties of this strain was tested by measuring the phosphate solubilization of Ca₃(PO₄)₂, FePO₄, AlPO₄ and indole-3-indol acetic acid (IAA) production. The genotypic characterization included DNA fingerprinting, 16S rDNA gene amplification and plasmid profile analysis. Tests of simple inoculation in plastic bags were carried out in order to study the kinetic of nodulation the total number of nodules was compared with the strain *S. meliloti* B399 (INTA). Competitiveness assays were performed by co-inoculation in glass tubes with mineral medium in which they challenged *S. meliloti* 3DOh13 and *S. meliloti* B399. We analyzed the occupancy percentage of nodules of each of the inoculated strains. The bacteria were differentiated using the markers of resistance to rifampicin (Rif) and streptomycin (Sm). The strain *S. meliloti* 3DOh13 solubilized Ca₃(PO₄)₂, and produces IAA. We did not detect any plasmid accessory when using the Eckhardt *in situ* lysis technique. The number of nodules and dry weight of shoots at the end of the assay was greater in plants inoculated with *S. meliloti* 3DOh13. The occupancy rate of nodules by *S. meliloti* 3DOh13 was lower than *S. meliloti* B399. We hope to obtain information on the response of alfalfa to inoculation with this strain and the results can be extended to the formulation of a new inoculant.

341.

PV19 - CROP OLIVE TREE AS DEVELOPMENT ALTERNATIVE FOR FRUIT GROWING OF NORTHERN SAN LUIS

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Fruit growing development in sparsely exploited sites requires preliminary studies ambient and economic viability. The proposal was obtain data climate, select cultivars of possible adaptation, and studies financial assessment in Quines. Thermal supply winterly Quines (87/08) was 425 +/-124 Hf, 510 +/- 202 Uf (Utah); average period vernalization 100 +/- 19 days, subtracting average 12 days warm. Interval frost 99 +/-35 days and low frequency. Coldest June, July and August did not exceed -4,2°C medium intensity. Loam sandy soil with optimal drainage; Proper depth without physical impedance, pH neutral 60 cm deep, and moderate risk erosion. Irrigation water at shallow depth and provision Quines and Conlara rivers. Cultivars Arbequina, Arbosana, Picual Coratina,, Barnea are appropriate. Semi-intensive system requires US\$ 5,000/ha in first three years, whose profitability is increases from 4% to 250% of the 4th to 8th year. The Provincial Fruit Promotion plan could enable soon recovery these costs, very important in the yield Olive tree, a kind of late start production.

342.

PV20 – PEACH (*Prunus persicae* L.) VARIETIES FOR QUINES-CANDELARIA ZONE IN SAN LUIS. PROVINCELucero RA¹, López P¹, Arjona C², Orta FJ¹, Rojas E¹, Consigli Robles FV¹.¹FICES. UNSL; ²Fac. Ciencias Agrarias. UNCuyo.

The peach (*Prunus persicae* L.) is the most important species of tree fruit in Argentina, are the new most dynamic varieties of market. The study area includes: the sites Quines and Candelaria, Ayacucho locations (north of the province of San Luis). Was revised and interpreted the characterization agro - climate regional by analyzing the parameters of winter chilling and soil properties, then relate this to eco-physiological requirements of the species and thus determine possible peach varieties for the region. It methodology was conducted by reviewing the requirements for hours of cold and climate data for the reference area of a record of 57 common varieties of Argentina. Was established maximum limit of 250 hours of cooling, as the average minimum value recorded in the area, thereby limiting the introduction of varieties with excessive cooling requirements for the area. The results are: The properties of the soils are characterized by light-textured sandy loam, good drainage, good depth, with no physical limitations and neutral pH up to 0.6 m; The winter heat supply Quine (1997/2008) was 425 +/- 124 Hf. The average frost regime was 99 +/- 35 days, with little daily basis, recorded during the period June, July and August months of highest intensity, yet they did not exceed -4.2°C medium intensity. 9 varieties were selected with a requirement less than or equal to 250 hours of cold: Flordastar, Tropicsnow, Flordaprince, Sunred, Sunsplash, Flordaglo, Flordagold, Tropic Beauty and UF Golg. Recommend adequate validation through field trials to determine their true behavior. In plants with more than 250 Hf requirement., varieties such as "San Pedro" and "Don Augustine" (300 chill hours), is recommended cyanamide applications when the cold winter might be insufficient.

343.

RE3 - OVARIAN HYPERSTIMULATION SYNDROME (OHSS): WHAT HAPPENS WHEN INHIBITS VEGF (vascular endothelial growth factor) SYSTEM?Scotti L¹, Abramovich D¹, Pascuali N¹, Pazos C¹, Bas D¹, Tesone M^{1,2}, Parborell F¹.¹IByME and ²FCEyN, UBA, Bs. As., Argentina.

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Ovarian hyperstimulation syndrome (OHSS) is an iatrogenic complication caused by the induction of ovulation in fertility treatments, which involves an overproduction of steroid hormones and vasoactive substances. The aim of this study was to analyze the effect of VEGF inhibition in a rat OHSS model on steroidogenesis, stability and vascular permeability. Sprague Dawley rats were injected prepubertal eCG (10 IU) and 48 hours later with hCG (10 IU) (Control). The OHSS group was injected with eCG (50 IU / day) for 4 consecutive days and 24 hours later with hCG (25 IU). The group OHSS + TRAP received TRAP (VEGF inhibitor) the day of hCG injection. The rats were sacrificed 48 and 72 hs. post hCG. The serum was obtained to measure estradiol (E2) and progesterone (P4) levels by RIA. Ovaries were isolated for histological sections to detect α -actin (cell periendothelial marker) by IHQ and to isolate proteins by western blot to measure Claudin-5 (vascular permeability marker). In the group OHSS+TRAP, a decrease of P4 and E2 levels at 48 hours (p<0.05) was observed. The cell periendothelial area decreased in the OHSS+TRAP group compared to the OHSS group without treatment (48h: p<0.01; 72h: p<0.05). In addition, TRAP increased the expression of claudin-5 compared to the untreated OHSS group (p<0.05). Conclusion: These results show that inhibition of VEGF in a rat OHSS model affects steroidogenesis and stability of blood vessels. In addition, this treatment reduces the vascular permeability which is a characteristic of this syndrome.

344.

RE4 - THE INHIBITION OF PDGF SYSTEM (platelet-derived growth factor) AFFECTS FOLLICULO-GENESIS IN OVARIES FROM HYPERSTIMULATED RATSPascuali N¹, Scotti L¹, Abramovich D¹, Tesone M^{1,2}, Parborell F¹.¹IByME and ²FCEyN, UBA, Bs. As., Argentina.

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Several angiogenic systems regulate selection of dominant follicle destined to ovulation. However, no study demonstrated the involvement of PDGF system on follicular dynamic. The aim of this work was to study the role of PDGF on folliculogenesis. eCG-treated prepubertal rats were injected with a selective inhibitor of PDGF receptor (AG1295 AG, 20 or 50 μ g/ovary) in one ovary and the contralateral ovary with vehicle (C). The rats were sacrificed 24 and 48 hs. post hCG. The ovaries were removed for histological sections. The ovarian sections were stained with H&E. The results showed a significant decrease in the number of preantral follicles (PF) and early antral (EAF) in the group treated with both doses of the inhibitor compared to the control group (p<0.05) (24-48hs). Only with the higher dose and 24 hs of treatment, a significant decrease in the number of antral follicles (AF) was observed (p<0.05). AG1295 inhibitor (20 and 50 μ g/ovary) increased the number of atretic follicles (Atret.F) (24-48hs) compared to the control group. In addition, we observed the presence of hemorrhagic follicles only in the group treated with the AG1295 inhibitor at 24 hours of treatment. Conclusion: PDGF receptor inhibition causes a decrease in the number of PF, EAF and AF, and an increase in the number of Atret.F. In addition, the AG1295 inhibitor causes the development of hemorrhagic follicles. The mechanism could be through a reduced vascularization and/or by a direct effect on ovarian follicular cells.

345.

RE5 - INCREASED SUPPLY OF DIETARY OMEGA 3 PUFAs AND REPRODUCTIVE FUNCTIONS OF MALE MICE

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Several nutritional factors can affect the mammal reproductive functions; among them, PUFAs $\omega 6$ and $\omega 3$ modify structural and functional characteristics of the sperm membranes. We analyzed the influences of cod liver oil (rich in $\omega 3$ PUFAs) on functional activity of epididymal sperm from *Albino swiss* mice. Two groups were considered: **C** (Control; $\omega 6/\omega 3$ ratio=19.87; n=10) and **P** (Problem; $\omega 6/\omega 3$ ratio=1.13; n=10). Daily food consumption and body weight were quantified during three months. Mice were sacrificed and concentration, motility (Makler chamber), viability (H33258 staining), hypoosmotic swelling test (HOST), spontaneous acrosome reaction and DNA fragmentation (TUNEL) were evaluated in epididymal gametes. An improvement tendency was found in P and C respectively: (Mean \pm SEM) concentration 25.3 ± 2.7 vs 23.5 ± 3.7 ($\times 10^6/\text{mL}$); motility 90.8 ± 1.5 vs 88.3 ± 1.5 (%); HOST 85.1 ± 2.9 vs 80.1 ± 5 (%). From these results, we can suggest that an increased supply of dietary PUFAs $\omega 3$ can optimize sperm functional activity. Further studies must be performed in order to confirm these effects.

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346.

RE6 - APOPTOSIS CASPASE-DEPENDENT IN CROSSBRED PORCINE PLACENTAS

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The apoptosis is a process in which several processes are involved related to maintain cell homeostasis. The mechanisms that regulate apoptosis are essential to the normal placental development. Extrinsic and intrinsic signaling pathways activate a group of amplifying enzymes, the caspases, that function as initiators, effectors and executioners in the cascade of apoptotic events. Our purpose was to study the fragmentation of DNA through TUNEL assay and the immunolocalization of caspase-3, executioner of the apoptotic process, by immunohistochemistry. Histological samples of $\pm 4 \mu\text{m}$ from placentas of 28, 60 and 114 days of gestation were used. TUNEL (+) nuclei were observed in placental cells at the beginning, the half and the end of gestation. The induction of caspase-dependent apoptosis was determined by the presence of the enzyme in villi, glands and blood vessels in the three gestational periods analysed. Caspase-independent apoptosis was detected in fetal mesenchyme at days 30 and 60 of gestation. We have previously determined the differential activation of the two apoptotic pathways in the remodeling of different porcine placental structures. In conclusion, in the present work we determined that the interconnection of specific apoptotic signals observed are executed, mainly, through the mechanisms dependent of caspases, by the exception of some placental structures in which apoptosis would be triggered by an independent activator.

347.

RE7 - HIGH SCORE OF VEGF RECEPTORS, FLT-1 Y FLK-1, DURING PORCINE GESTATION. PRELIMINAR STUDY

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Swines present a chorioallantoic placenta characterized by an apposition between uterine and chorionic epithelia, interdigitated by fetal and maternal microvilli, involving a vast increase in the area of contact without loss of continuity of membranes. Placental angiogenesis constitutes a pivotal factor during gestation in pigs to accompany the augmentation in blood flow with advancing gestation. This process depends on the expression of different molecules, as the vascular endothelial growth factor (VEGF) and their specific receptors, Flt-1 and Flk-1. Our purpose was to study the angiogenesis in placentas from crossbred swines. Placental samples of $\pm 4 \mu\text{m}$ from 30, 60, 80 and 114 days of gestation were used. The immunolocalization of VEGF receptors, Flt-1 and Flk-1, was performed by immunohistochemistry. The results were expressed as semiquantitative and the distribution of intensity of staining was determined by High Score. Flk-1 was expressed at days 30, 60 and 80 of pregnancy, being negative at term. Temporo-spatial distribution of Flt-1 was elevated at days 30 and 114 days, and negative at day 80. In previous studies, we have determined the distribution of immunointensity of VEGF throughout porcine placentation. According with the observations of the present study, the angiogenic events at the beginning and the end of gestation are associated with the co-localization of VEGF and Flt-1, and at days 60 and 80 Flk-1 would accompany the stimulation of placental angiogenesis induced by VEGF.

348.

RE8 - STRUCTURAL AND ULTRASTRUCTURAL PLACENTAL STUDY IN ANGLONUBIAN GOATS

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The caprine placenta is characterized by migration of binucleate fetal cells (BNCs) to the uterine epithelium. In the present work the structure of term caprine placentas was studied through Gallego's trichromic stain and by High Resolution Light Microscopy (HRLM), and the ultrastructure by transmission electron microscopy (TEM). Nineteen gestating primiparous Anglo-Nubian does (n=5) were fed according to their physiological state. Through trichromic stain trophoblastic epithelial cells, collagen fibers in the fetal mesenchyme and small and medium caliber blood vessels near the epithelium could be visualized. With HRLM, binucleated cells, some of them with numerous cytoplasmic granules, could be observed. Those cells could be ultrastructurally characterized through TEM, differentiating those young of those already mature in the trophoblastic epithelium. HRLM technique resulted in an excellent tool for description and analysis of cell structure and morphology in goat placentas.

349.

RE9 - IMPACT OF EXOGEN GHRELIN ON EARLY EMBRYO DEVELOPMENT AND IMPLANTATION IN MICE

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Embryos and endometrium express Ghrelin (Ghr) and its receptor. Our study evaluated the impact of Ghr and/or an antagonist (Ant=(D-Lys3)GHRP-6; 6nmol/animal/día) injection on embryo development and implantation. Adult female mice were injected (sc) from ovulation induction (Exp1) or from Day3 to Day7 of pregnancy (Exp2) with: Ghr₁ (2nmol/animal/day), Ghr₂ (4nmol/animal/day), Ghr₂+Ant, Ant or isotonic solution (Con=control) and sacrificed at 80hs from estimated ovulation (embryos retrieved by uterine flushing) or at Day18 of pregnancy respectively. Exp1 (67-136 oocytes/group): Antagonist enhanced embryos recovery from uterus (65±17%; p<0.05 vs Con 28±11%, Ghr₁ 23±9% and Ghr₂ 24±8%) and decreased *in vivo* fertilization indexes (75%; p<0.05 vs Ghr₁ 93%, Ghr₂ 92%). Ghrelin and/or Ant increased embryo degeneration and delayed embryonic development (% blastocytes: Ghr₂ 41%, Ghr₂+Ant 29%, Ant 37%; p<0.05 vs Con 66% and Ghr₁ 63%). Exp2 (6-9 females/group): Ghrelin and/or Ant increased the % of females with embryonic loss (fewer fetuses than corpora lutea) (Ghr₁ 33%, Ghr₂ 83%, Ghr₂+Ant 75%, Ant 67%; p<0.05 vs Con 13%) and with fetuses that had stopped growing (Ghr₂ 67%, Ghr₂+Ant 50%, Ant 67%; p<0.05 vs Con 0% and Ghr₁ 0%). These results suggest a biphasic effect of Ghr in modulating *in vivo* fertilization, embryo development and migration, and implantation.

350.

RE10 - EXPOSURE OF PHOSPHATIDYLSERINE IN PARTHENOGENETICALLY ACTIVATED MOUSE EGGS

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Phosphatidylserine (PS) is a phospholipid normally localized to the inner leaflet of the plasma membrane and it has been shown that its exposure is necessary for different cellular events. Recently, we have demonstrated that fertilization induces a transient exposure of PS in mouse eggs. In the present study we evaluated the exposure of PS in parthenogenetically activated mouse eggs. For this purpose, eggs were incubated with already known egg activators, and PS exposure was evaluated by incubation with fluoresceinated-Anexin5 (ANX-5), that binds PS, and microscopy. The incubation with 10 mM SrCl₂, 7% ethanol or 5 μM Ca²⁺ ionophore rendered a 94, 97 and 73% of activated eggs, respectively. Eggs activated by SrCl₂ or ethanol showed a high percentage of ANX-5 labelling (96 and 90%, respectively) and a fluorescent pattern similar to that previously observed in fertilized eggs. By contrast, only 6% of ionophore-activated eggs showed ANX-5 labelling, evidencing the existence of differences between parthenogenetic activation methods. Interestingly, when previously ionophore-activated eggs were inseminated, the fertilized ones showed ANX-5 labeling, suggesting that PS exposure not only depends on the elevation of Ca²⁺. Taken together, these results indicate that the PS exposure observed in mouse eggs would occur as a result of egg activation.

351.

RE11 - EFFECT OF GANGLIONIC CHOLINERGIC STIMULATION ON OVARIAN PHYSIOLOGY IN THE FIRST PROESTRUS IN THE RAT

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It recognized the involvement of nitric oxide (NO) in the ovarian steroidogenesis. Using the *ex vivo* coeliac ganglion-superior ovarian nerve-ovary (CG-SON-O) system, has been shown to stimulating the ganglion with cholinergic agents, modify the release of NO and progesterone (P) ovarian in prepubertal, pregnancy and adult rats. The aim of this work was to study in the same experimental scheme, if CG-cholinergic agents modify the release of NO and P, and the expression of the enzymes 3β-HSD and 20α-HSD (of synthesis and degradation of P respectively) in the Proestrus stage in the first estrous cycle. The system was incubated in Krebs Ringer at 37°C. The working groups were: a) control b) cholinergic agonist acetylcholine (Ach), c) muscarinic antagonist atropine (Atr) and d) nicotinic antagonist hexamethonium (Hex) all in 10⁻⁶M concentration. We determined NO (Griess technique) and P (RIA) in the ovarian compartment at 15', 30', 60' and 120'. The expression of enzymes was determined by RT-PCR, in the ovary to 120'. ANOVA 1 followed by Tukey test with a statistical significance of p<0.05 was used. Cholinergic agents favored the release of NO and inhibited P. Ach increased the expression of 3β-HSD (p<0.05) and antagonists generated an effect "*per se*" on the expression of 20α-HSD. The inhibitory effect exerted on the release of P, could be related to increased release of NO, with recognized antisteroidogenic properties. These parameters and the times studied, the mechanism of action of Ach would not be classical receptors pathway.

352.

RE12 - ADVANCES IN THE STUDY OF FMR1 mRNA EXPRESSION DURING RAT FOLLICULAR DEVELOPMENT

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Among the possible causes of the premature ovarian failure, premutation in the FMR1 gene is a genetic risk factor. In view that the role of the protein FMRP in the ovary is unknown, the aim of the present study was to analyze the expression of FMR1 mRNA during the follicular development in a rat model and to identify alternative *splicing* variants of the gene. Prepubertal rats without any treatment, rats injected with DES and rats superovulated with a unique dose of PMSG were used. mRNA expression was studied by real time PCR (qRT-PCR) in isolated follicles. *Splicing* variants were studied by RT-PCR covering different regions where alternative *splicing* has been described. qRT-PCR preliminary studies showed significant differences in FMR1 expression between groups, when isolated follicles were analyzed. In addition, we identified 3 isoforms resulting from exon 15 alternative *splicing*, besides those previously described resulting from exon 12 *splicing*. Our qRT-PCR results in isolated follicles contradict our previous observations by Western Blot, thus indicating that FMR1 regulation of expression during follicular development occur at different levels. The different FMR1 *splicing* variants found might indicate the existence of several protein isoforms that could be related to its function in the gonad.

353.

RE14 - PROLACTIN FROM COELIAC GANGLION DOES NOT AFFECT THE LUTEAL REGRESSION IN LACTATING RATS

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We have shown that prolactin (PRL), through neural pathway, promotes regression of the corpus luteum (CL) at the end of pregnancy in the rat. In the lactation, serum levels of the hormone are high and its neural effect on the CL of pregnancy, that is still present in the postpartum period, is unknown. The aim of this study was to investigate the effect of PRL in the celiac ganglion (GC), through superior ovarian nerve (NOS) on luteal regression on day 4 of lactating rats. The system *ex vivo* GC-NOS-ovary was used. The system was incubated in Krebs Ringer buffer at 37 °C, keeping the GC and the ovary connected by NOS, in separate compartments. PRL was added (10^{-7} M) into the ganglion compartment. Controls were not stimulated. Periodic extractions of the ovary incubation liquid were taken at different times along 240 min. to measure of progesterone and prostaglandin F 2α (PGF 2α) by RIA and nitrites by the Griess method. At 240 min, the luteal mRNA expression of 3 β -HSD and 20 α -HSD, PGF 2α receptor, ON synthase inducible (iNOS) and bcl-2 and bax (antiapoptotic and proapoptotic factor) were analysed by RT-PCR. ANOVA I followed by Tuckey test with a statistical significance of $p < 0.05$ was used. The addition of PRL in the ganglion compartment did not modify the studied parameters respect to the control group. This leads us to conclude that the neural pathway acts as a fine modulator, thus PRL from GC does not manifest effect because of its high levels endogenous.

354.

RE15 - CABERGOLINE TREATMENT REVERSES THE INFERTILITY IN TRANSGENIC MICE

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Transgenic mice hypersecreting β -subunit of human chorionic gonadotrophin hormone (hCG β +) produce elevated levels of hCG and prolactin. In addition, hCG β + female mice are infertile. The objective of this work was to analyze the influence of prolactin on the estrous cycle and infertility, by treating females with cabergoline, a dopamine agonist that inhibits the production and prolactin release. hCG β + female mice were injected with cabergoline (500 μ g/kg body weight) dissolved in 0,25% methylcellulose (vehicle). The protocol consisted of three doses per week during the fifth and sixth week of age. The estrous cycle was analyzed by daily vaginal smears. Cabergoline-treated hCG β + female mice partially reestablished the estrous cycling capacity throughout the period observed, whereas hCG β + mice injected with vehicle showed a continuous diestrus-type pattern. At eight-week-old, these females were treated with PMSG/hCG to induce ovulation and mated with WT adult male mice. Five over seven cabergoline-treated females showed vaginal plugs after mating and gave birth to living pups. In contrast, no vaginal plug or pregnancy was observed in control hCG β + female mice. In conclusion, cabergoline treatment reverses the acyclicity and infertility hCG β + mice.

355.

RE17 - INVOLVEMENT OF ESTRADIOL ON THE APOPTOSIS OF CORPUS LUTEUM

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Part of the peripheral innervations of the ovary is formed by the Superior Ovarian Nerve (SON), whose fibers originate in the coeliac ganglion (CG). The aim of this study was to analyze whether the presence of oestradiol valerate (E_2) in CG in *ex vivo* system and incubations of ovaries modifies the release of progesterone (P), the gene expression of the 3 β -HSD and 20 α -HSD enzymes, and the expression of genes involved in apoptosis of corpus luteum. We worked with rats on dioestrus II stage. *In vitro* ovarian incubations (experimental scheme A) and system *ex vivo* CG-SON-Ovary systems (experimental scheme B) were used. The incubations were performed in Krebs-Ringer solution pH 7.4 at 37 °C in metabolic bath. In scheme A, E_2 (10^{-8} M) was added in ovary and in scheme B in CG compartment. ANOVA test was applied with a significance of $p < 0.05$. The results showed that when E_2 was added in the ovarian incubation, P decreased, coinciding with the decreased expression of 3 β -HSD, and increased expression of 20 α -HSD ($p < 0.05$). In the *ex vivo* system, E_2 significantly stimulated the release of P at 60 and 180 min ($p < 0.001$), coinciding with the increased expression of 3 β -HSD. With regard of apoptotic markers in (A), Bax showed an increase ($p < 0.05$) while Bcl $_2$ decreased ($p < 0.05$). In (B) Bax and Bcl $_2$ not showed change. The results showed physiologically the presence of E_2 receptors in the CG demonstrating the importance of them in the ovarian steroidogenesis. In addition E_2 in CG, pathway SON protects the Corpus Luteum of events that lead to apoptosis of the same, in this stage of the estrous cycle.

356.

RE21 - DIFFERENTIAL CONTROL OF OVARIAN PROGESTERONE RELEASE BY VIP IN COELIAC GANGLION OF CYCLIC RAT

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We have previously reported that ovarian progesterone (P) in rats on diestrus day2 (D2) is decreased by vasoactive intestinal peptide (VIP) action in coeliac ganglion (CG). In this work we investigate whether ovarian P release is differentially modified in D1, D2 and estrus stages of estrous cycle by VIP effect on CG, and whether nitric oxide (NO)/inducible nitric oxide synthetase (iNOS) system is involved in the ovarian response. For that, the *ex vivo* CG-superior ovarian nerve (SON)-ovary system of Hotlzman virgin rats on D1, D2 and estrus, were used. CG and ovary were placed in separate compartments connected by the SON and incubated with K-R buffer (basal values). After addition of 50 ng/ml VIP in CG, samples from the ovarian compartment were taken at different times along 180 minutes to measure P (by RIA) and nitrite (by Griess reaction). Ovarian iNOS protein expression was determined by Western blot. VIP in ganglion induced an increase of P release ($p < 0,001$), and a decrease of ovarian nitrite levels ($p < 0.05$) and iNOS protein ($p < 0.05$) at D1 and D2, compared with the respective basal values. By contrast, at estrus, after VIP addition in ganglion, a decrease of ovarian P, without change in the NO levels and iNOS expression, were observed. VIP effect in CG could contribute to the increase and decrease of serum P levels, which are known to occur in the diestrus and estrus of the estrous cycle, respectively.

357.

RE22 - SUPERIOR OVARIAN NERVE CONTROLS THE SPLEEN MACROPHAGE APOPTOSIS AND STEROIDS OF POLYCYSTIC OVARY

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We have previously informed that progesterone release from polycystic ovary (PCO) is modified by secretions of macrophages (MΦ) from rat spleen. Now, we investigate whether the superior ovarian nerve (SON) section induces apoptosis and nitric oxide (NO) release in MΦ and also if this is related to the steroidogenic ability of MΦ secretions on ovaries. The PCO was induced in adult rats by a single i.m. injection of estradiol valerate, 2 mg/rat. After 2 months, rats were sacrificed. The *in vivo* SON section (PCO-SON) was performed 7 days before sacrifice. MΦ from control (C), PCO and PCO-NOS rats were isolated from spleen and cultured (1×10^6 cells) for 24 h in RPMI medium. MΦ secretions were used to stimulate androstenedione (A2) and estradiol (E2) release from C and PCO ovaries incubated for 3 h in a metabolic bath. Apoptosis was detected by TUNEL, NO by Griess reaction and hormones by RIA. Both PCO and PCO-SON (MΦ) secretions increase A2 ($p < 0.001$) and decrease E2 ($p < 0.01$) release from PCO ovaries, compared to C ovaries. These effects were higher with PCO than PCO-SON MΦ secretions, in coincidence with a MΦ apoptosis expression: $C < PCO-SON < PCO$, and a NO release: $C = PCO-SON > PCO$. The SON section reverts the MΦ NO release and decreases the apoptosis induced by PCO. The steroid ability of MΦ is modified accordingly.

358.

RE24 - ODF1, LABELLED BY MONOBROMO-BIMANE, IS A SPERM FLAGELAR PROTEIN, ALSO LOCALIZED IN KIDNEY MEDULLA

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Mammalian sperm proteins undergo thiol groups oxidation to form disulfides bonds as they travel through the epididymis during cell maturation. This proteins are involved in motility, sperm capacitation and acrosomal reaction. Our objectives were to determinate which proteins oxidate during epididymal transit, their physiology and if they are present in other tissue. In this work, we used a fluorescent thiol-selective labeling agent, monobromobimane, to study the protein thiol status of caput and cauda rat sperm. Fluorescence signal decreased along the epididymal trip, in sperm head and tail, indicating that both subcellular regions participate in the thiol changes. These sperm proteins also became resistant to be solubilized by SDS. Protein identification by mass spectrometry and sequence database searching correlated this protein with the ODF1. mBBBr specifically bound to N-terminal domain cysteine of ODF1. mBBBr reduce rat and human sperm motility, quantitatively and qualitatively, and the effects are dose dependent, without significant increasing the percentage of dead sperm. Finally, an ODF1 similar peptide were found in kidney, by western blotting techniques.

359.

RE26 - EFFECTS OF CASTRATION ON LYSOSOMAL PROTEINS IN RAT EPIDIDYMIS

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An important function of mammalian epididymis is the participation in sperm maturation, through the secretory activity. Several proteins are secreted actively by the epididymis (lysosomal enzymes). We here attempted to elucidate if secretion of cathepsin D (CatD) and prosaposin, (PSAP) in epididymis is a selective process mediated by receptors (sortilin and/or cation-dependent Man-6-phosphate receptor CD-MPR), and if these mechanisms are regulated by testosterone (Test). Three groups of rats were used; controls, castrated and castrated with hormone replacement (Test). After 48 hs, the epididymides were removed and the three regions (caput, corpus, and cauda) processed separately for the study (immunoblot) of proteins from tissue and the epididymal fluid. A decrease of sortilin expression was induced by castration in the three regions, but only in cauda the effect was partially reversed by hormone replacement, indicating that sortilin is regulated by Test in this zone. In contrast, the CD-MPRs tended to increase due to castration, but hormone replacement did not reverse that effect. In turn, the expression and secretion of CatD tended to increase due to castration and were reversed by the hormone replacement. Meanwhile, PSAP showed no major changes. We conclude that the expression and secretion of CatD in epididymis depend on Test and the increased secretion due to castration could be because a decrease of sortilin and/or a concomitant increase of CD-MPR.

360.

RE27 - CHANGES IN THE MEMBRANE OF BULL SPERMATOZOA DURING EPIDIDYMAL MATURATION

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The epididymis is involved in sperm maturation through the secretion of factors into the lumen; some of them are glycan-modifying enzymes that catalyze molecular changes in sperm membrane. We intend to study the molecular changes that occur in the plasma membrane glycoproteins of bull spermatozoa during their epididymal maturation. Fresh epididymides from bulls (Aberdeen Angus) were dissected into caput, corpus and cauda and the sperms were obtained by slicing of the tissue and centrifugation. We measured the activity of the glycosidases, βN-acetyl-glucosaminidase (NAG), α-mannosidase (MAN), β-galactosidase (GAL) and β-glucosidase (GLU) associated to the gametes. We also evaluated the content of fucose in sperm glycoproteins by using a FITC-conjugated lectin UEA-I (for Flow cytometry and microscopy) or biotin-conjugated UEA-I (for Western Blot). It was observed that NAG activity increased in spermatozoa from caput to cauda, whereas MAN was decreased along the epididymal transit. We also observed that content of fucose on spermatozoa glycoproteins decreased during their epididymal maturation, except a ~50 kDa protein, which is fucosylated in cauda. By fluorescence microscopy, we observed a redistribution of fucose from a strong post-acrosomal to a weak diffuse staining. These changes could provide new insights into molecular rearrangement that can be used as parameters for sperm maturation.

361.

RE28 - UNVEILING THE FUSION MACHINERY DURING CORTICAL REACTIONZanetti N¹, Bello O¹, Pavarotti M², Mayorga L², Michaut MA^{1,3}.¹Lab Biol Rep, ²Lab Biol Cel y Mol, IHEM-CONICET; ³ICB UNCuyo, Mendoza Argentina. E-mail: mnataliaz@fcm.uncu.edu.ar

Cortical granules exocytosis (CGE) is an event that blocks polyspermy. Enzymes released during this exocytosis, modify the zona pellucida of the egg, preventing other sperm fusion. The molecular mechanism by which cortical granules fuse with the plasma membrane has not been fully elucidated and so far only four proteins have been associated with this exocytotic process: Rab3A, Rabfilina 3, syntaxin 4 and SNAP-25. Our goal is to analyze the presence of other unidentified exocytotic proteins and study their role in the CGE, such as VAMP2 and NSF. To determine the involvement of these proteins in the CGE, we performed Western blot, indirect immunofluorescence, microinjection of antibodies and functional assays in mouse oocytes. The results showed that VAMP2 is present in mature and immature oocytes, and localized in the cortical region. Functional assays, using tetanus toxin (block VAMP2 function), showed that the CGE was inhibited. In addition, NSF was also detected in mature and immature oocytes, with an increase in expression and a redistribution of the protein during oocyte maturation. Functional assays, where anti-NSF antibody was microinjected, showed that the CGE was also inhibited. The results show that VAMP2 and NSF are expressed in mouse oocytes, localizes in the cortical region of mature oocytes, and that both proteins are required for the CGE during the cortical reaction.

362.

RE29 - SHORT NATURAL PHOTOPERIOD STIMULATES TESTICULAR ACTIVITY OF CHINCHILLABusso JM^{1,3}, Ponzio MF^{2,3}, Dominchin MF^{1,3}, Fiol de Cuneo M^{2,3}, Ruiz RD^{2,3}.¹Fac Cs Ex Fis y Nat, ²Fac Cs Méd, Univ Nac Cba, ³CONICET. E-mail: jmbusso@conicet.gov.ar

Hystriomorphic rodents are particularly well represented in South America; among them, *Chinchilla spp* is threatened and the presence of remaining colonies in the Argentinean Andes is uncertain, although a domestic form is widespread in breeding farms. *Chinchilla lanigera* exhibits reproductive rhythms with two breeding peaks in spring and summer; however, there are not studies about photoperiod effects on its reproductive physiology. Domesticated males (n=7) were studied to determine the effects of natural photoperiod upon testicular activity in Córdoba (31°S–64°W) during May, August, November, and February; urine (24h) was obtained before to semen collection by electroejaculation. Differences in androgen excretion (ng/mg of creatinine) were detected by repeated measures ANOVA, the highest values were found associated with "short" light/dark cycles (autumn-winter); acrosome intact sperm percentages were higher during winter-spring than summer-autumn, and the lowest sperm concentration was detected in summer (p<0.05). However, seminal volume and sperm functional activity showed non significant seasonal changes. These results suggest that the chinchilla male gonadal activity is affected by natural photoperiod; therefore, it is possible to postulate that this species would be a "short-day" breeder.

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363.

RE30 - INHIBITION OF GLYCOLYSIS AND KREBS CYCLE KEY ENZYMES. EFFECT ON CAPACITATION AND ACROSOME REACTION OF PORCINE SPERM

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The objective of this work was to determine the participation of glycolytic pathway and Krebs cycle in capacitation and AR in porcine spermatozoa, by inhibiting phosphofructokinase I, isocitrate dehydrogenase (IDH) and malate dehydrogenase. Sperm were diluted in PBS with 0,1% BSA, washed and resuspended in capacitation médium (mTBM) with bicarbonate. To evaluate capacitation, samples were incubated at 39°C and 5% CO₂ for 2 hours in the presence of different concentrations of specific inhibitors of the mentioned enzymes. Motility was evaluated by optic microscopy, viability by the eosin-nigrosin technique and capacitation by CTC. To evaluate acrosome reaction, sperm previously capacitated were incubated in the presence of follicular fluid at 39°C and 5% CO₂ for 15 minutes in the presence of the same concentrations of the enzyme inhibitors. Viability and true acrosome reaction were evaluated by DIC. The addition of high concentrations of all the inhibitors used diminished motility without affecting viability (excepting IDH) and produced a dose-dependent inhibition of capacitation and acrosome reaction. These results suggest that the presence of specific inhibitors of glycolytic and Krebs cycle key enzymes would negatively affect the capacitation and acrosome reaction processes, indicating that these pathways would be involved in the generation of the ATP necessary for these processes.

364.

RE32 - EFFECT OF HEMATOPOIETIC GROWTH FACTORS DURING *IN VITRO* MATURATION OF BOVINE OOCYTESMorado S², Conde P^{1,2}, Smitz J³, Cetica P², Gentile T¹.¹IDEHU- FFyB, ²INITRA-FCV, UBA; ³Vrije Universiteit Brussel.

Hematopoietic factors, such as erythropoietin (Epo) and kit ligand (KITL), and their receptors have been described in the ovary. However, their role is not clear enough. The aim of our study was to evaluate the effect of Epo and KITL during *in vitro* maturation of bovine oocytes. Cumulus oocyte-complexes (COCs) were recovered by puncturing ovarian antral follicles and matured at 39°C, 5% CO₂ in humidified air in medium 199 with gonadotropins supplemented with: 1) bovine serum albumin (BSA, negative control), 2) BSA + Epo (7, 14, 20 or 80 mU/ml), 3) BSA + KITL (5 or 50ng/ml), 4) fetal calf serum (FCS, positive control). After 22 hours of incubation, COCs were denuded to evaluate the presence of the 1st polar body, or co-incubated with a suspension of 1x10⁶ motile spermatozoa/ml for 20 hours in IVF-mSOF. Presumptive zygotes were cultured for 24 hours in IVC-mSOF to analyze the percentage of cleaved embryos. No significant differences were observed in the percentages of nuclear maturation with the addition of Epo or KITL. The percentages of cleaved embryos were higher with the addition of Epo in the maturation medium at all concentrations with respect to negative control (p <0.05). In the presence of KITL a trend to improved cleavage rates was observed. In conclusion, hematopoietic growth factors may not be involved on nuclear maturation but could have a positive effect on cytoplasmic maturation of bovine oocytes matured *in vitro*.

365.

RE33 - RYANODINE RECEPTOR EXPRESSION DURING OOGENESIS IN *Bufo arenarum*: IMMUNOHISTOCHEMICAL STUDY

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An increase in intracellular calcium concentration is critical for initiating egg activation and subsequent embryo development. Two families of channels are involved in the release of calcium from intracellular stores: the inositol 1,4,5-triphosphate (IP3) receptors, present in oocytes of all studied species and the ryanodine receptors, detected only in oocytes of some species such as sea urchin, mouse and human, but not in amphibians. Previous studies in our laboratory allowed us to determine that ryanodine receptors are present in *Bufo arenarum* oocytes and are involved in the maturation and activation process., together with IP3 receptors. There are no reports about which stage of folliculogenesis they can be detected at. The aim of this study was to analyze the expression of ryanodine receptors during oogenesis in *Bufo arenarum*. Samples of ovary from adult females in gonadal recovery period were processed by immunohistochemistry. By using a mouse monoclonal anti-ryanodine receptor (34C-SIGMA), we could establish that the expression of ryanodine receptors begins during folliculogenesis and they become evident from the stage of late vitellogenesis. The appearance of such channels during oogenesis would be one of the biochemical changes oocytes undergo, leading to the acquisition of competence to mature.

366.

RE34 - CONSERVATION OF PORCINE OVARIES AFFECTS REACTIVE OXYGEN SPECIES AND MATURATION OF OOCYTES

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We previously observed a decrease in oocyte nuclear maturation due to conservation of ovaries by long times at low temperatures (4-6 h at 15-25°C). The aim was to evaluate reactive oxygen species (ROS) production, redox state and cytoplasmic *in vitro* maturation of oocytes recovered from ovaries preserved at short times and high temperatures (2-4 h at 25-35°C). Cumulus-oocyte complexes were recovered from antral follicles and matured 48 h at 39°C, 5% CO₂ in medium 199 with gonadotropins. Fertilization was performed by 18 h in mTBM with fresh semen. Cytoplasmic maturation were evaluated by presence of decondensed sperm heads and/or pronuclei. ROS production and redox state were measured in immature and matured oocytes by 2',7'-dichlorodihydrofluorescein diacetate and 2,3,4,5,6-pentafluorodihydrodrotetramethylrosamine fluorochromes. There were no differences in cytoplasmic maturation rates when ovaries were conserved by 2 h at 25°C and 35°C, however a decrease was observed after 4 h at 35°C (P<0.05). Immature oocyte ROS level was higher in ovaries conserved by 4 h at 35°C (P<0.05), however after maturation decreased to similar values. Oocyte redox state increased after maturation regardless preservation conditions. ROS production seems to increase in oocytes due to storage of ovaries by long time at high temperature impairing cytoplasmic maturation. Oocyte redox state would not vary at these conservation conditions.

367.

RE35 - STUDIES ON THE EXPRESSION OF NEURAL CADHERIN IN MURINE GAMETES AND ITS PARTICIPATION IN FERTILIZATION

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Neural cadherin (Ncad) participates in cell-cell adhesion in a calcium-dependent manner. Since fertilization involves cellular adhesion and requires calcium ions, we propose the participation of Ncad in fertilization. The aims of the study were: 1) to assess Ncad expression in murine reproductive tissues and gametes, 2) to evaluate its participation in fertilization. Methods involved Western immunoblotting, immunocytochemistry and gamete interaction assays with anti Ncad antibodies. Ncad (135 kDa) was immunodetected in protein extracts from sperm, unfertilized eggs, granulosa cells, testis and ovary. Ncad was localized to the acrosomal region alone (A pattern) or with the equatorial segment (A+ES pattern) of non-capacitated (A: 40±8% and A+ES: 52±6%; mean±SEM, n=7) and capacitated (A: 42±8% and A+ES: 39±7%) intact sperm. The signal was found in the equatorial segment (ES pattern) in sperm that underwent acrosomal exocytosis (ES: 56±6%). Ncad was also immunolocalized in the egg plasma membrane (EPM). Sperm preincubation with anti Ncad antibodies (20 µg/ml) did not affect sperm binding to the EPM (IgG: 19±2, anti Ncad: 16±1 bound sperm), although it resulted in a reduction in the % of eggs with fused sperm (IgG: 74±4%, anti Ncad: 44±10; p<0.05). In conclusion, Ncad is expressed in murine reproductive tissues and gametes and would participate in events leading to sperm fusion with the egg.

368.

RE36 - GALECTIN-1 EXPRESSION IN HUMAN ENDOMETRIOTIC AND ENDOMETRIAL TISSUE

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A continuous effort is being done to elucidate the pathogenesis of endometriosis (EDT). Galectin-1 (Gal-1) plays key roles in immune tolerance, tumor-immune escape and neovascularization. The potential role of Gal-1 in EDT has not yet been reported, so our goal was to evaluate Gal-1 protein expression and its histological location in human endometriotic and endometrial tissue. Biopsies from endometriotic lesions (n=12) and eutopic endometrium (n=6) from women with EDT, and proliferative (n=10) and secretory (n=9) control endometrium, were analyzed by Western blot (WB) and immunohistochemistry (IHC). In addition, immunocytochemistry was performed on primary cultures of endometrial stromal and epithelial cells. IHC assays revealed a strong cytoplasmic expression of Gal-1 specifically confined to stromal cells from endometriotic lesions and eutopic endometrium from patients and controls. The same Gal-1 expression pattern was found in human primary endometrial cell cultures. However, we found no significant differences in the levels of Gal-1 protein among all groups. This is the first study reporting the stromal expression of Gal-1 in ectopic lesions and eutopic endometrium from women with EDT, supporting a potential role for Gal-1 promoting the establishment and development of EDT.

369.

RE38 - SEASONAL VARIATIONS IN mRNA β LH FROM MALE TOAD *Rhinella arenarum* (AMPHIBIA, ANURA)

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Rhinella arenarum expresses seasonal changes in androgen synthesis. Androgens fall in the reproductive period (R) (Sep-Dec) while progesterone and 5α -pregnanedione increase. In mammals, the hypothalamus-pituitary-testis axis (HPG) is regulated by testicular steroids through a negative feedback. In amphibians, however, the role of androgens and estrogens in the control of HPG axis is not clear. The objective of this work is to study seasonal changes in β LH mRNA to measure its normal values in each reproductive season. For this, adult males of *R. arenarum* were collected along the year. For β LH mRNA analysis, total RNA from each pituitary was extracted and β LH semi-quantified by RT-PCR. Specific primers were designed from previous sequencing analysis. The full β LH sequence (JN031567) is 93% homologous with the β LH precursor mRNA of *Bufo japonicus* (AB085662.1) and between 80% and 90% with other anurans. Results indicate that LH is elevated during the post-reproductive period (Jan-Apr) and decreases during the pre-reproductive period (May-Aug). In spring, when breeding season starts, LH increases at values higher than during the other two periods. Conclusions: Androgens could regulate β LH by negative feedback.

370.

RE39 - PENTOSE PHOSPHATE PATHWAY CONTROL IN PORCINE OOCYTE *IN VITRO* MATURATION

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Glucose consumption by cumulus oocyte-complexes (COCs) during *in vitro* maturation (IVM) may in part be fated to pentose phosphate pathway (PPP). We previously observed a statistical association between PPP activity and oocyte nuclear *in vitro* maturation. The control of the pathway by pharmacological modulators will contribute to the understanding of PPP participation on oocyte maturation. The aim of this work was to evaluate PPP activity, oocyte nuclear maturation, and glucose uptake during IVM in the presence of an inhibitor of the dehydrogenases of the pathway (6-aminonicotinamide, 6-AN). COCs were recovered by aspiration of antral follicles of ovaries from slaughtered gilts and matured 48 h at 39°C, 5% CO₂ in medium 199 with gonadotropins (control) and with different concentrations of 6-AN (0.01, 0.025, 0.05 and 0.10 mM). PPP activity was evaluated by Brilliant Cresil Blue stain, glucose uptake was determined spectrophotometrically, and oocyte nuclear maturation by the presence of metaphase II. There were dose dependent decreases in PPP activity and in oocyte nuclear maturation rates (P<0.05). However, glucose uptake diminished with 0.025 mM of 6-AN respect to control (P<0.05), but there was no further decrease at higher concentrations of the inhibitor. These results contribute with new evidences about the participation of PPP on porcine oocyte *in vitro* maturation process.

371.

RE40 - HEPARIN REQUIRES ADENYLATE CYCLASE PARTICIPATION TO MODULATE OXIDATIVE METABOLISM VARIATION AND TO INDUCE SPERM CAPACITATION

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Heparin is a glycosaminoglycan presents in genital tract of bovine female that induces sperm capacitation *in vitro*. It has been identified an adenylate cyclase presents in the membrane (ACm) that produces intracellular cAMP and induces capacitation and protein tyrosine phosphorylation. Capacitation of bovine sperm requires oxidative substrates in order to obtain energy. ATP production can be evaluated through the determination of oxygen consumption, an indicator of mitochondrial function. The aim of this study was to evaluate adenylate cyclase participation in capacitation induction and in oxidative metabolism variation of cryopreserved bovine sperm treated with heparin. 2',5'-dideoxyadenosine (100 μ M) was used as an ACm inhibitor. Capacitation was evaluated by the chlortetracycline epifluorescent technique and viability by Trypan blue stain. Oxygen consumption was measured polarographically with an oxygen electrode modified Clark type. Data was analyzed by ANOVA and Tukey test (P < 0.05). The ACm inhibition blocked capacitation induction and the respiratory burst produced by heparin. Oxygen consumption values were $7.91 \pm 2.38 \mu\text{LO}_2/\text{h}/10^8$ sperm vs $14.76 \pm 2.80 \mu\text{LO}_2/\text{h}/10^8$ sperm with heparin/2',5'-dideoxyadenosine and heparin, respectively. Sperm viability was similar in both treatments ($50.8 \pm 4.15\%$ vs $53 \pm 3.86\%$). Heparin requires ACm activity to modulate oxidative sperm metabolism and capacitation induction.

372.

RE41 - ENERGY METABOLISM ENZYMES: CAPACITATION AND ACROSOME REACTION IN CRYOPRESERVED BOAR SPERMATOZOA

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The aim was to determine the participation of glycolytic pathway and Krebs cycle in capacitation and acrosome reaction (AR) in cryopreserved with (VE) or without (C) α -tocopherol boar spermatozoa. Sperm were incubated with bicarbonate as capacitation inducer in the presence of different concentrations of specific competitive inhibitors of Phosphofructokinase I (PPKI), Isocitrate dehydrogenase (IDH) and Malate dehydrogenase (MDH). To evaluate acrosome reaction, capacitated sperm were incubated with follicular fluid in the presence of the same inhibitors. Motility was evaluated by optic microscopy, viability by the eosin-nigrosin technique, capacitation by CTC and true acrosome reaction were evaluated by DIC. All the inhibitors used diminished motility without affecting viability, and produced a dose-dependent inhibition of capacitation and AR. Optimal inhibition concentrations of capacitation and AR was different between C and VE for PPKI (C: 0,5mM y 1mM vs VE: 1mM y 2,5mM) and MDH (C: 7,5mM y 5mM vs VE: 10mM y 7,5mM, respectivamente), without differences for ICDH (5mM, p>0.05). These results indicate different participation of the glycolytic and Krebs cycle in the production of energy required for capacitation and AR in cryopreserved boar semen. The different inhibitory concentration between treatments (C and VE) could be due to the protective effect of α -tocopherol on sperm plasma membrane.

373.

RE45 - SUCKLING DIFFERENTIALLY MODULATES HYPOTHALAMIC HORMONE RECEPTOR EXPRESSION IN LACTATION DEFICIENT OFA *HR/HR* RATS

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OFA *hr/hr* rats present lactation deficit, elevated hypothalamic DA system sensitivity, overexpression of Tyrosine Hydroxylase, partially blocked suckling-induced prolactin and oxytocin release. In an attempt to determine possible causes of the altered sensitivity of DA system we measured hypothalamic expression of PRL (the long form, PRL_{long}), progesterone (PR total and PRB isoform), estrogen (ER α isoform) and thyroid hormones (TR α 1, TR α 2, TR β 1, TR β 2 isoforms) receptors using real time quantitative RT-PCR and serum PRL by RIA in mid-lactating rats separated from their pups for 12 h and subsequently suckled for 0 (S), 2 (S/s2h) or 4 h (S/s4h). 2 or 4 h suckling induced significant PRL release and decreased significantly mRNA levels of PRL_R, ER α , TR α 1, TR α 2, TR β 1 and TR β 2 without changes in the expression of PRs (total or B isoform). These results indicate that hypothalamic PRL_R, ER α , and TR expression are regulated by suckling in lactating OFA while PRs, one of the main modulators of dopaminergic hypothalamic neurons, do not change.

374.

RE46 - SERUM AND PORCINE PLACENTAL CONDITIONED MEDIUM EARLY PREGNANCY FACTOR QUANTIFICATION

Grosso MC, Martínez RA, Bellingeri R, Motta C, Alustiza F, Picco N, Moliner D, Busso L, Vivas B.
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Early Pregnancy Factor (EPF) is a molecule with immunosuppressor and growth factor activities involved in pregnancy. The main of this study was to quantify EPF in serum of pregnant sows and porcine placenta conditioned medium (PPCM). Crossbred non-pregnant sows sera (n=10) and from 10 (n=6), 30 (n=8), 60 (n=8) and 90 (n=7) days of pregnancy (dp) (San Ambrosio college farm) and placentas from 30 (n=6), 60 (n=5) and 90 (n=4) dp were used (Río Cuarto slaughterhouse). Placental tissue (5g) was homogenized, cultured in McCoy's 5A (5 days, 37°C), ultracentrifuged (30 min, 22.000g), supernatant filtered (0.22 μ m) and stored (-20°C) until used. EPF was quantified by densitometry. Western blot with rabbit anti-porcine EPF IgG was performed. EPF bands were digitized and processed by "ImageJ" analysis program (<http://rsbweb.nih.gov/ij/>). Color relative intensity of EPF pattern (1 μ g/ μ l), sera and MCP bands corresponding to EPF were determined. Different EPF concentrations were detected in sow sera: 0.017 μ g/ μ l (non-pregnant), 0.631 μ g/ μ l (10 dp), 1.39 μ g/ μ l (30 dp), 0.568 μ g/ μ l (60 dp) and 0.866 μ g/ μ l (90 dp). In PPCM, EPF concentrations were: 3.088 μ g/ μ l (30 dp), 0.702 μ g/ μ l (60 dp) and 0.185 μ g/ μ l (90 dp). The highest concentration of EPF in sera and PPCM was at 30 dp and could be related to immunomodulatory function at implantation time. In sera, a minor peak of EPF at 90 dp was found, probably related to growth factor role during exponential growth of embryos, it would be acting in a systemic manner.

375.

RE47 - SILDENAFIL AND L-NAME: EFFECTS ON PLACENTAL EFFICIENCY IN MICE. PRELIMINARY RESULTS

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The aim of this study was to determine the effects of Sildenafil and L-NAME on fetal growth and placental efficiency in mice. Pregnant Balb/c mice were used (n=16) and divided into 3 groups: 1) control, 2) L-NAME (50 mg / kg) applied on day 6 of pregnancy and 3) Sildenafil (10 mg / kg) administered on day 7 of pregnancy. The animals were euthanized on day 17 of pregnancy. Embryonic vesicles were counted in each female. In right horn uterine fetuses were measured and weighed with its corresponding placenta. Fetal weights in the L-NAME group were significantly reduced compared to control group (p = 0.0398) but did not alter placental efficiency. No significant differences were observed in fetal and placental weight, and placental efficiency in Sildenafil group. In conclusion, L-NAME could be used as a model of intrauterine growth restriction. The placental efficiency and growth restriction are not affected by Sildenafil in normal female mice.

376.

RE48 - RELATION BETWEEN BOVINE OOCYTE MATURATION AND MODULATION OF NITRIC OXIDE PRODUCTION

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Nitric oxide is a ubiquitous free radical that plays a role in oocyte activation and maturation, embryonic development and implantation. The aim of this work was to determine NO participation in the production of reactive oxygen species (ROS) and bovine oocyte *in vitro* maturation. Cumulus-oocyte complexes were recovered by aspiration of antral follicles from ovaries obtained from slaughtered cows and cultured in medium 199 (control), 0,5 mM N-nitro-L-arginine methyl ester (L-NAME), endothelial NO synthase inhibitor, 10 μ M sodium nitroprusside or 100 μ M detanonoate, NO donors, at 39°C, 5% CO₂ in humidified air for 22h. 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. ROS levels fluctuated during the 22h of maturation, showing a similar pattern with the different treatments. COCs cultured in the presence of NO donors showed a significantly lower maturation percentage (20% vs. 80%; p<0.05), with the remaining oocytes stopped at the germinal vesicle-MI stage. COCs cultured in the presence of L-NAME showed no difference in maturation. These results suggest that exogenous NO affects the process of bovine oocyte meiotic maturation *in vitro*.

377.

RE49 - CELL DEATH DURING FOLLICULAR ATRESIA IN *Dipetalogaster maxima* (HEMIPTERA: REDUVIIDAE)Aguirre S¹, Pons P², Settembrini B³, Rubiolo E¹, Canavoso L¹.¹Dpto Bioq Clin, CIBICI-CONICET, Fac Cs Qcas, UNC. ²Ctro Microsc Electr, Fac Cs Med UNC. ³Museo Arg Cs Nat-CONICET.

In insects, unfavorable physiological, environmental or nutritional conditions induce changes in the ovarian tissue, which in turn elicit the atresia of some terminal follicles and the final oocyte resorption. Although recent studies in some species indicate that during atresia, the follicular and/or nurse cells can be eliminated by programmed cell death mechanisms, these events have not been explored in Triatominae. The aim of this work was to investigate the cell death mechanisms operating during follicular atresia induced by nutritional deprivation in *Dipetalogaster maxima*, a vector of Chagas' disease. For the study we performed light, fluorescence and electron microscopy, TUNEL assays, western blot and immunofluorescence. The results showed that in *D. maxima*, morphological changes during follicular atresia were established gradually. While during early atresia some follicular cells exhibited nuclei with an apoptotic pattern, during late follicular atresia cells displayed pronounced changes, exhibited cytoplasm vacuolization and abundant autophagic vacuoles. As expected during a process of food deprivation, an intense fluorescence pattern compatible with LC3-II, a marker for autophagy, was observed at the atretic stages. The findings provide a better understanding of unexplored aspects of the biology of reproduction of Chagas disease vectors, which have a clear impact in the life cycle of the females.

378.

RE50 - ROLE OF ACID HYDROLASES DURING FOLLICULAR ATRESIA IN A VECTOR OF CHAGAS' DISEASELeyria J¹, Aguirre S¹, Fruttero L¹, Carlini C², Rubiolo E¹, Canavoso L¹.¹Dpto Bioquímica Clínica, CIBICI-CONICET, Fac Cs Químicas, UNC. ²Dpto Biofísica, UFRGS, Brasil.

In insects, unfavourable nutritional conditions induce biochemical and cellular changes in the ovarian tissue such as the early degradation of vitellin (Vt), the main oocyte protein, and the atresia of some follicles. In some species, the changes accomplished during atresia are elicited to promote the recycling of proteins to sustain younger follicles. In this study, we have explored biochemical changes that take place during the process of follicular atresia induced by a nutritional deficit in the hematophagous *Dipetalogaster maxima*, a vector of Chagas' disease. We have emphasized in the roles of two acid hidrolases, cathepsin D (CD) and acid phosphatase (AP). The study included assays of western-blot, the measurement of enzyme activities, immunofluoresce as well as *in vitro* assays of Vt proteolysis employing specific inhibitors of CD and AP. Results showed that proteolysis of Vt occurred gradually during the transition from vitellogenesis to follicular atresia. Higher activities of AP and CD were registered at early and late atresia, in comparison to pre-vitellogenesis and vitellogenesis. *In vitro* assays showed that AP is important in promoting Vt degradation, which in turn was mediated by CD. Altogether, the biochemical changes exhibited during follicular atresia in *D. maxima* could reflect the ability of the ovarian tissue to face a new vitellogenic cycle if nutritional conditions improve. The results also provide a better understanding of the biology of reproduction of Chagas' disease vectors.

379.

RE52 - SPERM SELECTION TECHNIQUES IN *Lama glama* FRESH SEMEN FOR *IN VITRO* EMBRYO PRODUCTIONSanta Cruz R¹, Carretero M^{1,3}, Arraztoa C^{1,3}, Miragaya M^{1,3}, Giuliano S^{2,3}.Cátedras de ¹Teriogenología y ²Física Biológica, ³INITRA, Facultad de Ciencias Veterinarias, UBA.

The objective of sperm selection is to separate spermatozoa from seminal plasma and to obtain a large percentage of motile spermatozoa. The objective of this study was to asses different sperm selection techniques in fresh semen of llama for *in vitro* embryo production. Using electroejaculation, a total of 21 semen samples were obtained from 7 llama males (n=7, r=3). The ejaculates were incubated in a solution of 0,1% collagenase and then split in 4 aliquots: one of them was layered over a column of Androcoll-E™ (A), another over a column of Percoll (45%) H-HAM-BSA medium (P), another aliquot was treated using swim up technique (S1) and the last one, was treated using swim up technique previously plasma seminal separation. Aliquots A y P showed higher percentages of progressive motility and plasma membrane functionality (p≤0.05) than raw semen. There was not significant differences (p>0.05) in sperm viability between raw semen and treatments, but A showed higher percentages of living spermatozoa than S1 y S2 (p≤0.05). Aliquot A had a higher number of progressively motile sperm (p≤0.05). According to this results sperm selection by colloidal centrifugation would be the indicate method to obtain a higher proportion of progressively motile sperm and integrity and functionality of plasma membrane.

- A**
- Abal A AH22
- Aberastain E FT55
- Abilleira E CL10
- Abramovich D RE3, RE45
- Abud J FT28, FT29
- Abud MA FA29, FA32, FA33
- Accialini P OC3
- Acevedo D BT20
- Acosta JC EB66, EB71, EB72, EB73, EB77, EB81, EB83, EB91, EB100, EB101, EB102, EB105, EB116, EB117, EB118, EB129, EB130
- Acosta M RE22, AH14, AH15
- Acosta N CL13
- Adamo MP MI58
- Adarvez S S4-2
- Agostini E BT28
- Agostinis F BF15, BT12
- Aguilar C BM33, BM36
- Aguilar EG BV15, BV19
- Aguilera AC RE26, RE27
- Aguilera JA BF8
- Aguilera M BT16
- Aguilera Merlo C AH9, AH17
- Aguirre S RE49, RE50
- Aiassa D FT45
- Ajmat MT RE33
- Albarracín G BV15
- Albarracín L FA31
- Alberto MR S3-1
- Alcoba A OC10
- Alé A FT27
- Aliandro OE MI12, MI9
- Allegretti L MI8
- Alonso AM MI24
- Alonso L RE6, RE7
- Altamirano L FT6, FT8
- Alustiza F BM32, RE46, RE47, MI68 BT20
- Alvarez GM RE34, RE39
- Alvarez JÁ EB7
- Álvarez MG BT4, BT5, BT6
- Amaiden MR CL16
- Amat G FT15, FT18
- Amaya C BM4, BM24
- Ambrosio LF BT28
- Amé MV FT41, EB125
- Andersen A RE24
- Andino N S6-3, S4-2, EB61, EB74
- Andrada N BV21
- Andrés JA PV18
- Angel SO MI24
- Ansaldo M FA11
- Antoniazzi CE EB89
- Anzulovich AC BM13
- Aostri CA EB113
- Aparicio JP EB127
- Aragón L BV36
- Arana MF EB26
- Aranibar JN EB7, EB44
- Aranovich EJ S2-2
- Arce O BD4
- Archuby D FA11
- Arenas GN BM17
- Arenas N MI8
- Arias AJ FA12
- Arias J Sat 1-2
- Arias R FA18, FA19
- Arjona C PV19, PV20
- Arranz S EM18
- Arraztoa C RE52
- Arredondo NJ EB67
- Arvia A BF12
- Astudillo GV EB129, EB130
- Attademo AM FT11, FT12
- Augusto M FT46
- Aun L EB4
- Avanza MV BV3
- B**
- Babelis G MI62
- Bacchetta C FT27, EB34
- Baeza M BT27
- Bagnis G OC10
- Baiardi G NQ17, NQ18
- Baigori M MI59, MI60, MI62
- Balboa E BT12
- Ballester S EB36
- Balmaceda RB FT47
- Baluszka I EB128
- Bannoud N RE26, RE27
- Bär Lamas M EB14
- Baraño RI RE36
- Baraquet M EB15, EB23, EB24
- Barbeito CG AH18, BD6
- Barbosa AO EB113
- Bárcena NM BV33
- Bas D RE3
- Bastón JI RE36
- Batalles SM CL7
- Batista S AH22
- Baudo J CL3, CL8
- Bazzoni G BF4, BF5, BF6
- Becker A EB9
- Beckwith EJ S2-2
- Beconi MT RE41, RE48
- Bedano JC EB9, EB13
- Bee de Speroni N BA6
- Bejarano C FT16
- Belgorosky D OC9
- Bellingeri R BT20, BM32, MI68, RE46, RE47
- Bellis LM EB76
- Bello O RE28
- Belotti M FT32, FT16
- Benardon ME BM19, BM30
- Benedetto M BA3
- Beninato V S4-2, EB75
- Benitez L FA12
- Benitez SG NQ12, NQ13
- Bensi N FA3, FA6, FA7, FA8
- Bentivegna DJ EB33
- Benuzzi D BM10, MI27, MI29, MI38
- Benzi VS PV12
- Bergero MF BT8
- Berli DE CL7
- Berli F BV20
- Berni J S2-2
- Bertiller M EB14
- Bertola Compagnucci A CL7
- Bertoldi MV BM4, BM24
- Bertolotti MA EB40
- Bertoluzzo MG BF15, BT12
- Bertoluzzo SM BF15, BT12
- Bettera S MI40
- Bianco M FA3, FA8, MI50
- Bianconi S RE5
- Bigatti G BM38
- Bilotas M RE36
- Binotti S FA3, FA6, FA7
- Biolé F EB25, EB37
- Bionda C EB16
- Biscaro AT CL13
- Bistoni MA FT41, FT42
- Blanco G EB66, EB72, EB73, EB91, EB105
- Blarasin M MI40, EB13
- Boarelli P Sat 1-3, RE24
- Bocanegra MV BM19
- Bocaccini AR BT29
- Boeris PS MI70, MI72
- Boetto N BM6
- Bogni C AH10, MI13, MI14
- Bollini A BF4, BF5, BF6
- Bolzán A BF12, BM31
- Bonamico NC BV9, BV25
- Bonanse M EB39
- Bonanse R EB125
- Bonanse SR BM39
- Bonaventura M EM5
- Bongiovanni M BV32
- Bonifacio AF EB4
- Bonilla F RE33
- Bonivardo SL EB94, EB96
- Bonomi M BM26, BT13, BT15
- Borghi C S4-2, S6-3, EB61
- Bosch B FT45
- Bosio A EB30
- Bosquiazzo VL FT24
- Bottini R BV20, BV40
- Bourguignon N EM5
- Brancolini F EB87
- Brasca AP CL7
- Bravo MA CL11
- Bregonzio C NQ19
- Breininger E RE30, RE41
- Bressan F BT16
- Bronzi D RE17
- Brun A AH28
- Bruno MA NQ15, NQ16
- Bühler Gramajo MI S3-2, FA12, RE33
- Bulfon M BA6
- Burton G NQ1, NQ5
- Busso JM EM6, RE29
- Busso L BM32, BT20, MI68, RE46, RE47
- Bustamante C CL10
- Buteler M PV11
- C**
- Cabagna-Zenklusen M FT12
- Cabrera GC EB56, EB57, EB58
- Cabrillana M RE24
- Cacciamani VE BM19, BM30
- Cáceres CS IM6
- Cadena V BT29
- Cagnolo SR CL17, EB40
- Calandra RS RE15
- Calderón C FT15, FT16, FT18, FT32, FT34, FT37, FT40
- Calderón ML FA29
- Callegari E IM6
- Calvente V BM10, MI29
- Calvo J MI27, MI38
- Cámara A MI58, MI69
- Cámara J MI58, MI69
- Camí G BV36
- Caminos T PV6, PV13
- Campos CM S6-3, EB74
- Campos V S4-2
- Campos VE EB74, EB75
- Canavoso L FA23, RE49, RE50
- Cánepa M EM18
- Cangiano MA FT43, FT44
- Cannizzo B BM40
- Cantero JJ BV6, EB46, EB47
- Cantón MA EB31
- Capelari DN FT22
- Capella P NQ10
- Capmany A BM17
- Cappa F S4-2, EB75
- Caraffa E EB16
- Carbone M MI50
- Carbonell X BT25
- Cardell L FT11
- Cardinali FJ FT47
- Cardoso YP EB87
- Carlier E BV34
- Carlini C FA23, RE50
- Carlini VP FA26
- Carmona N BM36

Carmona Y	EM21	Cohen DJ	RE10	Díaz de Barboza G	BA3
Carón RW	OC8	Coirini H	NQ1, NQ5, NQ15, NQ16	Díaz MC	BD6
Carosio C	EB34	Coll Araoz MV	AH24	Díaz SM	BV35
Carreño NB	RE45	Collado S	BT15	Díaz T	RE8
Carrera A	EB14	Combina M	Sat 3-2	Distel JS	MI36
Carretero MI	RE52	Concha ML	C1	Docherty NG	FT7
Carrizo D	RE11	Conde P	RE32	Dominchin MF	RE29
Carvelli L	RE27, NQ10, RE26	Condomí Alcorta S	NQ3	Dominguez G	CL3, CL8
Casais M	RE14, RE17	Consigli F	PV19	Dominguez S	AH9, AH17, NQ10
Casale CH	CL16	Consigli Robles FV	PV20	Donadel OJ	FT56
Casco C	BF4, BF5, BF6	Contreras HH	Sat 1-2	Donati E	S5-3
Cassano A	FT11	Córdoba M	RE40	Dorati P	BM34, BM35
Castagnolo BM	CL13, MI64	Córdoba MA	EB129, EB130	Dorfman VB	NQ14
Castellanos de Figueroa L	Sat 3-2, Sat 3-4, MI52	Correché ER	FT43	Duarte S	FT16, FT32
Castello L	EB62	Cortez R	EB121, EB64	Dubois D	RE30
Castellón EA	Sat 1-2	Cortiñas TI	FT13, IM6, MI34, MI35, MI66	Ducrós E	MI14
Castillo E	BV30, BV31	Corvi MM	MI24	Durando M	BD10
Castillo G	EB91, EB102, EB105	Costantino V	BM4, BM24	Durantini EN	BT4, BT5, BT6
Castillo V	Sat 1-2	Cozzolino ME	MI36	Durso G	AH22
Castro AE	NQ12	Crespo D	BT16	E	
		Crinó E	EB128	Echaniz S	EB55, EB56, EB57, EB58
Castro AE	NQ13	Cristofolini A	RE6, RE7, RE8	Edelsztein N	RE35
Castro C	BM11, BM40	Croce C	RE26	Eduardo S	EB90
Castro J	BT13	Croce MV	BD3	Egea V	MI8
Castro JF	FT17	Crosta H	EB33	Eiján AM	OC9
Castro M	MI66	Cruceño A	AH9, AH17	Eliás C	CL4
Castro NM	CL13, MI64	Cruzado M	BM11, BM40	Eliás FG	OC5
Castro S	BV34, EB83	Cuartas EI	FA9, FA10	Enriz RD	FT43, FT44
Castro SA	EB100, EB101, EB102	Cuasnicu PS	RE10	Escudero ME	MI19, MI20
Castro-Vazquez A	AH3, BM38, FA29	Cuello AS	S3-1	Escudero NL	BV15, BV19, FT49, FT50
Catalán C	FT17, FT20	Cueto M	S4-1	Espeche C	AH10
Catalan CAN	AH24	Curia A	RE10	Espino ML	FA10
Cataldo A	EB64, EB70	D		Estevez G	BT8
Catanesi CI	BM7	Daguerre P	OC13	Estévez S	PV6, PV13
Cavagnaro JB	EB31	Dain L	RE12	Estrella N	BM11
Cavallera M	FA7	Dalmasso A	PV3	F	
Caviedes Vidal E	BA5, AH28, FA17, FA18, FA19, FA28	Dalvit GC	RE34, RE39, RE48	Fabani MP	FT53, FT54
	Sat 2-2, FT27	Dambolena S	BV38, BV39	Fabra A	BV34
Cazenave J	AH23, FA20, RE38	Damiani MT	BM17	Falico D	EB89
Cecho A	CL3, CL8	Daneri C	RE17	Fassi MF	FT27
Cejas J	BM40	Dave MN	MI39	Fava G	EB72, EB77, EB81, EB102, EB116, EB117, EB118
Cejudo C	FA33	Dávila J	FA13	Favier GI	MI18, MI19, MI20,
Centorbi HJ	MI9, MI12	De Giovanini C	EB64, EB70	Favier LS	FT57
Ceriani MF	S2-2	de Jong L	MI50, MI26	Fenucci JL	FA9
Cerón M	MI8	de la Vega M	RE14	Ferder IC1	RE12
Cervio A	NQ3	De los Ríos C	S4-2, EB121	Ferder L	FT8
Cesari A	Sat 1-1	De Luca N	EB46, EB47	Feresin GE	FT51, FT52, FT53, FT54, FT55
Cetica P	RE32, RE34, RE39, RE48	De Pauw M	FT15, FT37	Fernández AO	EB33
Cevallos C	FA34	de Rosas I	BV29	Fernández Baldo M	BM10
Chaban R	FA26	Deanna R	BV10	Fernández Belmonte C	EB34
Chacón G	FT34	Debattista NB	MI3, MI4	Fernandez E	CL11, PV6
Chain FE	AH24	Degarbo SM	BM17, MI51	Fernandez EM	EB46, EB47, PV13
Chambó J	BM35	Del Vitto L	BV3, BV36, AH6, AH7, AH8, EB11, FT48	Fernández F	FT17
Chamut S	BD4	Delgadin T	EM18	Fernandez G	MI60, BM10
Chanique A	RE6, RE7, RE8	Delgado Marín L	BF7	Fernández L	EB64, FT46, FT9
Charreau E	RE12	Delgado SM	RE11	Fernández M	EB124
Chaves EM	AH9, AH17	Della Gaspera ME	L13	Fernández R	MI26, MI50
Chediack JG	FA17, FA18, FA19	Della Vedova1 MC	EM24	Fernández S	RE40
Chiapella J	EB62	Delocca M	CL10	Fernandez SS	BV19
Chiaranello A	MI22	Delsouc MB	RE11	Ferramola M	FT31, BM13
Cid FD	FA17, FA18, FA19	Demaestri M	EB30	Ferrari M	MI13
Cid-Barría JL	Sat 1-3	Demaldé S	FT34	Ferrari SG	MI37
Cid-Barría M	Sat 1-3	DePauw C	FT18	Ferré D	FA31
Cifuentes D	MI23, FT30	Depetris Chauvin A	S2-2	Ferreira A	BV30, BV31
Cioca DR	BM16	Di Fonzo C	FA11	Ferreira V	BV30, BV31
Ciocco NF	EB28	Di Genaro MS	MI39	Ferrero AA	BM15, EB53, PV11, PV12
Cioffi G	FT15, FT18	Di Genaro S	FT30, MI21	Ferretti E	RE39
Ciuffo GM	EB29, FT22	Di Palma MA	BT6	Ferretti VA	BD3
Ciuffo LEC	EB29	Di Pilla MR	NQ15, NQ16	Fidelio G	BV13
Civit E	CL4	Di Renzo MA	BV9, BV25	Figueroa MF	RE22
Cobos E	MI8	Di Salvi N	CL3, CL8	Filippa V	FA17
Cóceres VM	MI24	Dias I	PV16	Filippuzzi S	EM24
Cocito L	BD10	Díaz AC	FA9, FA10	Fingermann G	CL10
Cohen A C	BV40	Díaz AO	AH18		

- Fiol de Cuneo M RE9, EM6, FA26, RE29
 Fitt M NQ12, NQ13, NQ20
 Florida RA MI48
 Fogal T AH17
 Fonseca AM EB128
 Forneris M RE21, RE22
 Fornés M W Sat 1-3, EB99, RE24
 Fortuna AM S3-3
 Frattari A BT23
 Frigerio C MI40
 Frola I AH10, MI13, MI14
 Fruttero L FA23, RE50
 Fuchs DV EB114
 Fuentes L FT22, CL6
 Funes MB EB94
 Funes SC FA17
 Fusco M FT13, FT43
- G**
 Gagliardino JJ C4
 Gago FE OC13
 Gaido N BT13
 Galanti L EB125
 Galetto L EB62
 Galiana D NQ12
 Galoppo GH BD10
 Galvani G EB122, EB64
 Galvez MJ BM23
 Gambero L MI40
 Garavaglia S BV21
 Garbero M BV18
 García Aseff S FT5
 García DA BF7
 García IM FT6, FT7, FT8
 García LN IM3
 García M BM11,
 García M FA3, FA33
 García M MI51
 García Valdez MV BD4
 Gargiulo CN EB103
 Gargiulo PA NQ3, NQ17, NQ18
 Garraza M FT15, FT18
 Garraza MH RE21
 Garraza R BT26
 Garro MF FA32, FA33
 Gatica A FT56, FT57
 Gatica N BT24
 Gauna G OC13
 Gauna H FA6, FA7, FA8, FA3
 Gaurón C MI21
 Gayol MC BF4, BF5
 Gentile N FT45
 Gentile T RE32
 Gette M BV36, AH6, FT48
 Ghietto LM MI58
 Ghini A NQ1, NQ5
 Ghirardi R EB110
 Giannoni SM S4-2, S6-3, EB61, EB74, EB75
 Gibbons A S4-1
 Gigola G OC5
 Gil de Pertierra AA EB67
 Gil L FT5
 Gil Lorenzo AF BM19, BM30, CL11, NQ10
 Gil M BV20
 Giménez MS BM13, BM26, EM21,
 FA34, FT31, FT49, FT50
- Girardi VN BV9, BV25
 Giraud-Billoud M FA29
 Giraud J AH10
 Giuliano S RE52
 Gleiser RM EB126, EB127
 Godoy MS FA21
 Golombek DA S2-1
 Gómez Barroso JÁ BM33, BM36
 Gomez MA BM34, BM35
 Gomez ML BT4
- Gonzalez A RE36
 Gonzalez B RE15
 González Cortéz M MI50
 González E EB64, EB122
 González I FT33
 González II EM24
 González Jatuff A FA13
 González LE MI48
 González M FT52
 González N BD6
 González P BF12
 González PS BT28
 Gonzalez RP MI51
 Gorla N FA31, FT45
 Gorustovich A BT29
 Gouiric S BT16
 Grassi E BV30, BV31, BV32
 Grau A AH24
 Grenat PR EB15, EB19, EB20, EB24
 Grilli D BM17, MI8
 Grosso C RE47
 Grosso M BM32, BT20, MI68, RE46
 Guagliardo S EB25, EB37
 Guardia T FT25
 Guerra GB MI52
 Guerra RA IM6
 Guevara A EB44
 Guevara E PV7, PV8, PV9
 Guevara M NQ17, NQ18
 Guiñazú LB PV18
 Gutiérrez MH BV18
 Gutiérrez MM BM15, EB53
 Guyón NF FT41, FT42
- H**
 Haggie, E BM6
 Haro Durand L BT29
 Hasuoka R MI48
 Heredia R MI72
 Hermosilla PC RE33
 Hernández D EB124
 Hernández G BF4, BF5, BF6
 Herrera M EB64, EB90, EB93
 Herrero MF EB126
 Honoré SM BM13
 Huarte M BF6
 Hued AC EB125, FT42
 Huergo M BF12
 Huhtaniemi I RE15
 Huidobro C Sat 1-2
 Hynes V FA31
- I**
 Ibarra J OC13
 Ibarra ME NQ14
 Ighani M FT53, FT54
 Iglesias G RE24
 Inzirillo G BV20
 Irigoyen S CL10
 Irusta G OC3
 Isaguirre A MI18
 Isla MI S3-1
 Ituarte LME BF14
- J**
 Jacob A FT28, FT29
 Jácome N L S4-3
 Jahn G BD4, OC8, RE45
 Jiménez Baigorria L NQ18
 Juan HV MI23
 Juan V EB33
 Juárez AV BM6
 Junges CM FT11, FT12
 Junqueras MJ EB34
 Juri Ayub M BM37
- K**
 Katz D NQ3
 Kruse MS NQ15, NQ16
- L**
 Laciari A MI9, MI22, MI23
 Lahoz V S4-2, EB61
 Lajmanovich R S5-2, FT11, FT12
 Lambrese YS MI29
 Landa AI NQ18
 Lapadula WJ BM37
 Lapierre A MI48
 Lapyckyj L OC9, RE35
 Larregle E FA34
 Larreguy C EB14
 Larroudé M MI13
 Larrusse A BV21
 Lartigue C BT27
 Lascano HR BV10
 Laspina CA EB52, EB74
 Laspiur A EB66, EB71, EB72, EB81, EB83,
 EB91, EB100, EB101, EB102,
 EB105, EB116, EB117, EB118
- Lauret CM BV38, BV39
 Laurito SR NQ20
 Lavilla E O C3
 Lazarte Otero V MI18, MI19, MI20
 Lazarte V MI21
 Ledesma C EB39
 Ledesma CR PV13
 Leimgruber C IM3
 Lemes J PV3
 Leporati J BT24, BT27
 Lescano JN EB76
 Leuzzi M OC13
 Levy M EB110
 Leynaud GC EB76
 Leyria J RE50
 Libertun C EM5
 Licata L AH22
 Licera CM CL17
 Liendo A EB36
 Liffourrena AS BT8, MI16
 Lillo J Sat 1-2
 Lima B FT55
 Liv Nardello A MI51
 Lizarraga E FT17, FT20
 Lizarralde MS EB87
 Llaver AP FT57
 Llompert G AH22
 Llompert J AH22
 Lodillinsky C OC9
 Loidl CF NQ14
 Lopez Appiolaza C BM30
 López D BT12
 López D BV6
 López Fontana C OC8
 López GA MI72
 López HL EB114
 Lopez J EB110
 López JA EB89
 López L BA5
 Lopez L BM4, BM24
 Lopez LE FA28
 López Lucero VR BV19
 López ML BV38, BV39
 López P PV20
 López Rivilli MJ BV6
 López Soto EJ BM7
 Lorenzatti EA FT12
 Losinno A BM4, BM24
 Losinno L S1-3
 Loto F MI59, MI60, MI62
 Lozano E BM27
 Lucchesi G I S5-1, BT8, MI16, MI70, MI72

- Lucero C MI21
 Lucero Estrada C MI18, MI19, MI35
 Lucero López VR FT49, FT50
 Lucero R FT33, PV19
 Lucero RA PV20
 Lucifora LO Sat 2-4
 Luconi M FT9
 Ludueña Almeida F CL17
 Luján A MI50
 Luna L FT52, FT53, FT55
 Luna LC FT51
 Lupi G MI71
 Luque EH BD10, FT24, FT28, FT29
 Luque EM RE9
 Lux-Lantos V EM5
- M**
 Machado EE BV16, BM39
 Machevsky EJ BV3
 Ma?kovi? M BT29
 Madoery R BV13, BV14
 Maffrand C OC10
 Maggioni T EB125
 Magnano G AH10
 Maiocchi MG BV3
 Maldonado CA IM3
 Malossi E RE26
 Mañas F FT45
 Mancini M EB22, EB25, EB36, EB37, EB57
 Mancuso P CL10
 Manella Hoyos JL BV11
 Manes ME BD4
 Mangione A FT44, BV35
 Mangudo C EB127
 Manucha W FT6, FT7, FT8, FT40,
 Manzano H BV29
 Manzano J MI50
 Marchese N NQ19
 Marchessi J EB33
 Marchetti CS PV13
 María A FA32, FA33
 Mariani ME BV13
 Marín Briggiler CI RE35
 Marinero V EB64, EB121
 Márquez F FT33
 Marra C EM21
 Martín Arrieta G OC5
 Martínez AN EB94
 Martínez C AH22
 Martínez CF EB31
 Martínez L BV29
 Martínez NA EB96
 Martínez RA RE46
 Martínez V Sat 1-3
 Martini AC RE9
 Martino AL EB15, EB16, EB19, EB20,
 EB22, EB23, EB24
- Martori R EB4
 Marún A MI14
 Maselli ME OC8
 Mattana C MI23
 Mattar MA IM6
 Maturano YP Sat 3-2, BT17
 Mayocchi K BM34, BM35
 Mayorga L RE28
 Mayorga LS BM17
 Mazzei L FT6, FT7, FT8
 Mazzeo M CL10
 Medawar MV FT30
 Medina E MI59, MI60, MI62
 Medrano M MI34
 Meglioli PA EB7, EB44
 Melatini G OC5
 Méndez E EB6
 Mendoza G FT33
 Mengarelli G BF6
- Mercado MI AH24
 Mercado S EB113
 Meresman G RE36
 Meringer MV BV16
 Merkis C RE6, RE7, RE8
 Mestre MV BT17
 Miatello RM BM19
 Michaut MA BF14, RE28
 Micinquevich S BM34, BM35
 Milanesio ME BT4, BT5, BT6
 Milesi S BM33
 Minchiotti M BV13, BV14
 Miquelarena AM EB114
 Miragaya M S1-2, RE52
 Mirande JM Sat 2-3
 Mitjans N BT13, BT15
 Mock FM EB115
 Moglia MM EB128, EB68
 Mohamed F AH14, AH15, FA17, RE22
 Molina A BT20
 Molina Arias SM BV12
 Molina Marino LM BA5, FA28
 Molinero D RE46, MI68, BM32, BT20, RE47
 Moliva M RE6, RE7
 Monclus MA RE24
 Monesterolo NE CL16
 Monetta P BT16
 Monferrán M EB125, FT51, FT54
 Montagna DR RE48
 Montoro A BV37
 Mora SJ BT4, BT5
 Morado S RE32, RE48
 Morales DP EB115
 Moreno D EB64, EB70
 Moreno L MI58
 Moreno M BV31
 Moreno MT BV17
 Moreno VM EB77, EB81
 Moretti H BV32
 Morosano G CL7
 Morra G EB25, EB36, EB37
 Motta A RE14
 Motta C BM32, BT20, FT24, MI68,
 RE46, RE47
- Muñoz del Toro M FT28, FT29, BD10
 Muñoz EM S2-3, NQ12
 Muñoz MA Sat 3-4, MI56, MI57
 Muñoz NB BV10
 Muraro N S2-2
 Murray AP PV12
 Mutto A S1-1
 Muzzio N BF12
- N**
 Nader-Macias F AH10
 Nadin SB BM16, OC13
 Najle R BD6
 Nally MC Sat 3-2, Sat 3-4, MI52, MI56, MI57
 Nanfaro F FA33
 Navarta G MI27, MI38
 Navas A EB64, EB90, EB93
 Negro A FT11
 Niebyski A FA3, FA6, FA7, FA8
 Nieva RA EB83, EB100, EB101,
 Nievas RP EB68
 Nigra A CL16
 Nuño G S3-1
- O**
 Ochoa AL BT6
 Odierno L MI14
 Odoñez RM S3-1
 Ojeda M FT33
 Ojeda MS EM24
 Ojeda RA S6-1
 Olivares B FT5
- Olivares C RE36
 Oliveros L BM26, RE21, RE22,
 Oliveros LB EM21
 Oliveros LB FT31
 Olivieri G PV15, PV16
 Ontañón OM BT28
 Orta FJ PV20
 Ortega HH FT22
 Ortiz Maldonado V NQ13
 Ortiz SG EB52
 Ortolan T BV9, BV25
 Ortuño MN EB75
 Ortuño N S4-2
 Ostinelli CA BF15
- P**
 Paccapelo H BV30
 Padrón JM FT30
 Padrones N FA18, FA19
 Paez S MI8
 Paglini MG MI58
 Paisio CE BT28
 Palchetti MV MI58
 Palermo JA BM27
 Palomo V FT40
 Panigoni MR PV18
 Panini A FT15, FT18, FT37
 Pappano NB MI3, MI4
 Paracampo A EB87
 Parborell F RE3, RE4, RE12
 Paredes J FT13
 Pareja V MI26, MI50
 Parera CA BV33
 Parma MJ FT27
 Paroldi HE MI59, MI60, MI62
 Pascuali N RE3, RE4
 Pasquale M BF12
 Pastor A MI49
 Pastor P EB64, EB70
 Patterson SI NQ20
 Patti G FT34
 Pavarotti M RE28
 Paviolo NS BM31
 Paz MC NQ19
 Pazos C OC3, RE3
 Pedernera AM FT25
 Pedranzani HE BV11, BV12, BV18
 Pellegrino M MI13, MI14
 Pellerano RG BV3
 Peltzer PM FT11, FT12
 Pelzer L FA32, FT5, FT13, FT14, FT25
 Peña E NQ14
 Pennacchio GE RE45
 Peralta L FT45
 Peralta M BT20
 Pereira C BM33
 Peretti S CL16
 Pérez Díaz MF FT31
 Pérez Díaz J P BT25
 Pérez GA PV18
 Perez JE OC5
 Pérez Sirkin D EM18
 Perillo MA MI71
 Perotti M FT17, FT20
 Perroud HA CL7
 Pesce VM Sat 3-4, MI52, MI56, MI57,
 Petenatti E BV36
 Petenatti EM AH6, AH7, AH8, FT48,
 BV3, EB11
 Petenatti M BV36
 Petenatti ME EB11, AH6, AH7, FT48
 Pezzotto SM CL7
 Phiel L BF6
 Piaggio LE EB71
 Piazza LA BV6
 Picco N BM32, BT20, MI68, RE46, RE47

- Piccoli P BV20
 Piccoli PN BV40
 Piedrabuena J EB103
 Piezzi R AH17
 Piola H BV15
 Pires DA EB126
 Pirola CJ C5
 Pittaro MG PV4
 Pittau R BM6
 Pizarro M CL13
 Pizarro MA MI51
 Plaul SE AH18
 Pollo FE EB22
 Ponce C FA33
 Ponce M BV29
 Ponnasa G AH24
 Pons P BM6, RE49
 Ponzio MF EM6, RE29
 Popovich M BV36, FT48, AH6
 Porcel R BV18
 Poretti MB FA26
 Poutamen M RE15
 Prat MI BM23
 Presello DA BV25
 Previtali G CL16
 Puebla M FA3
- Q**
 Quero M FA31
 Quiran EM EB12, EB115
 Quiroga AM BV11
 Quiroga ED BT5
 Quiroga L EB64, EB70, EB90, EB99, EB110, EB121
 Quiroga LV AH7
 Quiroga M PV19
- R**
 Rabinovich GA RE36
 Racagni GE BV16
 Radicetti DS Sat 3-4, MI56, MI57
 Ramos GC CL13, MI64
 Rasia M BF6
 Rask-Andersen M FA26
 Rasmussen J NQ12, NQ20
 Raspanti C MI13
 Rastrilla AM RE11, RE14, RE17
 Ratner LD RE15
 Ray AM MI37
 Razzeto GS FT49, FT50
 Reboreda JC C2
 Redondo A BM11, BM40
 Redondo E OC10
 Regueira E AH23
 Reiner G BF7
 Reinoso H BV17
 Remes Lenicov M Sat 2-1
 Reta GF FT56
 Reta Moyano A RE21
 Reus L S6-3
 Reus ML EB52, EB74, EB75
 Rey M NQ1, NQ5, NQ16
 Rey-Funes M NQ14
 Rezza I BT13, BT15
 Ricci A RE36
 Rigatuso R BT12
 Ríos ML CL4
 Ríos-Luci C FT30
 Rivelli JF CL16
 Robbio G FA13
 Rodríguez Assaf LA Sat 3-2, BT17
 Rodríguez C AH3
 Rodríguez C EB36, EB39, EB55
 Rodríguez Echandía E FA13
 Rodríguez J NQ18
 Rodríguez N EB11
- Rodríguez PC RE30
 Rodríguez PE MI58
 Rodríguez Rivera M BV12
 Roggio MA FT41, FT42
 Roig FA EB31
 Rojas E PV20
 Rojas JÁ BV17
 Rolando A RE8
 Romanini M RE47
 Romero A Sat 1-3
 Romero M BT29
 Romero MC BV17
 Romero N EB96
 Ronchi G MI6, MI7, MI48
 Ronchi GD FA18, FA19
 Roquer S FT16, FT32
 Rosa GA FA11
 Rosas SB PV18
 Rossi A FT27
 Rosso M OC9
 Rossomando P MI22
 Rotelli ME FT25
 Rubin de Celis E BF6
 Rubiolo E FA23, RE49, RE50
 Rueda EC EB87
 Ruiz AM CL11, NQ10
 Ruiz Lozano JM BV11, BV12, BV18
 Ruiz M BF4, BF5, BF6
 Ruiz M BV33, BV37, PV15, PV16
 Ruiz RD EM6, RE5, RE9, RE29
 Rulli SB RE15
 Ruttler ME CL13, MI51
- S**
 Saad JR FA32, FA33
 Sáez Lancellotti TE Sat 1-3, RE24
 Sagua MD CL4, CL13
 Sahrawy M BV17
 Saidman E BV36
 Salas NE EB15, EB16, EB19, EB20, EB22, EB23, EB24
 Saldeña E FA31
 Saldeña TA BF14
 Salinas A FT44
 Salinas AG FT13, MI18, MI34
 Salinas E BM10
 Salinas V EB25, EB36, EB37, EB57
 Salinas ZA EB23
 Salinas, AG MI35
 Salinero MC EB24
 Salomon MV BV20
 Salvadores C BV40
 Sanabria E EB64, EB93, EB99, EB110, EB121
 Sanabria EA BF8, EB122, EB70
 Sanchez JM MI71
 Sanchez S FT22
 Sánchez SS BM13
 Sánchez-Borzone M BF7
 Sánchez-Puerta MV BM37
 Sanchis E RE6, RE7, RE8
 Sansone G BM10
 Sansone MG MI29
 Santa Cruz R RE52
 Santamaría C FT28, FT29
 Santander VS CL16
 Santillán ME RE5
 Sanz Ferramola M BM10, MI27, MI38
 Sanz MI MI29
 Saraví FD BF14
 Sartor T FA13, NQ10
 Sassi PL S6-2
 Sasso CV OC8
 Sassone A AH23
 Satorre MM RE41
 Satorres S MI6, MI7, MI22
 Savastano LE NQ13, NQ20
- Sawant R FA26
 Scaia F AH23
 Scaia MF FA20
 Scappini EG EB26, EB113
 Scarabotti P EB64
 Scardapane L AH9, AH17
 Schiöth H FA26
 Schleef N OC10
 Schnittger L MI8
 Scopel AL BV6
 Scoppa G FA7, FA8
 Scoppa H FA3
 Scotti L OC3, RE3, RE4
 Segal-Eiras A BD3
 Segatto R CL10
 Seltzer AM NQ13
 Selvlever G NQ3
 Semino S OC8
 Sereno C NQ3
 Serra E BM36
 Serrano L NQ3, NQ17, NQ18
 Serrato AJ BV17
 Settembrini B RE49
 Siewert S FT33, EM24
 Silva HJ MI34, MI36, MI37
 Silva MP BV6
 Silva PG MI36, MI37
 Silva RL MI51
 Singla J RE36
 Slanis A FT52
 Smitz J RE32
 Soaje, M RE45
 Somma C PV6, PV13
 Soria J MI19
 Sosa A FT13, FT43
 Sosa MA BM27, FA13, NQ10, RE26, RE27,
 Sosa ME FT56
 Sosa Paredes E AH15
 Sosa VL AH8
 Sosa Z RE17
 Sottile ML BM16
 Spallek S BT29
 Spiecker E BT29
 Stadler T PV11
 Stagnitta P BT13, BT15, MI66
 Stagnoli S FA6
 Stariolo R FA23
 Stefanazzi N BM15, PV11, PV12
 Stoker C BD10
 Stutz G RE5
- T**
 Taleisnik E PV4
 Talia J FT15, FT18
 Talia JM MI3, MI4
 Talio M FT9, FT46
 Tamiozzo LV PV7, PV8, PV9
 Tanevitch A AH22
 Tanzola D EB25, EB37
 Tapia A FT51, FT52, FT53, FT54, FT55
 Taricco E EB36
 Tavecchio NE BV18
 Telechea A BM17, CL4, MI64
 Tellado MN RE34
 Tereschuk ML FT52
 Tesone M OC3, RE3, RE4, RE12
 Teves M FT14, FT15, FT18
 Teves MR AH6, FT48
 Thevenon MA FT47
 Tinnirello B NQ18
 Tione ML EB13
 Tolosa de Talamoni N BA3
 Tomás A PV4
 Tonn C FT56, FT57, MI3, MI4, MI23
 Toro ME Sat 3-2, Sat 3-4, BT17, MI56, MI57, MI52, MI59, MI60, MI62

Torres A	BM6	Vega AE	FT13, MI34, MI35, MI66	Vincenti LM	RE9
Torres Basso MB	EB29	Vega Avila AD	MI59, MI60, MI62	Violante G	PV6
Torres C	FA3, FA6, FA7	Vega I	BM27	Violante MG	PV13
Tosti SB	CL3, CL8	Vega IA	AH3, AH15, FA21, FA29	Visconti M	BF4, BF5, BF6
Tourn MG	BV6	Vega M	FT46	Vissio P	EM18
Travaglia C	BV17	Vega MC	NQ15, NQ16	Vita F	PV15, PV16
Troncoso N	BT15	Vega V	BM26	Vivas A	BM32, BT20, MI68, RE47
Trotteyn MJ	EB42	Veiga MF	RE35	Vivas B	RE46
Trujillo L	CL6	Velazquez G	MI16	Volonteri C	AH23
Turchetti J	PV15	Velázquez L	MI18, MI19, MI20, MI21	Volonteri MC	RE38
Turco MD	BV6	Veleiro AS	NQ1, NQ5		
Turiello P	RE8	Velurtas SM	FA9	W	
Tuseddu M	FT34	Vendramín M	FT16, FT32	Wendel G	CL6, FT5, FT13, FT14
		Vera Mesones R	BT29	Werdin González J	EB53
U		Verdes P	BT23, BT24, BT25, BT26, BT27	Werdin J	BM15
Ulloa A	MI64	Verdugo R	CL6	Wunderlin D A	Sat 3-1, FT41, FT51, FT53, FT54
Uñates MA	BV19	Verga EG	EB76		
Uribe Echevarria E	IM3	Viale S	EB30		
Usorach JI	BV16	Viale SN	PV7, PV8, PV9	Z	
Usorach MN	BM39	Victorica AE	EB71, EB116	Zago MP	BT29
		Vidal Bravo M	BM31	Zalazar C	FT11
V		Vidal E	FT11	Zambrano K	FT9
Valdez SR	RE45	Vidal EA	FT52	Zampini IC	S3-1
Valenzuela R	Sat 1-2	Vidal M	EB121	Zanetti MN	BF14
Valcaneras S	RE14	Viera TB	BF14	Zanetti N	RE28
Vallejo M	BT16, BT17	Vigezzi L	FT24	Zapata MJ	BT17
Vallés P	BM19, BM30	Vignatti A	EB55, EB56, EB57, EB58	Zappala C	FA11
Vaquero F	EB9	Vilches JY	EB12	Zárate JM	BM37
Vargas GE	BT29	Villa MC	IM6	Zayas MA	BD10
Vargas L	BV14	Villagra PE	EB7, EB44		Zelarayán LI
Vargas-Roig LM	BM16, OC13,	Villamil CB	BM23	FA12	
Vazquez F	Sat 3-2, Sat 3-4, BT17, MI52, MI56, MI57, MI59, MI60, MI62	Villar J	FT34	Zubiria MG	OC8
Vazquez M	BM36	Villarrubia O	EB30	Zuliani M	EB93
Vazquez ML	EB128	Villavicencio HJ	EB66, EB77	Zunino MP	BV38, BV39
Vazquez-Levin MH	OC9, RE35	Villavicencio R	CL7	Zygodlo J	Sat 3-3, BV38, BV39
		Vincenti A	RE24		

POSTER PRESENTATIONS INDEX

	Abstract N°
AH ANATOMY AND HISTOLOGY	1 - 14
BA ANIMAL BIUCHEMISTRY	15 - 17
BD DEVELOPMENTAL BIOLOGY	18 - 21
BF BIOPHYSICS	22 - 29
BM GENERAL, MOLECULAR AND CELLULAR BIOLOGY	30 - 54
BT BIOTECHNOLOGY	55 - 71
BV PLANT BIOCHEMISTRY, PHYSIOLOGY, PATHOLOGY AND GENETICS	72 - 99
CL HUMAN, VETERINARY AND ODONTOLOGIC CLINICAL INVESTIGATION	100 - 109
EB ECOLOGY, ETOLOGY AND BIODIVERSITY	110 - 191
EM ENDOCRINOLOGY AND METABOLISM	192 - 196
FA ANIMAL PHYSIOLOGY	197 - 218
FT PHARMACOLOGY AND TOXICOLOGY	219 - 262
IM IMMUNOLOGY	263 - 264
MI MICROBIOLOGY	265 - 309
NQ NEUROCHEMISTRY-NEUROSCIENCES	310 - 322
OC ONCOLOGY	323 - 328
PV VEGETAL PRODUCTION	329 - 342
RE REPRODUCTION	343 - 379

Erratum from XXVIII Annual Scientific Meeting, Cuyo Biology Society (Sociedad de Biología de Cuyo)

1.

THE TRANSPORT OF CATHEPSIN D IN RAT EPIDIDYMAL CELLS

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The epididymal epithelium secretes proteins into the lumen, some of which are acid hydrolases (such as cathepsin D). In other cell types, hydrolytic enzymes are transported to lysosomes mediated by receptors for mannose-6-phosphate, the cation-dependent and independent ones (CI-CD and MPRS). In some cellular models, procathepsin D (PCD) is complexed with prosaposin (SAP), which are transported to lysosomes or secreted by sortilin (Sort). Here, we have attempted to identify the transport pathway for PCD (CD-MPR and / or Sort) in epididymal cells and how it could be regulated by steroid hormones. We evaluated the expression and secretion of PCD and SAP, and the expression of CD-MPR and Sort in RCE-1 line cells (rat epididymis), subjected to hormone treatments (estradiol or tamoxifen), and in the presence or absence of NH₄Cl. The proteins under study were detected by immunoblot from cells or culture medium. Estradiol induced an increase in the expression and secretion of PCD, but no changes in the expression of Sort or the CD-MPR. In turn, treatment with NH₄Cl decreased the secretion of PCD, with greater intracellular retention, accompanied by an increase in the expression of Sort and CD-MPR. These preliminary results suggest that PCD is not secreted by "default" in the presence of estradiol and that an alternative route of transport, mediated by Sort and regulated by estrogen may be involved.

2.

TRYPANOCIDAL EFFECT OF TERPENOID ACIDS AND DERIVATIVES OF BILE ACIDS OBTAINED FROM PLANTS

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Trypanosoma cruzi is the causative agent of Chagas disease. When cultured, this parasite cycles between epimastigote form and a low percentage is transformed to the infective form trypomastigote. Recently, we have found that some sesquiterpene lactones (STLS) and hydroxylated diterpene quinones obtained from native plants affect the growth of epimastigotes at very low concentrations and with low toxicity on mammalian cells. This work was focused on evaluating the effect of other families of natural compounds on *T. cruzi*, to assess which were active against the parasite. Epimastigotes (Dm28C strain) were cultured in liquid media in the presence or absence of different concentrations of the compounds (2-10 µg/ml). Among the compounds tested, quinones coupled with steroids and terpenoids showed some antiproliferative effects on parasites. We have shown that seco-chiliolid acid, its derivative secochiliolidquinone and, to a lesser extent, secochiliolidhydroquinone and lithocholic acid exert an important antiproliferative effect on epimastigotes. It remains to determine the IC₅₀ values of these compounds and to evaluate the cytotoxicity on mammalian cells, in addition to the identification of molecular targets for the action of these molecules.

3.

USE OF PSYCHOTROPIC IN MENDOZA

Grilli S, Montiel A, Kaiser L, Cannutt B, Lentini A.

This project involves the study of drug utilization in private pharmacies in Mendoza, through a retrospective analysis performed in different branches of "Del Centro" Pharmacy chain. Students regularly attended the branches. Data of each branch were transcribed to a form. Importantly, 15554 prescriptions were processed manually. All branches, irrespective of geographical area, there is an alprazolam consumption that exceeds the reference dose. The collected data allowed us to see that the female sex has the highest prevalence of use of psychotropic drugs. The age range in which they consume more psychotropic drugs ranging from 40-70 years. Most recipes containing psychotropic drugs are prescribed by doctors who are not psychiatrists or neurologists specialty and in many cases, they are not clinicians. The collected data have identified different types of failures such as the existence of many prescriptions does not match with the diagnosis. We have identified patients that consumption of certain psychiatric drugs exceeds normal doses because in the same month they have several recipes prescribe with the same drugs by the same medical doctor. Certain prescriptions of psychotropic drugs do not match the diagnosis. There are patients who abuse of certain drugs because we found recipes with the same active ingredients, prescribed by different medical professionals, thus demonstrating that consume two or three times what is recommended according to pathology.

4.

DISTRIBUTION OF CD-MPR AND PROSAPOSIN IN THE EPIDIDYMIS OF SORTILIN-KNOCKOUT MICE. POSSIBLE HORMONAL REGULATION

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The epididymis is involved in sperm maturation through the secretion of factors into the lumen, and its integrity and functionality depend on steroid hormones. Epididymal secretion is enriched in acid hydrolases. In most cell types, these enzymes have a residue mannose-6-phosphate (M6P) to be transported to lysosomes via M6P receptors (CI-MPR or CD-MPR). Other lysosomal proteins (such as prosaposin, PSAP) are transported (complexed with procathepsin D, PCD) through the sortilin receptor. This study set out to determine whether PCD is transported and secreted through the CD-MPR, sortilin or both pathways in mouse epididymis, and if these mechanisms are altered by hormone deprivation. We used transgenic sortilin-knockout mice (SKO) and evaluated the effect of castration on the expression and distribution of proteins in epididymis. After 48 hours, the epididymides were removed, fixed and processed for immunohistochemistry. Besides morphological changes, castration induced changes in the expression and distribution of proteins in the epididymal epithelium. We observed increased immunoreactivity of CD-MPR and PSAP in SKO mice, indicating a likely compensatory mechanism to the absence of sortilin. Furthermore, immunoreactivity for CD-MPR and PSAP increased due to castration in both controls and SKO, and there was a redistribution of CD-MPR to apical areas in the epithelium. We conclude that CD-MPR and sortilin are alternative routes for some lysosomal proteins in the epididymis.