SECOND JOINT MEETING
OF THE BIOLOGY SOCIETIES
FROM ARGENTINA
(Segunda Reunión Conjunta de Sociedades de Biología de la República Argentina)

XIII Multidisciplinary Workshop, Argentine Society of Biology
(Sociedad Argentina de Biología)

XXIX Annual Scientific Meeting, Cuyo Biology Society
(Sociedad de Biología de Cuyo)

XVIII Scientific Workshop, Córdoba Biology Society
(Sociedad de Biología de Córdoba)

Abstracts

August 17-19, 2011
San Juan, Argentina

The abstracts were evaluated by a scientific committee prior to publication
C1. DECIPHERING THE ORIGIN, EVOLUTION AND FUNCTION OF EPITHALAMIC ASYMMETRIES
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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
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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
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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 therapeutical strategy to maintain the effective beta cell mass in order to obtain an adequate diabetes control.

C5.
GENETICS AND EPGENETICS OF THE METABOLIC SYNDROME
Pirola CJ

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 polychistosis 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 PPARγ, 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 TNFα, 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, PPARα and PPARγ, already targets for approved drugs may suggest new avenues for MS pharmacological treatment.
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.

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 luteolysis 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 vitro 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 (busceral 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-ETM) 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.

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.
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 nematode, 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
Depetris Chauvin A, Berni J, Aranovich EJ, Muraro N, Beckwith EJ, Ceriani MF.

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 s) are clearly crucial. The preservation of molecular oscillations within the sLN s is key to command rhythmic behavior under free running conditions. The s-LNvs 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 s (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 s 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 promising; 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.
BIOLGICAL PROPERTIES OF MEDICINAL PLANTS FROM HIGH ALTITUDE ECOSYSTEM AND POTENTIALS USES IN THE PHARMACEUTICAL INDUSTRY
Zampini IC, Cueto AS, Nuño G, Odoñez RM, Alberto MR, Isla MI.

Plants from arid and semi-arid 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 bolivien sis, Chuquiraga atacamensis, Parastrephia lucida* (Asteraceae family) and *Fabiana punensis, F. bryoides, F. densa* and *F. patagonica* (Solanaeace 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 pour 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 acto on the cellular cycle. In this regard, active vegetal compounds have demonstrated potent control of cell proliferation. The utilization of ovocite 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 cytoplasmatic compound, maturation promoter factor (MPF). Our group has demonstrated that DhL and its hydrogenated derivative, in which the alpha-methylene lactone (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 permitsmeiosis 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 attributes 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, in *Tricitum 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.
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
Giannoni SMA,2, Beninato V1, Ortúñio N1, Adarvez S, Cappa F1,2, de los Ríos C1, Lahoz V1,2, Campos V1,2, Andino N1, Borghi C1,2.
1Grupo INTERBIODES, Departamento de Biología y Museo, UNSJ. 2CONICET.

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 Biocenmic 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.

S4-3.
ANDEAN CONDOR CONSERVATION PROGRAM: JOINING SCIENCE WITH ANCESTRAL TRADITIONS
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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.

Jardín Zoológico de la Ciudad de Buenos Aires & Fundación Bioandina Argentina.

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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.

Jardín Zoológico de la Ciudad de Buenos Aires & Fundación Bioandina Argentina.
**ECOLOGY AND ENVIRONMENT**

**SS-1. STRATEGIES OF P. PUTIDA TO COUNTERACT THE EFFECT OF TOXIC IONS AND UNDESIRABLE COMPOUNDS FROM CATIONICS DETERGENTS DEGRADATION**

Lucchesi GI.

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-monoxygenase activity resulting in the formation of tetradecylalkanal and trimethylamine (TMA). The TMA is metabolized to NH₃ through oxidation and demethylation, and also accumulated inside the cell, affecting the bacterial growth, an effect counteracted by the addition of AlCl₃, Al³⁺ acted as a Lewis’ acid, playing a role in the control of TMA intracellular levels by the formation of Al³⁺: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 phosphatidylylglycerol to neutralize the positive charge of TTAB. In the presence of AlCl₃, an increase in phosphatidylethanolamine (PE) was detected, which led us to propose a physiological role for PC as a temporary reservoir for Al³⁺ through the formation of Al³⁺:PC complexes. We have cloned and sequenced a gene in this strain that encodes a phosphatidylethanolamine 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 Al³⁺ supporting that the PC acts as a temporary reservoir for available Al³⁺ through the formation of Al³⁺:PC complexes. These complexes are utilized as an Al³⁺ 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.

**SS-2. ECOLOGICAL RISK OF AMPHIBIANS IN AGROECOSYSTEMS OF CENTRAL-EASTERN ARGENTINA**

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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.

**SS-3. MINERAL BIOLEACHING AND HEAVY METALS BIOREMEDIATION**

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Metals can be extracted from ores through the microbial action on the sulfides oxidizing them to sulfate and releasing the metals. This process called bioreaching 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 bioreaching 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 develop 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.
DESSERT: “ALTERNATIVES ADAPTED TO LIVE IN HIM AND THREATS TO THEIR CONSERVATION”

S6-1.

SELECTION PRESSURES, CONVERGENCE AND COEXISTENCE OF DESERT SMALL MAMMALS

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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.

Parcialmente financiado por PIP- CONICET 2884, 5944, 0325, y PICT-AGENCIA 11768, 0455.

S6-2.

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

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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.

Study funded by CONICET, Agencia-Secyt and FONDAP.

S6-3.

DISTURBANCES IN THE DESERT AND THREATS TO CONSERVATION

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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
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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

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 orchietomy 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.

Funding: FONDECYT Project 1100183.

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.
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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
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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
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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 and 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
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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.

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.
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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, Gualeguaychú 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, Gualeguaychú 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.
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 control yeast fermentation processes 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 vinacea BHv438 and Torulaspora 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.

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 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.
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 which forms all of the floor and part of the roof of 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)
Gette MA, Petenatti ME, Popovich MC, Teves MR, Del Vitto LA, Petenatti EM.
Herbarium/Proj. 22/Q-016 SPU-ME, UNSL. Ej. de los Andes 950, 5700 San Luis, Argentina. E-mail: mgette@unsl.edu.ar

Schkuhria pinnata (Lam.) Kuntze ex Thell., commonly known as “canchalagua” or “matapulgas”, is a South American annual herb used in popular medicine as a deparative, antiseptic, for slimming, and as insecticide against fleas and lices. It has shown antimarial 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 micrographic 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, filiarial dotted-glandular. Flowers pistillate 1, bilabiate, and perfect 5-6, wide blade. Cipselae pyramidal, papus 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; Sls =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 megapotamicum, “TÉ PAMPA” (ASTERACEAE)
Quiroga LV, Petenatti ME, Petenatti EM, Del Vitto LA.
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Thelesperma megapotamicum (Spr. Kuntze “té pampa” or “té indio”, Asteraceae:Coreopsisae) is a perennial herb with bicentric American distribution, native to Central and Southern Argentina and Uruguay. It grows from Santiago del Estero to Rio 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 fistulose. 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)
Sosa VL, Petenatti EM, Del Vitto LA.
Herbarium UNSL/ Proj. 22-Q-616 SPU-ME, Ej. de los Andes 950, D5700HHW San Luis, Argentina. E-mail: elipete@unsl.edu.ar

Gochnatia glutinosa (D. Don) Hook. & Arn. (”jarillilla”, “jarilla pispia”) 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 amastomatous (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 IMMUNOHISTOCHEMICAL DESCRIPTION OF TESTIS VISCACHA (Lagostomus maximus maximus) IN RELATION TO AGE**

Cruceño A, Aguilera Merlo C, Chaves EM, Domínguez S, Scardapane L.

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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 keys 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 constitute of numerous primitive germ cells and Sertoli cells. The diameter of the seminiferous tubule in immature animals was 59.7 ± 0.75 μm, achieving a value of 137.6 ± 1.67 μm in adult animals. The RA in immature animals are mainly in germ cells, whereas in adult animals are found in the Sertoli around 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**

Frola I1, Magnano G2, Giraudo J3, Espeche C3, Nader-Macias F1, Bogni C1.

1Dpto Microbiol and 2Dpto Animal Patol UNRC, 3Dpto Prevent Microbiol CERELA-CONICET. E-mail: cbogni@exa.unrc.edu.ar

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 produced inflammation, three quarters were inoculated (IQ) with 1ml of 107, 108 and 109 CFU/ml and one quarter as control. Other animal was used for histological analysis and was IQ with 108 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 108 CFU/ml did not produce clinical signs in the udder or changes in milk. The SCC increased up to 48th 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.

<table>
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<th>Pars Distalis</th>
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<th>Pars Intermedia</th>
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<tbody>
<tr>
<td></td>
<td>Control</td>
<td>Melatonin</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>1.08 ± 0.11</td>
<td>0.65 ± 0.05</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>0.73 ± 0.06</td>
<td>0.47 ± 0.05</td>
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<tr>
<td>3</td>
<td>0.34 ± 0.04</td>
<td>0.20 ± 0.03</td>
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<tr>
<td>4</td>
<td>0.41 ± 0.02</td>
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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 - INMUNOHISTOCHEMICAL CHARACTERIZATION OF THE INTERSTITIAL CELLS PRESENT IN THE PINEAL GLAND OF Lagostomus maximus maximus**

Sosa Paredes E1, Acosta M1, Vega IA2, Mohamed F1.

1Cát. de Histología. Proy. 22/Q003. UNSanLuis. 2Lab. de Fisiología (IHED-CONICET). UNCy.

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 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 (Lm.n/m) IN RELATION TO REPRODUCTIVE CYCLE AND AGE
Proy 22/Q003, Cátedra de Histología. UNSL.
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The objective of this work is to study morphological variations seasonal in seminal vesicles (SV) of vizcacha and describe the SV in immature males. The vizcacha is a seasonal reproduction rodent, with maximal activity in summer and minimal in winter. SV of adult and immature male vizcachas 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 nuclei, 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 immature male viscachas. 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 apismatic 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.

10. AH18 - VARIATIONS IN THE EXPRESSION OF CARBOHYDRATES IN THE INTESTINE OF Corydoras paleatus JENYS, 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, telesost of cosmopolitan distribution of South American, which use 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 hematoxylin 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*; KOH/PA*; PA/Bh/KOH/PAS; KOH/PA*/Bh/PAS; Alcian Blue and Toluidine Blue at different pH. 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
Facultad Odontología Universidad Nacional de La Plata UNLP.
E-mail: gracieladurso@yahoo.com.ar

We analyze the microstructure of enamel according to levels of increasing complexity: crystals, prisms, enamel, pattern and dentin. 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 apismatic 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.

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Depto Biodiversidad y Biologia Experimental FCEN-UBA.

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. Steroidogenics 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)
Chain FE, Mercado MF, Coll.Araoz MV, Catalan CAN, Grau A, Ponessa G.
1Inquinoa-Conicet; 2Inst de Ecologia Regional, Fac de Cs Nat e Inst Miguel Lillo, UNR; 3Inst de Morfologia Vegetal, Fund Miguel Lillo. E-mail: fernandochain@hotmail.com

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 differentiation periderm 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
Brun A, Caviedes-Vidal E.
IMIBIO-SL, CONICET – UNSL.

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 develop by Campbell et al. 2008. The culture medium used, D-MEM supplemented with 2.5% FBS, insulin 10 µg/ml, transferrin 5.5 mg/ml, selenium 0.67 mg/ml, porcine mucosal heparine 50 µg/µl, penicillin 50 U/ml and streptomycin 50µg/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 that lasted up to 22 days. On the surface PET Grenier Bio-One, cell proliferation was apparent on PET Nunc surface. We obtained similar results were obtained according to the surface to be evaluated

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

Glutamine (GLN) is an aminoacid 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 2.5µmol 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 90min 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)
Molina Marino LM, López L, Caviedes Vidal E.
IMIBIO-SL, CONICET - UNSL. San Luis. Argentina. E-mail: lucianamolina.3@gmail.com

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 we 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.
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 transosomas, involved in early yolk deposition, were the most evident organelles. In yellow vitellogenic follicles ≥ 4 mm, there were yolk granules ruptured 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 of ≥ 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 regresion process characterized by cell death by apoptosis and necrosis, among others.

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It has been suggested that the number and complexity of carbohydrates 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.

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

Durando M, Cocito L, Zayas MA, Galoppo GH, Stoker C, Luque EH, Muñoz-de Toro M.
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Caiman latirostris exhibit temperature-dependent sex determination (TSD) and sexual reversion has also been demonstrated after 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 E, exposed eggs incubated at the male-producing temperature). Serum levels of E, 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 E, 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

Casco C², Visconti M¹, Ruiz M², Gayol M C², Hernández G¹, Bazzoni G², Bollini A³.

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 the initial rate (EA). 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

Ruiz M², Casco C², Visconti M¹, Gayol M del C², Bollini A³, Hernández G¹, Bazzoni G².

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: xₜₜ (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: xₜₜ (mM):74.76 (58.06-84.19) “; β: 0.064(0.062-0.082)“; beta line: xₜₜ (mM):67.05(58.25-88.16)”; β:0.061(0.047-0.07)“. ES: stomatocytes 4 and 3 in both lines. Although the beta line at 100 days-old presents higher body mass, its 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 MODIFIED BY ARSENIC (V)

Hernández G, Rasia M, Bollini A, Mengarelli G, Casco C, Ruíz M, Visconti M, Huarte M, Philé L¹, Rubin de Celis E², Bazzoni G. Biofísica, Medicina, UNR. ¹Física, Bioquímica, UBA.

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 \( y = 598 \times 10^{-0.01x} \) for controls and \( y = 1257 \times 10^{0.01} \) 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.
The enzyme alpha-amylose 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-amylose 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-amylose in healthy adult smokers. Saliva samples were collected in a sterile disposable polypropylene tube of 15 ml. The hydrolysis of starch catalyzed by alpha amylose 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 amyllose forms in the hydrophobic regions, giving a blue coloration. The presence of alpha amylose degrades amyllose, 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.

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.
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 hoty). Red Delicious apples were wounded with a punch, and inoculated with 20 μl of a conidial suspension (10^6 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 transposa 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.

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 associated 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 coincubation 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 synergetic 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.

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 hematoxilin-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.

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 value 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.

Financial support was provided by SECYT-UNS (P.G.I. 24/B 142).
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 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 GTases 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.
41. BM24 - DEHYDROEUCODINE DELAYS HeLa CELLS PROLIFERATION IN G1 OF THE NEW CELL CYCLE

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 ± 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.

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 secocchioliloquinone (PQ) and, to a lesser extent, secocchiolilhidroquinone (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 μg/ml) of the compounds. Among the compounds tested, PPBuC, PPU, PP AZ 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.

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 RARα, RXRα and RXRβ, and an increase of PPARα and PPARβ, which regulate many gene involved in FA oxidation, contribute to explain the effects of vitamin 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.

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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FCM, UNCuyo. IMBECU - CONICET. Mendoza.

Objective: Here we examined the Angiotensin II AT1 receptor inhibitor Losartan effect on Hsp70, Nox4 and CHIP protein expression in SHR primary culture proximal tubule epithelial cells (PTCs)Four GroupsControl (C), Angiotensina II (AII: 1x10-7 y 1x10-11M ), Losartan (L: 1x10-5M, 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 (1x10-5M, 90 min) we observed increased Cav-1expression 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 Hsp70 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 vs1.00±0.05, p<0.001 n=3 and also lower Nox4 protein levels in SHR+Los+All (Los1x10-5M 90min, All:1x10-3 M 60min) vs SHR+All (1x10-60min) 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

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The chromosome damage and its relationship with the telomeric sequences was analyzed in the progeny of mammalian cells exposed to the antimitonal 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 Chines 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

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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 hrs. 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

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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-his-tag 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

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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. Al TEM the changes were evident in the basal layer. There were few hemidesmosomes defined in some areas, irregular shape of keratinocytes and electrondensity 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 electroforetic run in minigels of poliacrilamida to 6 per cent and stained with silver nitrate. Results: of the LP were amplified 5/15 (33%); CAR bucal 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 HPV.Vs. 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 Western Blot. 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 protein 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 site 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 (spermduct, epithelial cover and glands, irritation 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 spermduct 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.
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 InsP₃ was accumulated in response to hyperosmolarity. InsP₃ 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 highosmolarity on the functionality of *T. cruzi* epimastigotes Na⁺/H⁺ exchanger. We found the alkalinization 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 para-

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55. **BT14 - ELIMINATION OF YEAST PHOTOTOXIC SITIZED 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 silesquioxane films doped with 5-(4-carboxyphenyl)-10,15,20-tris (4-methylphenyl)porphyrin, in order to obtain an 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.

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56. **BT15 - PHOTODYNAMIC INACTIVATION OF YEAST SENSITIZED 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-dimethylaminoproxy)phenyl]porphyrin (TAPP) and 5,10,15,20-tetrakis [4-(3-N,N-trimethylaminepropoxy)phenyl]porphyrin (TAPP*) were compared on *Candida albicans*. The quantum yields of singlet molecular oxygen were 0.74 and 0.72 for TAPP and TAPP*, respectively. The cultures treated with 5 μM porphyrin showed a binding of 1.04 for TAPP and 1.5 nmol/10⁶ cells for TAPP* 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 sensitzers are located in an irregular manner into the cells. TAPP* 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 PHOTHALOCYANINES 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 we studied the spectroscopic, photodynamic properties and the PDI of C. albicans sensitized by Zn(II)2,9,16,23-tetrakis(N,N,N′,N′-trimethylamine) phthalocyanine (ZnAmPc4+) and Zn(II)2,9,16,23-tetrakis(N,N,N′-methylpyridyloxy)phthalocyanine (ZnPPyPc4+). 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 illumination showed a binding of ~4.80 and ~0.17 nmol/10^6 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 - TETRADECYLTETRAMETHYLAMMONIUM DEGRADATION BY FREE AND IMMobilIZED Pseudomonas putida
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Tetradecytrimethylammonium bromide (TTAB) degradation by immobilized P. putida 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 30ºC 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 PLANTS
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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 anti-chlorine 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, lcsSL11 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.
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.

Modern olive-oil mills generate a semi-solid residue commonly called “alperujo” (AL) that presents serious environmental problems related to its disposal. It is high phenolic load due to an important source of water and soil pollution when it is improperly treated. 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.

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 spectrometry 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.
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 organogenic 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-diclorofenoxiacetyc acid (2,4-D) and acetic indol acid (NAA). The cultivations stayed in cultivation camera with a temperature of 24±/2ºC; 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.

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 summy 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 aseptic appropriate to achieve a germination rate in vitro efficient. The seeds were disinfected with alcohol 70%, sodium hypochlorite and methyl benclidazol-2-1 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 ± 2ºC 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.

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 conservation 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 nutritional means was of Murashige and Skoog (50%), supplemented with 30 sucrose g/l, 6 agar g/l and different combinations of 2,4-diclorofenoxiacetyc acid (2,4-D). The cultivations were incubated in cultivation camera to 24 ± 2ºC 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.
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.

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.

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.
73. **BV6 - SECONDARY METABOLITES WITH PHYTOTOXIC ACTIVITY IN FLOURENSIA CAMPESTRIS (ASTERACEAE)**

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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 phytoxic effects from *Florenisia campesris* (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 (I)) was isolated as the most inhibitory active principle (EC\(_{50}\)) 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 I was found to be distributed throughout the plant, with a remarkable high concentration (1.6%) in leaves and in florescences. Leaf essential oils (HD) between 0.5 and 1.5 μl ml\(^{-1}\) did not show herbicidal effects and I 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 I 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 I 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 by polymer morphological 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 descript 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 30, 60 and 120 min respectively. Inoculated and DPI-treated plants at both leaf and root showed hydroperoxide 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 STRESS**

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The aim of this study is to investigate the association of arbuscular mycorrhizal (*Glomus intraradices*) with plants of *Medicago sativa* var. Verdor and analyze their effects on a biotic stress mitigation. *Medicago* transplanted at pots of 1000 ml with soil/sand (1:1, v/v) with 10 gr *G. intraradices* inoculums. The AM and non AM plants grew in a chamber at 23:21°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 nom 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.
Digitaria eriantha cv. Sudaficana has high sensibility to different biotic stresses. In this study the aims was study the response of different morphometric and physiological parameter in mycorrizal and non mycorrizal plants under drought, salinity and cold conditions. Seedling of Digitaria eriantha cv. Sudaficana was 17 days 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 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 mycorrizization, stomata conductance (SC), photosynthetic efficiency (PE) and % dry matter in leaves and roots (DM) were measurement. The inoculated plants showed 70% of mycorrizization and don’t decreased with a biotic stress. The SC showed a significant decreased in all stress in AM and non AM plants, however without stress, the SC of AM plant increased. The PE don’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 don’t change, only in drought. The roots DM%, in AM and no AM plants decreased in salinity and cold, but in drought increased. The AM plants DM% doesn’t change showed the mitigation effects of arbuscular mycorrized.

78. BV13 - IMMUNOPURIFICATION OF SECRETORY Glycine max PHOSPHOLIPASE A2
Minchiotti M, Mariani ME, Fidelio G, Madory R.
Fac Cs Agropecuarias, CIQUIBIC-Fac Cs Químicas. UNC.

Secretory Phospholipase A2 (sPLA2, EC 3.1.1.4) from soybean (Glycine max) was partially purified and biochemically characterized to the authors. PLA2 promotes the formation of lysophospholipids, strong bioemulsifyers. The objective of the present work was: to obtain GmsPLA2 pure to homogeneity with the aim of develop structural and kinetic studies. Polyclonal antibody IgG anti sPLA2 Bothrope alternatus was covalent 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 PL2A units (purification factor= 6.1, yield= 19.1%) and Ac-Eupergit C250 was bound 17 ± 1 PL2A units (purification factor= 5.1, yield= 11.2). SDS-PAGE electrophoresis showed two bands of 11 and 13 kDa proteins related with GmsPLA2 isoforms or homologue groups, as we demonstrate by recent molecular biology studies. It is the first time that GmsPLA2 was obtained to homogeneity.

79. BV14 - RESISTANCE INDUCTION IN SOYBEAN: PHYTOALEXINS DETERMINATION
Minchioti M, Vargas L, Madory R.
Facultad Cs Agropecuarias. UNC.
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 ±2 μg/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 inductor 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%) y 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.
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 phosphotase 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 PH4-P 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 MGR, 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 reducunt 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.
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 (2x10^7 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 was the synthesis of the monoterpines 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 54 (B) and DM3702 (C)) harvested in “Valle del Conlara” (San Luis) during the 2010 was performed. A total of 300 seeds per vc were sowed in APG 2% medium (10 seeds/Petri dish of 9 cm Ø). Incubation conditions were 7-9 days at 25ºC + / - 2ºC. 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*.

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 synthesis. 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, Quiné-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 (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 choose. This group of strains will be including 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 alloagamous 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 forced selfing significantly the number of seeds per spike (H = 27.84***; H = 12.41**) and the relation grains per spikelet (H = 24.68***; H = 14.67**). In the other cultivars, Yagán and Don Santiago, the differences among treatments were not significant. Regarding 1000-grain weight, the differences among treatments were significant in all cultivars and the treatments natural pollination (25.3±6.8, 34 grains) and forced selfing (21.7±3.7, 36 grains) exceeded significantly the induced cross-pollination (5.9±4.2, 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 ANOVA and Duncan test. The interactions were not significant for all traits analyzed. The total green biomass weight was 5276.5 ± 1646.4 g/ha with significant differences for the situation with supplementary irrigation (p=0.0023), whereas the total dry biomass weight was 1751.1 ± 818.8 g/ha with significant differences in favor of the situation with supplementary irrigation (p=0.001) and the late cut in milkdough 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. crenatiifolia* 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) at noon, osmotic potential (Ψo), 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 Ψo and Ψ 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 adequate their potentials Ψ and Ψo and made greater AO by increasing salinity. These mechanisms would allow them to tolerate stressful conditions and survive in arid and saline environments.
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 (10^6 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 H_2O_2, 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 H_2O_2 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 H_2O_2 content.

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 15). 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.

93. BV34 - EVALUATION OF NATURAL PEANUT NODULAR SENESCE
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94. BV35 - REORIENTATION OF LEAVES OF Larrea cuneifolia Cav.
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95. BV36 - HPLC & FTIR PROFILES OF Melissa officinalis AND Nepeta cataria (LAMIACEAE) OF ARGENTINEAN MARKET
Popovich M1, Gette M2, Saidman E1, Aragón L1, Petenatti M2, Petenatti E1, Del Vitto L1.
1Proj. 22/0-016; 2Quim. Inorg.; 1Lab. C.C. Medic. UN San Luis. E-mail: mcpopovich@unsl.edu.ar

96. BV37 - EFFECT OF WATER STRESS ON STOMATAL DENSITY AND TRANSPERSION IN MAIZE
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97. **BV38 - EFFECT OF TEMPERATURE, WATER AND SOIL MICRO-BIOLOGICAL ACTIVITY ON THE LOSS OF MONOTERPENES FROM Tagetes minuta L. FRUITS**

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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 lost 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, ocimenones (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 PROCESS**

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Monoterpenes are secondary metabolites with recognized 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 chemotaxonomic 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-ocimenene (60.1%), (E)-ocimenone (62.6%), (Z)-ocimenone (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 PLANTS**

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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 DESEASE**

Tosti SB, Baudo J, Cecho A, Dominguez 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 Mullheman Index was used for the determination of gingival disease degree. Results: Estradiol levels founded in saliva of adolescents aged 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 cathetism (0 degree index) in 12 adolescents (40%), and bleeding in cathetism 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 ASTHMACTIC PATIENTS WITHOUT EXACERBATING BACTERIA**

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Area Microbiologia y Farmacologia, FCMedicas. UNCuyo. E-mail: mdsguad@yahoo.com.ar

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 conductive to renal stone formation. The aim was to evaluate metabolic abnormalities in renal lithiasis in pre-menopause post-menopause 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 potassium (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: phosphorus 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); hypocraturia (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**

1Inst Inmunol, Fac Cs Medicas, UNR. 2Fondación Villavicencio.

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 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, Ceicho 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 Radioimmunoassay. 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**

Irigoyen S, Abilleira E, Bustamante C, Segatto R, Mancuso P, Fingermann G, Mazzeo M, Delocca M.

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Oral Implantology Dentistry is the science that deals with the replacement of missing teeth by artificial substitutes. Its 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. El 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. Para work were used 50 skulls obtained from the Osteoteca Faculty of Dentistry, UNLP. They were numbered with Arabic numerals. Panoramic radiographs were obtained in the radiology Course Faculty of Dentistry, UNLP. They were used for measurements of optical magnification element graph type and size. Los results demonstrate the existence of a statistically significant magnification for this sector of the massif facial. Se 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 sugar 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 Odontología 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 (81.1±5.83 vs 112.8±49.45* 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 sniff 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**

Ramos GC, Biscaro AT, Ruttler ME, Castro NM, Castagnolo BM, Pizarro M, Acosta N, Della Gaspera ME, Sagua MD.

Area Microbiología, FC Médicas. UNCuyo, Mendoza. E-mail: gcramos968@hotmail.com

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 of Moore and Immuno- magnetic beads and the toxin by PCR. Suspicous 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 coliformes. 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**

Nigra A, Peretti S, Rivelle JS, Amaiden MR, Monestero NE, Santander VS, Previtali G, Casale CH.

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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 membrana 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.
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 ($X^2$) 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 ($X^2 = 0.05$; $gl = 2$; $P = 0.7763$). 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 Tucuman. In Argentina, as in most Latin American countries, prevalence rates vary.

Enterobius vermicularis larvae preference of suitability of a species for biological control. Food composition of each species were placed in each enclosure. The individual fishes were starved for a period of 72 hs before experiments. Experiments lasted 2 hours. Cladocerans were eaten most by both species, followed by culicidae larvae for J. multidentata and Cnesterodon decemmaculatus and Culicidae larvae for J. multidentata and J. multidentata present the higher food items richness and absolut numerosity and frecuency 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 make us underestimate its biocontrol skills, as it's been shown in others studies.

This study seeks to gain knowledge of cactaceae populations to aid in their management. A comparative analysis is conducted between Echinopsis leuocantha 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 leuocantha 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.

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 J enynia multidentata and Cnesterodon decemmaculatus were analyzed in mesocosm experiences to determine the relative preference for mosquitoes 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 mosquitoes 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 absolut numerosity and frecuency 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 underestimated C. decemmaculatus biocontrol skills, as it's been shown in others studies.

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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 ($X^2$) 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 ($X^2 = 0.05$; $gl = 2$; $P = 0.7763$). 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 Tucuman. In Argentina, as in most Latin American countries, prevalence rates vary.

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 J enynia multidentata and Cnesterodon decemmaculatus were analyzed in mesocosm experiences to determine the relative preference for mosquitoes 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 mosquitoes 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 absolut numerosity and frecuency 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 underestimated C. decemmaculatus biocontrol skills, as it’s been shown in others studies.

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113. **EB9 - CONTRIBUTION OF EARTHWORMS (ANELLIDA: OLIGOCHAETA) TO ECOSYSTEM PROCESS IN AGRICULTURAL SOILS**

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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, burial and mixing of vegetable 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 Vito LA, Petenatti EM, Petenatti ME, Rodriguez 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 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 arborescent. Shubby belt extends above 850 m in most windy exposures, up to 1,500-1,600 m, with *Heterothalamus alieanus* and *Eupatorium bunifolium var. bunifolium*. 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 *Polyplepis 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**

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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 Mansioave (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, Blarasin MT, Bedano JC. Fac. Cs. Ex F-Q y Nat. UNRC; CONICET. 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-. We conclude that land use influences groundwater geochemistry and the presence/abundance of copepods, which could be used as bioindicators of groundwater quality.
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.

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 acoustics 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.

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 Rio 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.

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 base don 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 function. The reclassification 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 sympathy. Thus the release call could mean a diagnostic character for distinguishing these two cryptic species.
121. EB20 - MORPHOMETRIC ANALYSIS OF Odontophrynus CRYPTIC SPECIES
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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 SYSTEMS
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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 (33º 07' W, 64º 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 smears were fixed and subsequently stained with 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 Dalc ar. The ratio N/L (neutrophils/lymphocytes) was 0.17 for the pond Las Brujas and 0.24 for Lake Villa Dalc ar. 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.

123. EB23 - HEMATOLOGICAL BIOMARKERS AND CYTOMORPHOLOGIC IN THE COMMON TOAD Rhinella arenarum
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The differential amount of white blood cells and cytometerphy 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 Dalc ar, 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 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 Dalc ar. The ratio N/L (neutrophils/lymphocytes) was 0.17 for the pond Las Brujas and 0.24 for Lake Villa Dalc ar. 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)
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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.
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: 33º59'W, 65º24'S, 20 ha, 4 fish species) and Córdoba province (L2: 34º46'S, 63º38W, 190 ha, 13 species and L3: 33º48'S, 64º51'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 Oligosurus 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.

Solanaceae is a monophyletic family and in Argentina is represented by 33 genera including 328 species, being one of the best represented in argentianian 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.
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 candelabrum-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.

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, Platymis 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 65%) and T3 Severe water deficit (reposition 33%). The quantified growth 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 species and the treatment. The ANOVA statistical evaluation (α ≤ 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.

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.

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 semi-arid 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.
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 *Asyntaxus eigenmanniorum* and *Asyntaxus 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, coincided 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 Saprolegniass 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.

Species of the genus *Contraecuem* (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. australis* in corroman *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 microscope and scanning electronic microscope (SEM). Birds dissected were positive for *C. australis*, 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 *Contraecuem australis* species. The wide distribution of *P. brasilianus* contributes with the dispersion and life cycle continuity of these nematodes.

**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 (cl-a) in Rio 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 cl-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 cl-a measured. The estimated response (R2=0.73) for multiple linear regression model of the parameter cl-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 cl-a in Rio Tercero dam, providing information relating to the distribution of this parameter over the entire reservoir.**
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 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, Proterochampsa barrionuevoi and Chanareuchus sp nov. The sediments which contained the specimens here analyzed represent a fluvial sedimentary succession. This evidence indicates that proterochampsids were aquatic or semi-aquatic 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.

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 phreatophyte 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 nutrient 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.

Proterochampsids are members of Triassic ecosystems from South America

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EB42 - PROTEROCHAMPSIDS, MEMBERS OF TRIASSIC ECOSYSTEMS FROM SOUTH AMERICA

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.
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 (Polypogum 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 Polypogum 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.
In southern San Luis province in central semiarid Argentina, more than one hundred shallow lakes are located in aridic 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 gl -1 and transparency from 0.3 to 2.3 m. There were seven fish species. Jenynsia multidentata was found in the six lakes and Odontesthes bonariensis in four. The zooplankton richness was 20 taxa: five cladocerans, six copepods and nine rotifers. Species common to the six lakes were Bosmina huanorensis, 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: 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.

Hypsersaline 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 gl -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.

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.
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), Diplomystidae (1), Trichomycteridae (3) and Pimelodidae (1). Of which 11 belong to the native fauna and 6 were introduced for different reasons. This listing is an update of a total of 17 species belonging to the taxonomic arrangement follows: Atherinopsidae (2), Characidae (3), Cyprinidae (2), Anablepidae (1) Poeciliidae (1), Percichthyidae (1), Diplomystidae (1), Trichomycteridae (3) and Pimelodidae (1). Of which 11 belong to the native fauna and 6 were introduced for different reasons. This listing is an update of 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.

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 abscessus, Harriscoleus kaparari, Megathylacus travassosti, Monticellia spinulifera, Nomimoscolex pertieriae, Pelidocotyle 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. abscessus 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.
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 to 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±8.3cm.) sub-adults 12 (LHC=6.8±4.0cm.) metamorphosed 9 (LHC=3.3±1.42cm.).
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), Aranae (25.8), Diptera (17.2), Collembocha (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); Anura (4.7). The ontogenic 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 it 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 Fértil, 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, Myiopitta monachus calita y Molothrus bonaerensis. Conservation priority species are Vultur grphus categorized as “almost threatened” in globally level and Spizipapteryx circumcinctus, as “Vulnerable” nationwide. We cite for the first time Columba picazuro and Craniolaeca ptyrhophia for San Juan province and 14 new species for Valle Fértil department. This result represents the first data on Valle Fértil park’s avifauna and will be part of base line for the purpose of declare the protected area as “Reserva de Usos Múltiples Valle Fértil”.

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 compara- tive study of five species from three genera (Liolemaus, Homonota and Cnemidophorus) for two consecutive years, analyzing gonadal patterns and cycles. There were gender differences about cycles and patterns, while intrageneric differences (Liolemaus) 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.
**EB74 - Lepus europaeus (European hare): DIET AND FOOD AVAILABILITY IN THE HYPER-ARID MONTE DESERT**

Reus ML1, Laspina CA1, Campos VE3, Andino N3, Giannoni SM1, Campos CM1.
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_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 shrubs and cactaceae, and only in the wet season for grasses and epiphytes. The hare has been considered a generalist and opportunistic herbivore because it 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%).

**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 could be observed a _Turdus chiguacu_ 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” prey 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 HILIDS (ANURA: HLILIDAE) 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 (Ojk: 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 (Ojk=0.89), Dn/Hp (Ojk=0.78) and Hp/Hpu (Ojk=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.
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 Simpon (D = 0.411) and Margalef index (l = 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.

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* (LIOLAEMIDAE) IN SAN JUAN, ARGENTINA. 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 Sorensen 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 (Is = 0.66) between periods. Dominance in certain items was in reproductive period (PR). No differences in trophic amplitude for both periods were founded. *L. parvus* is a generalist and omnivore species that consumes arthropods generally in both periods.

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 *Liolaemus 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<sub>Subadult</sub>: R=0.47; p<0.004; Spearman<sub>Adult</sub>: R=0.87; p<0.0009. *L. parvus*: Spearman<sub>Subadult</sub>: R=0.69; p<0.02; Spearman<sub>Adult</sub>: 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.

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.*
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: Rhacarcis punctatus, Rhacoporus pratensis and D. pratensis; a few Spyllina spp. and Neopedies spp. in 1941; Staurorhectus longicornis, Scyllina variabilis, C. speciosa, D. pratensis, D. vittatus, Rhamnatocerus pictus, Zenipoda 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 Acriid species. Specimens were identified using the taxonomic key from Salto (1999). The following species of the Acriididae family were identified: D. elongatus, Baeacris punctatus, Ronderosia bergi, D. pratensis on alfalfa crops and on grasslands: D. pratensis, R. bergi, D. conspersus, B. punctatus, Allotruxalis strigata, C. miles, D. elongatus, R. pictus, Borellia pallida, S. longicornis, Z. tarsata. The survey was carried out during the campaign 2010-11, using drag net entomology. Of 15 species observed only five of them agree with those identified in previous studies.

Global climate change will affect more to the communities of sub-tropical and tropical ectotherms that communities are distributed at higher altitudes. The aim of this study is compare the 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 sub-tropical and tropical ectotherms that communities are distributed at higher altitudes. The aim of this study is compare the thermal safety margin (TSM) and heat tolerance (HT) of tadpoles of Rhinella spinulosa and Odontophrynus 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 ± 0.3ºC and O. cf. occidentalis = 15.2 ± 0.5ºC. In addition, we detected significant changes in the parameter HT (Mann Whitney U = 4.15, p <0.00001) where: R. spinulosa = 12.9 ± 0.3ºC and O. cf. occidentalis = 3.8 ± 0.4ºC. Amphibian communities in lowland are apparently more susceptible to global warming that amphibian communities found at highland. 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.
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 Escorpionesidae “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.

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. Courtship 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 (51%) 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.

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.

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 high prevalence 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.
Seven terrestrial species of the family Orchidaceae are cited for the province of San Luis. They are: Scolicia lanceolata (Aubl.), Habenaria gourleiana Gillies ex Lindl., Habenaria hexaptera Lindl., Pelexia bonariensis (Lindl.) Schltr, Pteroglossapis argentina Rolfe, Aa achalenisis 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. Scolicia 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 hillside in the “Sierra de San Luis”, as Habenaria gourleiana not found. Pteroglossapis 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 lithosols, d) on hillsides, valleys and plains barely grazed, which recognizes the vulnerability of these species to trampling and overgrazing.

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 submersed and floating vegetation. Species of the genus live from cold deep areas to warm top areas of the soil. That in field conditions may thermogulates by vertical movements temperature in similar manner those epigeal reptiles. We believe to conclude that these fossorial reptiles are capable of regulating body remarkable higher than those reported for other amphisbaenids. We was tigmotherm obtaining heat by substrate contact (Spearman, Ts: rs =0.80, P=0,00001). Tb mean was 28.5 (DE: 3.01°C), this value is understood because the observations and captures are difficult. 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 Ambisphaenidae plumbea in lab conditions. During May-2007 the experiment was performed in a sample of amphibiaenids (N=4) placed in open-top terrarium (120x60x40 cm) with sandy-loam substrate (3cm depth) 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 amphibiaenids was obtained every 10 m for 5 hr. A. plumbea was tigmotherm obtaining heat by substrate contact (Spearman, T_s: rs =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 amphibiaenids. 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
Laspiur A*, Acosta JC, Ibargüengoytía NR*, Fava G. Dpto Biologia. FCEFyN UNSJ CRUB UNC Comahue *CONICET. E-mail: laspiursaurus@gmail.com

Given their dependence on temperature and solar radiation, the lizards constitute an interesting biological model for determine individual responses as the ultimate adaptation modulators and, therefore, it is possible to determine different species life histories and physical environment aspects-related. Therefore, our aim is contribute to knowledge of thermal relationships 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, being a “good” 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
Laspiur A*, Acosta JC, Ibargüengoytía NR*, Fava G. Dpto Biologia. FCEFyN UNSJ CRUB UNC Comahue *CONICET. E-mail: laspiursaurus@gmail.com

Temporal and spatial activity constitutes two of three dimensions of the resources partition that the species may segregate and explain 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 (χ²) 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
Cortez R, Marinero V, Sanabria E, Quiroga L, De los Ríos C, Vidal M. FCEFyN-UNS J. E-mail: cortezricard@gmail.com

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 squijugal. 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)
Galvani G, González E, Sanabria EA. FCEFyN UNSJ IHEM-UNC; IZA-UNC; CONICET. E-mail: guillegalvani.sj@gmail.com

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 20.72±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 ventral 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±1.13. Vent tube medial, attached to the ventral fin. Oral disc anterodorsal. Labial tooth row formula 2 (2)/2. The present study extends the knowledge of the tadpole of P. nebulosum allowing unmistakable identification.
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 maduration 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.

The aim of this study was to evaluate the quality of the lower-middle basin of Suquía River integrating information from different biomarkers. We selected 3 sites along the basin (before Córdoba 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 *Lepidophyllum 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.

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 (≤15 days of water permanence), while generalists were associated with longer permanence water. Collectors, *Chironomidae* and *Stratiomyidae* were associated with intermittent water bodies, *Psychodidae* and *Syrrhidae* 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.

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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.
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.

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 parameters herpetofaunística biodiversity conservation and environment. The aim of this work is to present the first data trophic parameters herpetofaunística biodiversity conservation and environment. The aim of this work is to present the first data trophic parameters L. eleodori (IGUANIA: LIOLAEMIDAE), SAN JUAN, ARGENTINA Astudillo GV, Córdoba MA, Acosta JC. Dpto. Biología. Fac. Cs. Exactas Físicas y Naturales UNSJ. E-mail: astudillo.vanesa@gmail.com

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.
That the urinary route is preferential in detecting in equal amounts of both excretes. Therefore, we proposed EM18 - EFFECT OF FASTING ON GROWTH PARAMETERS

Supported by grants from SECyT-UNC, CONICET, FONCyT. tested by matching our results and those reported for other rodents ered the the and guinea pigs; currently considered cavy-like forms) while in this time, blood samples were collected to quantify cortisol and restriction. Two groups of fish were fed (F , control) or unfed (UF) growth pattern and organosomatic indexes (OI) in response to food extracted to determine the OI. Somatic growth parameters were calculated BMI, HOMA-IR and HOMA-β.

DIABETIC PATIENTS SYNDROME IN FIRST DEGREE RELATIVES OF GENE AND ITS RELATIONSHIP WITH METABOLIC

The cholesteryl ester transfer protein (CETP) plays a key role in the metabolism of the HDL particles. One of the polymorphisms studied was 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 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 (PPARα) 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 LDL-c, 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 PPARα were 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 PPARα, 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.

Whether the steroid hormone is primarily passed into urine or fe-

ces is species-dependent, and this information is useful to a precise
docrine monitoring; in excretes metabolite concentrations are
frequently several times higher than the concentration in blood.

Wild Chinchilla lanigera is threatened (CITES I), although, do-

mesticated 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 cortico-
terone 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 "Hystrixcognaeth" (chinchilla and guinea pigs; currently considered cavy-like forms) while in the "Sciurognatha" (squirrel and mouse-like forms, formerly considered "Sciurumorpha" 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.

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), weight (p=0.0001), and body length (p=0.0001). No differences were detected in the condition factor [(P/L)3*1000], glucose and cortisol levels (p>0.1). The hepatosomatic index was reduced in UF fish (p=0.0001) 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. 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.
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 thioacetothiuric 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 chemical 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.

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) 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 (RGMa) 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 RGMa and RGMin were observed in the HFC group. IMO increased RGMa in both diets and RGMa 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 CAROTENOIDS
Díaz AC1,2, V elurtas SM1, Cuartas E1, Fenucci JL1,2.
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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-picrylhidracil (DPPH), using EPR. The 96-h LT90 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 AC1,2, Cuartas E1.
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 all the salinity conditions (extremely low temperatures and high UV radiation) that the shrimp are at the top of the Antarctic food chain.

203. FA11 - CLIMATE CHANGE, FEEDING AND BLOOD ANTI-OXIDANTS IN THREE ANTARCTIC PENGUIN SPECIES

Pygoscelis antarctica (Chinstrap), Pygoscelis papua (Gento) y Pygoscelis 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 (GIPx) 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 GIPx 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 OOCYTES
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Amphibian ovarian oocytes do not usually resume meiosis unless stimulated with steroids. However, demed 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 anehtory 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 hypertrophia was observed in all groups. Testis weight and epidydimal sperm count were not affected as well. However, NS and Sep treatments caused a rearrangement in the epidydimal 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 E, Caviedes-Vidal E.


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 dehydroxlated and embedded in paraffin. Histological studies were performed on 5 μm 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. Supported by CyT-UNSL 22/QT51 to ECV. and PIP No. 11220090100998 CONICET and CyT-UNSL 0110 to JGC.

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

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

Laboratorio de Biología Integrativa. IMIBIO-SL. CONICET. UNSL. San Luis. Argentina.

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.

Laboratorio de Biología Integrativa. IMIBIO-SL. CONICET. UNSL. San Luis. Argentina.

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.
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 reproductive season (PreR, 0.39±0.01 ng/ml), they increase during the reproductive season (R, 0.90±0.02 ng/ml) and achieve highest levels during the post reproductive season (PostR, 1.34±0.04 ng/ml). Moreover, during PreR season total aromatase activity is significantly lower than during the rest of the year (PreR, 3.66±1.46 pmol/min; R, 13.56±2.15 pmol/min; PostR, 16.20±9.47 pmol/min). Taken together, these results suggest that the BO is the main source of plasma estradiol.

**FA21 - EXTRACELLULAR 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 intestine of *P. canaliculata*. It was significantly higher 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 (serine-protease inhibitor) inhibited the 60% of protease activity in crop and stomach contents. This inhibition was positive in all soluble proteases found (32, 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 carried out protein digestion using soluble and membrane associated proteases (leu-N-aminopeptidases).

**FA23 - THE PHYSIOLOGICAL RELEVANCE OF MIDGUT MEMBRANE PROTEINS IN THE PROCESS OF LIPID TRANSFER IN A VECTOR OF CHAGAS’ DISEASE**

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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*.
213.

**FA 28 - 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 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.

**Funded by PICT 2007 1320 to EC-V.**

214.

**FA 29 - 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 and is an agricultural plague. Its 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 three-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 37°C 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 genes for urogenital development.

215.

**FA 31 - CHROMOSOME STUDIES IN CRYPTORCHID DOGS***

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Cryptorchidism is the failure of the testes 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 three-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 37°C 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 genes for urogenital development.

216.

**FA 32 - 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* 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.

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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, rubefascient, 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. a.) 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 ± 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 INFLAMATION 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 glutathion 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 oxidative stress and inflammation were analysed on 2% agarose gels containing Gel Red. RT-PCR. Tissue total RNA was isolated using TRIzol. The expression of NRF-2 and TNF-α were analysed on 2% agarose gels containing Gel Red. RT-PCR. Tissue total RNA was isolated using TRIzol. The expression of NRF-2 and TNF-α were increased in Cd treated 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 TNFα 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, glutamal transferase and bilirubin were determined. The Ad infusion produced reduction of AST and ALT (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 rosvastatin (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, afterRos treatment. A dose dependent effect of rosvastatin 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.
Congenital obstructive nephropathy is characterized by tubular apoptosis and fibrosis. Stains 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-β1, WT1, Snail, BMP-7 and E-cadherin protein expression. Results: UUO significantly increased tubular apoptosis, dilatation and tubulointerstitial fibrosis coupled to increases in TNF-α and TGF-β1 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.

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, AT₅ receptor, TGF-β and Renox (NOX) 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 AT₅ expression and NADPH activity were reverted 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 AT₅ receptor dependent at the mitochondrial level.

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.
In our country, in late 2007, new strobiurin and triazole based biocidal 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 preliminary determined an inhibitory effect of TFT on BChE activity of *S. nasicus* tadpoles.

**FT12 - EFFECT OF THE FUNGICIDE TRIFLOXYSTROBIN ON B-ESTERASE ACTIVITY IN TADPOLES OF Scinax nasicus (AMPHIBIA: ANURA)**


**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.
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.


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.

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, artherosclerosis 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 Ibuprofene 1.5, Lansoprazole and Enalapril hydroxychlorohidize (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.
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 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-postively 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.

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 BP A/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 ovarioctomized 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 significantly 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.

In previous studies we reported antulcer 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.
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 (ERα) and beta (ERβ), androgen receptor (AR) and p27 by immunohistochemistry. The expression of ERα, ERβ 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.

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), 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.

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 conform 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 ppm of Cd (as Cl2Cd) in the drinking water, respectively, for 60 days. In toraxic 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 gluthation 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.
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-butilhioscina 5.1, propinox-clonixinate lyisne 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 than 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 connotations for the health which this misuse implies.

The Transcription Factor7 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 two hospitals (A, B) and a social security (SS) of the city of San Luis. 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% for TT in controls, in diabetics were 19.2% GG, 69.2% and 11.5% GT % TT. The presence of “T” allele of both rs12255372 (G/T) and rs7903146 (C/T) polymorphisms of TCF7L2 gene confer susceptibility to T2DM.

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.

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 classi- fied according to ATC (Anatomical Therapeutical 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) 5.7, 2.4, 5.8; 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, 5.5; genitourinary (N) 2.5, 7.2, 5.8; alimentary tract (K) 6, 1.9, 29.4, cardiovascular (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, 5.5; genitourinary (N) 2.5, 7.2, 5.8; alimentary tract (K) 6, 1.9, 29.4, cardiovascular (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.
245. **FT40 - HYPERTENSION, CO-MORBIDITY AND USE OF ANTIHYPERTENSIVE DRUGS IN A HEALTH CENTER OF GODOY CRUZ (MZA.)**

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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: 45% 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% 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 17β-ESTRADIOL AT ENVIRONMENTAL LEVELS**

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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 17β-estradiol (E2) on cyp19α1a (gonadal aromatase) and cyp19α1b (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 days. 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 cyp19α1b transcript abundance in a concentration-dependent manner was observed. However, no effects on gonadal aromatase expression were observed. The remarkable changes observed in cyp19α1b 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 reproductible 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 17β-estradiol (50, 100 and 250 ng/L), 17α-ethinylestradiol (10, 75 and 250 ng/L) and 4α-nonilfenol (1 and 10 μg/L). 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 4α-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 JUNGLA POLITA USING ALLIUM CEP A 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.
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 Allium 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 cystotic 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%.

Using tests of genotoxicity in peripheral blood of people exposed to pesticides is possible to determine the effects of such exposure on the 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. We were carried out tests for Chromosomal aberrations (CA), Micro-nuclei (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 ± 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.

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 surfase spectrofluorimetry ($\lambda_{em} = 532$ nm; $\lambda_{ex} = 548$ nm). At optimal experimental conditions, quantitative recovery was reached with a detection limit of 0.662 pg L$^{-1}$ and quantification limit of 2.20 pg L$^{-1}$. The calibration sensitivity was of 1.22 L pg$^{-1}$ for the new methodology with a linear range of 2.20 pg L$^{-1}$ to 779 pg 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, Argentina) 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.
Many plants or their active ingredients are known for their anti-allergic 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%), Lamiales & Plantaginaceae (10%), and Ephedraceae, Fabaceae, Magnoliaceae, Polygonaceae, Rosaceae & Verbenaceae (3%). 26 families bring one species each. Four species (6%) are excluded for phytotherapics (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 traditional therapies in 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.

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 7α-hydroxylase (CYP7A1). The animals were divided into four groups of six subjects each, two of those groups were fed diet AIN-93 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.
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 to 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 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 (PICT 2008-0554), UNT. CIUNT 1IBT. ICB-UNSJ, San Juan 2FACET-UNT, 3FCNeIML-UNT y FML, Tucumán. E-mail: lluna@unsj.edu.ar

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 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, Ti 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.
261. FT56 - MORTALITY EFFECTS OF SEMI-SYNTHETIC DITERPENES ON Tenebrio molitor
Reta GF, Gatica A, Sosa ME, Tonn CE, Donadel OJ.
INTEQUI-CONICET, FQByF, UNSL, San Luis. E-mail: greta@gmail.com

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 derivates 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 AEIC (PCI A / 025750/09).

262. FT57 - BIOACTIVITY AND PRELIMINARY PHYTHOCHEMICAL STUDIES OF Nepeta cataria L.
Llaver AP, Favier LS, Gatica A, Tonn CE.
INTEQUI-CONICET. FQByF. Universidad Nacional de San Luis. San Luis, Argentina. E-mail: lfavier@unsln.edu.ar

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 nepetolactone 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 nepetolactone 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 nepetolactone, 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 AEIC (PCI A / 025750/09).*

263. IM3 - DIFFERENT PROFILE OF RESPONSE TO Th1/Th2 STIMULUS BY CLARA CELL
García LN, Leimgruber C, Uribe Echevarría E, Maldonado CA. Centro Microscopía Electrónica UNC. E-mail: lgarcia@cmefcm.uncor.edu

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 IMMUNE REACTIVE PROTEINS BY MASS SPECTROMETRY
Villa MC, Mattar MA, Guerra RA, Caceres CS, Callegari E, Cortinas TI.
Área Microbiología, UNSL. E-mail: ticor@unsln.edu.ar

*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 antimicrobial, 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 (34 - kDa); flagellin (23 k-Da). 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. M13 - EFFECTIVE BINARY AND TERNARY ANTIMICROBIAL FLAVONOIDS COMBINATIONS WITH NALIDIXIC ACID AGAINST Escherichia coli ATCC 25 922
Talia JM, Debattista NB, Tonn CE, Pappano NB.
UNSL. 5700 San Luis. E-mail: jmtalia@unsl.edu.ar

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.

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<thead>
<tr>
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<th>2’,3-(OH)2-chalcone</th>
<th>2’,4-(OH)2-chalcone</th>
<th>2’,4’-(OH)2-chalcone</th>
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<tbody>
<tr>
<td>0</td>
<td>NA</td>
<td>0</td>
<td>NA</td>
</tr>
<tr>
<td>121.5</td>
<td>76.5</td>
<td>52.5</td>
<td>75.8</td>
</tr>
<tr>
<td></td>
<td>44.2</td>
<td>28.1</td>
<td>74.8</td>
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<tr>
<td></td>
<td>57.5</td>
<td>40.0</td>
<td>57.5</td>
</tr>
</tbody>
</table>

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. M14 - 2’,4-DIACETYLCHALCONE PREPARATION AND ANTIBACTERIAL ACTIVITY AGAINST Escherichia coli ATCC 25 922
Talia JM, Debattista NB, Tonn CE, Pappano NB.
UNSL. 5700 San Luis. E-mail: jmtalia@unsl.edu.ar

In the present study, 2’,4-dihydroxychalcone, a compound with higher antibacterial activity against E. coli, was selected for to obtain 2’,4-diacetilchalcone. For dihydroxychalcone 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-diacetilchalcone 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) were evaluated and informed for both compounds alone and its binary and ternary combinations.

<table>
<thead>
<tr>
<th></th>
<th>2’,4-dihydroxychalcone</th>
<th>2’,4-diacetilchalcone</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>NA</td>
<td>NA-Ru</td>
</tr>
<tr>
<td>75.8</td>
<td>44.2</td>
<td>28.1</td>
</tr>
<tr>
<td>151.0</td>
<td>91.4</td>
<td>139</td>
</tr>
</tbody>
</table>

The derivative obtained showed less antimicrobial effect against E. coli (MIC: 151 μg/mL) and only 2’,4-diacetilchalcone-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 diphenyletherbutylnsilyl to enhance the bacteriostatic activity of hydroxylated flavonoids.

267. M16 - PREVALENCE OF Streptococcus agalactiae COLONIZATION IN PREGNANT WOMEN IN THE CITY OF SAN LUIS, SENSITIVITY PROFILE AND RESISTANCE TO MACROLIDES
Ronchi G1, Satorres S2.
1UNSL. & Clínica y Maternidad CERHU. 2San Luis Argentina. E-mail: gdronchi@unsl.edu.ar

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%). Suspicous 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 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. M17 - PREVALENCE OF Streptococcus agalactiae COLONIZATION IN PREGNANT WOMEN IN THE CITY OF SAN LUIS ACCORDING TO AGE AND UNDERLYING DISEASES
Talia JM, Debattista NB, Tonn CE, Pappano NB.
UNSL. 5700 San Luis. E-mail: jmtalia@unsl.edu.ar

Streptococcus agalactiae is 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%). Suspicous colonies were identified by conventional biochemical tests. Patients were divided in 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).
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 cellulolytics 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 *Pseudobutyrivibrio ruminis*, which is closely related to *Butyrivibrio 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.

**M112 - GROWTH AND PROTEASE PRODUCTION BY NON-O1/ NON-O139 Vibrio cholerae USING FAST AND SLOW ASSIMILATION CARBON SOURCES**

Centorbi HJ, Aliendro OE.

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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 1:1000 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.

**M113 - Lactobacillus ADHESION TO EPITHELIAL CELLS FROM BOVINE TEAT CANAL**

Frola I, Pellegrino M, Larrardé M, Ferrari M, Raspanti C, Bogni C. Fac Cs Exactas, Fco-Qcas y Naturales, UNRC. E-mail: ifrola@exa.unrc.edu.ar

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 (BTC). Co-agggregation between LAB (n=6) and the BMP (n=20) was assay by co-incubation of 10⁶ UFC/ml of LAB and BMP, Gram-stained and microscopic observation. For adhesion assays, 10⁶ cells/ml obtained by scraping 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, *Weisella cibaria* CRL1840 and *Pediciococcus 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
Pellegrino M, Froila I, Marín A, Ducrós E, Odieiro L, Bogni C. Fac. Cs. Exactas, Fco-Qcas. y Naturales, UNRC. E-mail: mpellegrino@exa.unrc.edu.ar

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). The 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 BAL 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, Weissella 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 - TETRADECYLTRIMETHYLLAMMONIUM DEGRADATION BY P. PUTIDA AND RELATION TO GLOBAL REGULATORY SYSTEMS
Liňfourrena AS, Velázquez G, Lucchesi GI. Dpto. Biología Molecular. FCEFyQyN. UNRC. 5800-Rio Cuarto. Córdoba. Argentina. E-mail: aliffourrena@exa.unrc.edu.ar

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 kk::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
Favier GI, Isaguirre A, Lazarte Otero V, Lucero Estrada C, Salinas AG, Velázquez L. Microbiología General. Fac. de Qca. Bioqca y Fcia. UNSL. E-mail: gfavier@unsl.edu.ar

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 XbaI, 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 where 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

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 XbaI (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
Lazarte Otero VS, Favier GI, Escudero ME, Velázquez L.
Microbiología General. Fac. de Qca Bioqca. y Fcia. UNSL. E-mail: vs lazarte@uns.edu.ar

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, fosfomycin, chloramphenicol, gentamicin, colistin, furazolidone, aztreonam, erythromycin, tetracycline. The disk diffusion method according to Kirby-Bauer was used and bacterial sensitivity was 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
Lazaro C¹, Gaurón C¹, Lazaro V², Di Genaro S², Velázquez L¹.

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 cryoviolent technique. The culture medium was trypticase soy broth (TSB) added with 0.25% glucose. Furthermore, two strains were studied under different concentrations of KHP04 (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 KHP04. The highest ability to form biofilm of Ye C1 strain was observed with null glucose or sucrose, 2% NaCl and 27 mM KHP04. On contrary, the ability to form biofilm of Ye 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 antiinflammatory and expectorant, as well as as antiinfective, antiparasitic, anti-inflammatory and hypoglycemic. 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 10⁶ UFC/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, P aeruginosa 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 “arcueja” is used in Argentine in folk medicine as hepatic, anti-inflammatory and eutopic. 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.
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*.

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 -20ºC or -70ºC, 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 (-70ºC) before the freeze-drying with SMYG was of 98% (p<0.05); while when they were frozen slowly (-20ºC) 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 (-70ºC) before the freeze-drying. 

Supported by SeCyT-UNSL 22/Q541.

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 vischaca (*Lagostinus 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 *Clostridium 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.

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 commercial 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. 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 23º Brix, at 28ºC in Erlenmeyer flasks plugged with a special glass device (Müller valve), for 16 days. Results showed that two strains of *Saccharomyces cerevisiae* had fermentative kinetic and fermenting power similar to commercial strains. Whereas two strains of *Saccharomyces exigus* 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.

**Support by SeCyT UNSL 22/Q541**.
Morphological conversion from spiral to coccoid forms has been described for Helicobacter pylori under several conditions such as aerobicobiosis 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.

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-familial 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 1.5x10⁶ 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 10⁵ 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.

Bacillus species produce a large number a bioactive 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 glucose 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.
The absence of Y opH in C57BL/6 mice. The profile of Y ops by SDS-PAGE confirmed the aim of the present work was to investigate the degree of at-
cytosis of Yere with immune mechanisms in the host. Y op H prevents phago-
mid (pYV) that encodes YeW A-314 infected mice (50%). Accordingly, at days 7
infected with Ye, infected mice (<0.001). The mutant strain was com-
deficient in Y op H (ΔyopH). The mutant strain was com-
pared with YeW A-314 infected mice (1-5 x 10^8 CFU).

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 plas-
mid (pYV) that encodes Yersinia outer proteins (Yops) which inter-
fere with immune mechanisms in the host. Yop H prevents phago-
cytosis of Ye by macrophages and blocks the lymphocyte activation.

The aim of the present work was to investigate the degree of at-
tenuation of Ye deficient in Yop H (ΔyopH) after oral infection in C57BL/6 mice. The profile of Yops by SDS-PAGE confirmed the absence of YopH in YeΔyopH strain. C57BL/6 mice were orally infected with Ye wild-type (WA-314) or ΔyopH (1-5 x 10^6 CFU).

Percent survival was higher (80%) in YeΔyopH infected mice com-
pared 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
YeΔyopH infected mice (p<0.0001). The mutant strain was com-
pleted clarified from these organs 21 days after infection. We con-
clude that YeΔyopH was markedly attenuated after oral infection.
The results suggest the potential use of YeΔyopH as vaccine carrier for oral immunizations

291.
MI40 - LAND USE AND BACTERIA ANTIBIOTIC RESIS-
TANCE IN GROUNDWATER. RIO CUARO, CORDOBA
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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 ground-
water samples (n=25), and the relation between bacterial indica-
tors 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 suscepti-
Bility testing of E. coli was performed by a disc diffusion method.
The highest value of RT was 6x10^10 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 peni-
cillin, 29% to ampicillin. None was to tetracycline, ciprofloxacin,
amoxicillin+clavulanic acid, chloramphenicol and cephalothin.
The antibiotic resistance pattern showed high percentage of E. coli re-
sistant to common antimicrobials in veterinary medicine revealing
mainly the impact of livestock in groundwater. The spread of resist-
ant bacteria in aquatic environments could pass through the food
chain to humans.

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' Can-
dida, with decreased resistance towards antifungal agents, espe-
cially fluconazole. Our objectives were to characterize the differ-
ent 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 vulvovagi-
tis, and for its identification Sabouraud dextrose agar (British),
CHROMagar Candida, Nickerson Agar (Merck), Api Candida
(BioMerieux), urease broth (Britania), Pope agar (Britania) Neo-
Sensitab (25ug Fluconazole (Flu), Voriconazole 1UG (Vo), ampho-
tericin 10ug (A), 5ug caspofungin (Cas)) were used. Of 46 samples
studied, the Candida’s species isolated in percentage were: albicans
70%, glabrata 15%, guillermondii 11%, kruised 4%. Resistance to
fluconazole was of a 100% in C. krusei. C. glabrata and C.
guillermondii 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 rel-

evant 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.
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 enteritis (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.

Shiga toxin producing Escherichia coli (STEC) constitutes an important cause of diarrheal 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 slaughtering 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 was 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 0157: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 dendogram 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.

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: 10⁶ 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. and 42% against A. c. Index 5 was observed in four yeasts against A. c. and only one against A. c. Torulaspora delbrueckii Td129 reduce 64.5% growth of A. c. and Pichia membranifaciens BPm6 reduce 52.7% growth of A. c. 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.
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 Botrytis 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 inhibition (S. cerevisiae, 1 S. kluveri, 1 S. bayanus, 4 T. delbrueckii, 1 C. sake, 1 D. vanrijae, 1 S. cerevisiae). 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. kluveri, 1 H. uvarum, 1 D. vanrijae). 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.

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 (10^6cfu/mL) was inoculated in wounds. A suspension (20μL) of B. cinerea (10^4spore/ml) was inoculated into a wound. After 2 and 24h 20μL of yeasts (10^6cfu/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 (S. cerevisiae, 1 S. kluveri, 1 S. bayanus, 4 T. delbrueckii, 1 C. sake, 1 D. vanrijae, 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. kluveri, 1 H. uvarum, 1 D. vanrijae). 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.

297. MI56 - IN VIVO CURATIVE ACTIVITY OF ENOLOGICAL YEASTS AGAINST Botrytis cinerea 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 (10^6cfu/mL) was inoculated in wounds. A suspension (20μL) of B. cinerea (10^4spore/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. kluveri, 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. vanrijae, 1 K. marxianus, 1 H. uvarum, 1 C. albida, 1 P. 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.

298. MI57 - BIOCONTROL IN VIVO OF DIFFERENT STRAINS OF Botrytis cinerea USING ENOLOGICAL YEASTS IN GRAPE
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In 2001 van den Hoogen discovered Human Metapneumovirus (hMPV, RNA virus, single negative chain) in Holland. Located in Paramyxoviridae 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 Santisima 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 taxa (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.; 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.
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 different respect yeasts and filamentous fungi in April 2010. 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.

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.

Denture stomatitis is an inflammatory process of the oral mucosa. Mucous Atrophic (with erythematous lesions) and hyperplastic 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%.

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. perfringes. An enterotoxigenic strain of C. perfringes, 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-log10 and 1-log10 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. perfringes.
PCR (n=156), a 69.87% were non-pathogenic E. coli Citrobacter spp. (1.13%). From were pathogenic strains compatible with Mc Conkey agar. Lac (+) colonies compatible with rectal swabs (n=620) were collected from dairy calves and cultured on pathogenic bovine strains of Universidad Nacional de Río Cuarto. Ruta 36 Km 601. Río Cuarto. Molinero D, Vivas A, Picco N, Alustiza F, Bellingeri R, Motta C, Grosso MC, Busso L, AND ROTA VIRUS STRAINS IN DAIRY NEONATAL CALVES

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 Al³⁺. Only 50% of the mutant cells managed to grow in the presence of the ion (the amount of viable cells decreased from 10¹² ufc ml⁻¹ to 10⁶ ufc ml⁻¹ 15 min after Al³⁺ addition). Also, Al³⁺ in the membrane of wild-type and mutant strains was visualized using the fluorochrome 2′,3′,4′,5′-pentahydroxylavone (morn 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 Al³⁺. These results support that phosphatidylcholine is involved in the response of P. putida to Al³⁺ and acts as a temporary reservoir of available ions.

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 BETC (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.

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 Gender 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 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.
309. M172 - RESPONSE TO BROMIDE-TETRADECYLTRIMETHYLMAMMONIUM IN A P. putida CARDIOLIPIN SYNTHASE DEFICIENT

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Pseudomonas putida A ATCC 12633 responds to tetradeacyltrimethylammonium (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 pps364 as cardiolipin synthase (cls). The P.putida A ATCC 12633 cls homologue was disrupted to generate the corresponding mutant, P.putida A ATCC 12633 Δcls. 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 NH₄Cl 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 phosphatidyglycerol 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 GABAₐ RECEPTOR

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Previously we described the binding behavior of synthetic steroids (SE) structural analogues to Allopreganone (Allo) and Pregnanolone (Pregna), using specific agonists of GABAA 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 ³¹S-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 (IC₅₀= 66,29; 89,12; 92,89 / 133,65; 141,25 y 489,7 nM respectively), although the Ns2 was the least effective. While the values are most 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 Allo. 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

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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±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 received 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 3βHSD ENZYME ACTIVITY IN HIPPOCAMPUS AND CEREBELLUM OF RAT

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The 3 beta-hydroxysteroid dehydrogenase (3βHSD) is present in different tissues, it catalyzes Pregnenolone(P5) into Progestrone(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 synthetys 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 3β-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 (10μM, 50μM y 100μM) 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.
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±15.84* and 100±15.30 vs 191.16±11.30* respectively. Acci of treated rats shows a significant increase 100±14.00 vs 179.15±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.

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/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.

In the experimental model of ROP, and to evaluate hypothermia as a possible protective treatment. We used retinas asphyxia develop retinal lesions consistent with ROP. Our aim was to study the impact of hypoxia-ischemia on retinal neovascularization and astrogial 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±3% 7d, 38±6% 15d, 33±4% 21d, 35±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 increase of GFAP expression was observed by WB in PA vs CTL from day 7 ahead, with a peak at 15 d (235±68%, 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 vessels/field, p<0.01), while HYP was similar than CTL. In conclusion, AP results in angiogenesis and gliogenesis in the inner retina and hypothermia showed to be protective to avoid the damage.
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 female 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 rats, 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.

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

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 neuroadaptative 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 neuroadaptative 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 tumor angiogenesis and it also participates in the survival of tumor 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 blots.

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 DIMETHYLDIZAMINE
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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 pathologically 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.
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 weighed 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.

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 MB49 and MB49I cell cultures and orthotropic 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.

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 admitted 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.
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, *L. multiflorum* and *L. multiflorum* Axcella in the south center of Cordoba. 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 Rio Cuarto, where crop producers use soil-applied manure avaries. 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.m⁻²) and a urea to the control (12 and 24 g.m⁻²). 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 havervest 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.
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 acceptable for human consumption.

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.

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.

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 (Aply Aloysia citriodora (Ac) against Tribolium castaneum. 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).
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* and *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.

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 europaea* 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. 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.

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.
Fruit growing development in sparsely exploited sites requires pre-
liminary studies ambient and economic viability. The proposal was
obtain data climate, select cultivars of possible adaptation, and stud-
ies financial assessment in Quinces. Thermal supply winterly Quines
(87/08) was 425 +/- 124 Hf; 510 +/- 202 UF (Utsah), 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 en-
able soon recovery these costs, very important in the yield Olive
tree, a kind of late start production.

432.
PV20 – PEACH (Prunus persicae L.) VARIETIES FOR
QUINCES-CANDELARIA ZONE IN SAN LUIS, PROVINCE
Lucero RA1, López F3, Arjona C1, Orta F1, Rojas E1, Consiglhop Robles 1F1,
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 mar-
ket. The study area includes: the sites Quinces and Candelaria ,
Ayacucho locations (north of the province of San Luis). Was re-
vised 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. Its
metodology 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 exces-
sive cooling requirements for the area. The results are: The prop-
ties 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 inten-
sity. 9 varieties were selected with a requirement less than or
equal to250 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 Au-
gustine” (300 chill hours), is recommended cyanamide applica-
tions when the cold winter might be insufficient.

434.
RE4 - THE INHIBITION OF PDGF SYSTEM (platelet-derived
growth factor) AFFECTS FOLLICULO-GENESIS IN
OVARIES FROM HYPERSTIMULATED RATS
Pascuali N1, Scotti L1, Abramovich D1, Pascuali N1, Pozas C1, Bas D1, Tesone M2, Parbarell F1,
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Several angiogenic systems regulate selection of dominant follicle
destined to ovulation. However, no study demonstrated the involve-
ment 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 μg/ovary) in one ovary and the con-
tralateral 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
obtained to measure estradiol (E2) and progesterone (P4) levels by
RIA. Ovaries were isolated for histological sections to detect t-actin
(cell periendothelial marker) by IHQ and to isolate proteins by west-
ern 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 treat-
ment (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 ves-
sels. In addition, this treatment reduces the vascular permeability
which is a characteristic of this syndrome.
Several nutritional factors can affect the mammal reproductive functions; among them, PUFAs ω6 and ω3 modify structural and functional characteristics of the sperm membranes. We analyzed the influences of cod liver oil (rich in ω3 PUFAs) on functional activity of epididymal sperm from Albino Swiss mice. Two groups were considered: C (Control; ω6/ω3 ratio=19.87; n=10) and P (Problem; ω6/ω3 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 trend was found in P and C respectively: (Mean ± SEM) concentration 25.3 ± 2.7 vs 23.5 ± 3.7 (x106/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 ω3 can optimize sperm functional activity. Further studies must be performed in order to confirm these effects. 

Supported by grant SECyT-UNC. We acknowledge contribution of Avendano C.

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. Extrinsice 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 ± 4 µm from placentas of 28, 60 and 114 days of gestation were used. TUNEL (+) nuclei were observed in placentals cells at the beggining, 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.

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 ± 4 µm 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 placentaion. According with the observations of the present study, the angiogenic events at the beggining 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.

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 mesenchyma and small and medium caliber blood vessels near the epithelium could be visualized. With HRLM, binucleated cells, some of them with numerous cytoplasmatic 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.
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: Ghr, (2nmol/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%, Ghr 23±9% and Ghr, 24±5%) and decreased in vivo fertilization indexes (75%; p<0.05 vs Ghr 93%, Ghr 92%). Ghrelin and/or Ant increased embryo delay and delayed embryonic development (% blastocysts: Ghr 41%, Ghr+Ant 29%, Ant 37%; p<0.05 vs Con 66% and Ghr, 63%). Exp2 (6-9 females/group): Ghrelin and/or Ant increased the % of females with embryonic loss (fewer fetuses than corpora lutea) (Ghr, 33%, Ghr 83%, Ghr+Ant 75%, Ant 67%; p<0.05 vs Con 13%) and with fetuses that had stopped growing (Ghr 67%, Ghr+Ant 50%, Ant 67%; p<0.05 vs Con 0% and Ghr, 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) ovarián 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 (Art) 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.
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 (SNO) on luteal regression on day 4 of lactating rats. The system ex vivo GC-SNO-ovary was used. The system was incubated in Krebs Ringer buffer at 37 °C, keeping the GC and ovary connected by NO. In separate compartments, PRL was added (10 μM) 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 F2α (PGF2α) by RIA and nitrites by the Griess method. At 240 min, the luteal mRNA expression of β3-HSD and 20α-HSD, PGF2α receptor, ON synthase inducible (iNOS) and bcl-2 (antiapoptotic and proapoptotic factor) were analysed by RT-PCR. ANOVA I followed by Tukey 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.

**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 (SNO) on luteal regression on day 4 of lactating rats. The system ex vivo GC-SNO-ovary was used. The system was incubated in Krebs Ringer buffer at 37 °C, keeping the GC and ovary connected by NO, in separate compartments. PRL was added (10 μM) 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 F2α (PGF2α) by RIA and nitrites by the Griess method. At 240 min, the luteal mRNA expression of β3-HSD and 20α-HSD, PGF2α receptor, ON synthase inducible (iNOS) and bcl-2 (antiapoptotic and proapoptotic factor) were analysed by RT-PCR. ANOVA I followed by Tukey 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 TRANSGENICS MICE**

Ratner LD, González B, Huhtaniemi I, Poutamen M, Calandra RS, Rulli SB. IByME, Argentina; Turku University, Finland; Imperial College London, United Kingdom

Transgenic mice hypersecreting β-subunit of human chorionic gonadotrophin hormone (hCGβ+) produce elevated levels of hCG and prolactin. In addition, hCGβ+ 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. hCGβ+ female mice were injected with cabergoline (500 μg/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 hCGβ+ female mice partially reestablished the estrous cycling capacity throughout the period observed, whereas hCGβ+ 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 hCGβ+ female mice. In conclusion, cabergoline treatment reverses the acyclicity and infertility hCGβ+ mice.
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 steroidogenenic 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 (1x10^6 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.

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 dilucidate 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.

358. **RE24 - ODF1, LABELLED BY MONOBROMO-BIMANE, IS A SPERM FLAGELAR PROTEIN, ALSO LOCALIZED IN KIDNEY MEDULLA**


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 capacitación 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. mBBr specifically bound to N-terminal domain cysteine of ODF1. mBBr reduce rat and human sperm motility, quantitatively and qualitatively, and the effects are dose dependent, without significal 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 LYSOSONAL PROTEINS IN RAT EPIDIDYMIS**

Carvelli L, Malossi E, Aguilera AC, Bannoud N, Croce C, Sosa MA. IHEM–CONICET–ICB–UNCuyo. E-mail: lorenecarvelli@gmail.com

The epididymis is involved in sperm maturation through the secretion of factors into the lumen; some of them are glycan-modifying enzymes that catalyse 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 sperm were obtained by slicing of the tissue and centrifugation. We measured the activity of the glycosidases, βN-acetil-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.

360. **RE27 - CHANGES IN THE MEMBRANE OF BULL SPERMATOZOA DURING EPIDIDYMAL MATURATION**

Aguilera AC, Carvelli L, Bannoud N, Sosa MA. Lab. Biol Fisiol Celular. IHEM–CONICET. ICB (UNCuyo)- Mendoza. Argentina E-mail: caguilera@fcf.uncu.edu.ar

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 dilucidate 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.
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, Rabflilina 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, were 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.

Hystricomoroph rodents are particularly well represented in South America; among them, Chinchilla spp is threatened and the presence of remaining colonies in the Argentinian 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 percent ages 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.

Supported by grants from SECyT-UNC, CONICET, FONCyT.
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.

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 localization 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.
RE38 - SEASONAL VARIATIONS IN mRNA βLH FROM MALE TOAD Rhinella arenarum (AMPHIBIA, ANURA)
Volonteri MC, Ceballos NR.
Lab Endocrinología Comparada, DBBE, FCEyN, UBA. E-mail: claravolonteri@bg.fcen.uba.ar

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 βLH mRNA to measure its normal values in each reproductive season. For this, adult males of R. arenarum were collected along the year. For βLH mRNA analysis, total RNA from each pituitary was extracted and selected. LH semi-quantified by RT-PCR. Specific primers were designed from previous sequenciation analysis. The full βLH sequence (JN031567) is 93% homologous with the βLH 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 βLH by negative feedback.

RE39 - PENTOSE PHOSPHATE PATHWAY CONTROL IN PORCINE OOCYTE IN VITRO MATURATION
Ferretti E, Alvarez GM, Dalvit GC, Cetica PD.
Biochemistry, INITRA, School of Veterinary Sciences, UBA.

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.

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 adenylyl 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 μM) was used as an ACm inhibitor. Capacitation was evaluated by the chlorotracycline 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±2.38 μLO₂/h/10⁸ sperm vs 14.76±2.80 μLO₂/h/10⁸ sperm with heparin/2’,5’-dideoxyadenosine and heparin, respectively. Sperm viability was similar in both treatments (50±2.15% vs 53±3.86%). Heparin requires ACm activity to modulate oxidative sperm metabolism and capacitation induction.

RE41 - ENERGY METABOLISM ENZYMES: CAPACITATION AND ACROSOME REACTION IN CRYOPRESERVED BOAR SPERMATOZOA
Satorre MM, Breininger E, Beconi MT.
INITRA-Química Biológica-FCV-UBA. E-mail: msatorre@vet.uba.ar

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 IDH (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 concentrations 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 oF EGF/HR RATS
Pennacchio GE1, Carreño NB1, Jahn GA2, Valdez SR2, Soaje, M1,2
1IMBECU-CONICET-CCT-Mendoza, Argentina, 2ICB, UNCuyo.

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, PRLR[long]), progesterone (PR total and PRB isofrom), estrogen (ERα isofrom) and thyroid hormones (TRα1, TRα2, TRβ1, TRβ2 isofroms) 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 PRLR, ERα, TRα1, TRα2, TRβ1 and TRβ2 without changes in the expression of PRs (total or B isofrom). These results indicate that hypothalamic PRLR, ERα, 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 MC1, Martínez RA1, Bellingeri R1, Motta C1, Alustiza F1, Picco N1, Moliner D1, Busso L1, Vivas B2.
Anatomia Animal, EAV, UNRC.

Early Pregnancy Factor (EPF) is a molecule with immunosuppressors and growth factor activities involved in pregnancy. The main of this study was to quantify EPF in serum of pregnant sows and porcine placentas from 30 (n=6), 60 (n=5) and 90 (n=4) dp were used (Río Corts, 50, 000g), supernatant filtered (0.22 μm), aspirated from 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 μM sodium nitroprusside or 100 μM detanonoate, NO donors, at 35°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.

375. RE47 - SILDENAFIL AND L-NAME: EFFECTS ON PLACENTAL EFFICIENCY IN MICE. PRELIMINARY RESULTS
Motta C1, Picco N1, Alustiza F1, Grosso C1, Bellingeri R1, Busso L1, Moliner D1, Vivas A1, Romanini M2.
FAV, UNRC.

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 MATUREATION AND MODULATION OF NITRIC OXIDE PRODUCTION
Morado SA1, Cetica PD1, Montagna DR1, Beconi MT1, Dalvit GC1.
Química Biológica, INITRA, FCV, UBA.
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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 μM sodium nitroprusside or 100 μM detanonoate, NO donors, at 35°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 Bioquímica Clínica, CIBICI-CONICET, Fac Cs Qumicas, UNC. 2Ctro Leyria J1, Aguirre S1, Fruttero L1, Carlini C2, Rubiolo E1, Canavoso
378.  RE50 - ROLE OF ACID HYDROLASES DURING FOLLICULAR ATRESIA IN A VECTOR OF CHAGAS DISEASE
Leyria J1, Aguirre S1, Fruttero L1, Carlini C1, Rubiolo E1, Canavoso L1.
1Dpto Bioquímica Clínica, CIBICI-CONICET, Fac Cs Qumicas, UNC. 2Dpto Biofísica, UFRGS, Brasil.

In insects, unfavorable physiological, environmental or nutritional conditions induce changes in the ovarian tissue, which in turn elicited 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 Triatominie. 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-I, 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.  RE52 - SPERM SELECTION TECHNIQUES IN Llama glama FRESH SEMEN FOR IN VITRO EMBRYO PRODUCTION
Santa Cruz R1, Carretero MI1, Arraztoa C1,2, Miragaya M1, Giuliano S2.
Cátedras de *Teriogenología* y *Física Biológica,* 1INITRA, 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 assess 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 these 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.
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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.

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 secochiliolilquinone 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.

USE OF PSYCHOTROPIC IN MENDOZA
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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 off each branch were transcript 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.

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.