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Subjects



Education

Environment

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LECTURES AND SYMPOSIA

A1

BIOETHICAL CHALLENGES AGAINST TECHNOLOGICAL ADVANCES IN REPRODUCTIVE BIOLOGY

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Bioethics is a space for study and reflection in the construction of positions based on unstoppable scientific and technological advances. Thanks to the advances of science, solutions have been discovered for numerous problems of humanity with clear positive consequences, but also, there have been situations with undoubted bioethical objections. Given these challenges and dilemmas, bioethics analyzes the cost and benefit of each advance. The question arises, "Is everything technically possible ethically acceptable?" The bioethics debate about the human right to decide about your body, sexuality, reproductive rights, how to face the disease, aging, and the right to die with dignity. It provides tools and builds a regulatory framework on which laws can be generated to achieve general welfare within the framework of freedom. Reproductive biology exhibits numerous bioethical challenges such as assisted fertilization, gamete manipulation, and genetic editing. The genetic edition of human embryos using the CRISPR Cas9 technique has caused a scientific revolution. CRISPR CAS 9 means short, palindromic, grouped and regularly interspaced repetitions. The genetic edition of human embryos raises the possibility of avoiding diseases of genetic transmission, improves xenotransplants, improves the quality of milk and meat, but also, the possibility of changing the genome and choosing traits special features is very risky. Many scientists fear eugenics (good origin) or the application of the biological laws of inheritance to the perfection of the human species. So, ¿are all possible applications of CRISPR Cas 9 known today? What are the limits? There is still much to define, for example, the ecological risk of altering natural evolution and use in biological terrorism. An impasse is required to reflect on the consequences and where we want to go as a species.

A2

SEXUALLY TRANSMITTED INFECTIONS: WHAT S NEW ABOUT THE CONTROL OF CHLAMYDIA TRACHOMATIS

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Chlamydia trachomatis (Ctr) is the most common bacterial cause of sexually transmitted infections (STIs). The World Health Organization (WHO) estimates that 131 million are infected each year, mainly young people of reproductive age. In women, Ctr causes cervicitis, endometritis, salpingitis, which frequently persist along time leading to serious complications such as pelvic inflammatory disease, spontaneous abortions, and tubal infertility. Newborns, when infected in the birth canal, can develop conjunctivitis and pneumonia, whereas men can suffer urethritis, prostatitis, and epididymitis. Ctr is the main cause of preventable blindness or trachoma worldwide. There was an epidemiological alert in Argentina in August 2018 for the appearance of venereal lymphogranuloma. The asymptomatic nature of most of the infections makes diagnosis and treatment difficult. Besides, the lack of a preventive vaccine and the antibiotic resistance increase both reveal the need for new tools for the prevention and control of chlamydial infections. Ctr invades cervical epithelial cells through numerous receptors, many of them glycosylated, and survives and multiplies intracellularly in a vesicle called inclusion. We have shown the release of a glycan-binding protein, galectin 1 (Gal1), in cervical tissues under inflammation. This lectin engages glycosylated bacterial proteins, like MOMP (Major Outer Membrane Protein) and OmcB, to glycosylated cervical epithelial cell receptors such as PDGFR and various integrins. Acting as a bridge between bacterial and eukaryotic glycans, Gal1 promotes invasion, increasing not only the number of infected cells but also the number of inclusions per cell and the number of bacteria per inclusion. Lactose, glycanases, or neutralizing antibodies against glycosylated receptors decrease the magnitude of chlamydial infections. In agreement, mice KO for complex N-glycan-forming enzymes and Gal1 are less susceptible to infection. These findings suggest that hijacking bacterial glycan-Gal1-glycosylated receptors bridge could be a new tool to prevent cell invasion and overall Ctr infection. Once inside the cell, Ctr avoids its degradation in the phagocytic pathway by hijacking Rab proteins, the main controllers of intracellular transport. By bacterial-driven mechanisms, certain Rabs are recruited to the chlamydial inclusion while others are excluded. We have described that Ctr intercepts Rab14-mediated transport not only to evade fusion with lysosomes but also to acquire sphingolipids synthesized at the Golgi apparatus. Molecular mechanisms underlying how these bacteria manipulate intracellular transport are a matter of intense study. We demonstrate that Ctr provokes Akt phosphorylation along its entire developmental life cycle and recruits phosphorylated Akt (pAkt) to the inclusion membrane. As a consequence, Akt Substrate of 160 kDa (AS160), also known as TBC1D4, a GTPase Activating Protein (GAP) for Rab14, is phosphorylated and therefore inactivated. Phosphorylated AS160 (pAS160) loses its ability to promote GTP hydrolysis, favoring Rab14 binding to GTP. Akt inhibition by an allosteric isoform-specific Akt inhibitor (iAkt) prevents AS160 phosphorylation and reduces Rab14 recruitment to chlamydial inclusions. iAkt further impairs sphingolipids acquisition by Ctr-inclusion and provokes lipid retention at the Golgi apparatus. Consequently, treatment with iAkt decreases chlamydial inclusion size, bacterial multiplication, and infectivity in a dose-dependent manner. Similar results were found in AS160-depleted cells. By electron microscopy, we

observed that iAkt generates abnormal bacterial forms as those reported after sphingolipids deprivation or Rab14 silencing. Taken together, our findings indicate that targeting the Akt/AS160/Rab14 axis could constitute a novel strategy to limit chlamydial infections, mainly for those caused by antibiotic-resistant bacteria.

A3

A MOLECULAR APPROACH TO FEMALE AND ASSISTED REPRODUCTION

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In many modern societies, the proportion of women who delay childbearing beyond the age of 35 years has increased greatly in recent decades. They are falsely reassured by the popular belief that advances in new reproductive technologies can compensate for the age-related decline in fertility, but science cannot beat the biological clock yet. Age is the single most important determinant of female fertility, either natural or treated, and it mainly impacts the quality of the oocyte. Even though the advances in science to know the factors that determine oocyte quality are constantly growing, there is still much to discover. Our group is interested in characterizing the cortical reaction, a process in which cortical granules fuse with the plasma membrane to avoid polyspermy. Thus, the cortical reaction is the only defense mechanism that the oocyte has to prevent the penetration of two or more sperm cells and guarantee the development of the preimplantation embryo. Using techniques of molecular biology, indirect immunofluorescence, live imaging, transmission electron microscopy, and animal models, we are contributing to the characterization of the cortical reaction and the biology of cortical granules. Based on our findings, we propose that cortical granules are fundamental organelles in determining oocyte quality.

A4

NANOTECHNOLOGY AND ITS IMPACT IN THE HEALTH SCOPE

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Nanotechnology is the study, development, and manipulation of systems at a nanometric scale. Furthermore, nanomedicine is the branch of medicine that takes advantage of nanotechnology knowledge in health care procedures. Nanomedicine is primarily concerned with the study of three fields of application of nanotechnology to the biomedical field: regenerative medicine, diagnosis, and treatment of different pathologies. A newly emerging field of application that arises from the combination of two of the traditional areas mentioned is teragnosis, where nanotechnological tools are enhanced and complemented to achieve a therapeutic and diagnostic effect simultaneously. In general, the behavior of the matter in the nanoscale offers multiple advantages over traditional therapies and diagnostic methods. There are various types of nanostructures, both organic and inorganic, where their composition, morphology, and different specific physicochemical properties directly or indirectly influence their pharmacokinetic and pharmacodynamic behavior, and finally, on their biomedical application. Another essential aspect for taking into account when developing nanostructured systems for use in health is the study of several characteristics related to its biocompatibility and its nanotoxicity. Currently, few biomedical nanotechnological alternatives have been approved for commercialization, and most of them are oriented to their application in cancer disease. Despite this, the versatility of nanotechnology allows its application in practically all health fields. In particular, our laboratory is working on the search for new nanotechnological alternatives for cardiovascular therapy, with promising results so far.

A5

MYTHS AND TRUES ABOUT HEREDITARY BREAST CANCER

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Five to ten percent of breast cancer (BC) cases are inherited and they are associated with the inheritance of a gene that has a pathogenic variant and therefore does not fulfill its function. Hereditary (H) BC/Ovary (CO) Syndrome, caused by pathogenic variants in *BRCA1* and *BRCA2* genes, is the most common and the risk of BC in individuals carrying pathogenic variants in these genes can reach values of up to 80% throughout life. Numerous myths about HBC generate confusion and uncertainty regarding the real risk of developing this disease. Cancer genetic counseling (CGC) identifies people who are at risk of developing BC. Based on the calculated risks, it is recommended to carry out molecular studies, especially of the high penetrance genes *BRCA1* and *BRCA2*, to determine the existence of germLine variants. CGC allows us to eliminate the myths about HBC, some of which are: (1) My BC is not H if I do not have relatives who have had this disease. (2) I should not perform genetic studies if my BC relatives belong to the paternal branch. (3) My risk for developing BC is low because I do not have *BRCA1* and *BRCA2* pathogenic variants. (4) As I already had BC, it makes no sense to carry out the genetic study even if I have clinical criteria of suspected HC. (5) Men of a family with a known pathogenic variant should not be tested.

A6

IMPLEMENTATION OF THE TICs, ARTIFICIAL INTELLIGENCE, BIG DATA AND ANALYSIS OF BIOMEDICAL IMAGES IN THE ANDROLOGICAL DIAGNOSIS. TOWARDS A SCIENTIFIC EVIDENCE-BASED MEDICINE

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The emergence in recent years of new strategies of information and communication technologies (ICTs), artificial intelligence (AI), and big data (BD) has allowed its implementation in multiple contexts of human development. Among the most important is health. Currently, we can access constantly updated sources of information, so the machine learning process is inexhaustible. The integration of Health Sciences with the development of ICTs in combination with Medical Informatics and its application in the different activities of the health sector made it possible to conceptualize the term of e-health, which is defined as the use of ICTs to provide services for diagnosis, treatment, and prevention of diseases, as well as research, I+D, evaluation, and continuing education, all for the development of health. Its applicability has been seen in a large number of specialties within medicine, including Psychiatry, Dermatology, Cardiology, Radiology, Biochemistry, etc. Semen analysis is the first diagnostic tool to evaluate the male factor in an infertile couple. For many years, its evaluation was carried out through manual, subjective methods, which produced highly variable and operator-dependent results. The incorporation of methods that use mathematical algorithms, AI, and biomedical image analysis has allowed standardizing semen analysis. The Teleanalysis and Translational Research Laboratory (LaTIT) is the first link of the andrological observatory, which through the use of mathematical formulations and computational algorithms plus image analysis, quantifies sperm parameters of concentration, motility, vitality, morphology and DNA fragmentation regardless of the geographic location of the patient. The incorporation of this new technology ensures reliable and reproducible results in the measurement of the different parameters of the spermogram that will bring significant improvements in the andrological diagnosis directed towards a medicine based on scientific evidence.

A7

TECHNOLOGICAL IRRUPTION... EDUCATIONAL DISRUPTIONS?

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REDIRECTING THE JOURNEY So much has been said, written, spoken, produced, reflected on the digital technologies impact in the school environment that it may be time, at least for this writing, to order according to personal and situated criteria, some points of the reflective happening on the subject. The educational processes have gone from the solid to the liquid, from the earth to the water, transit between spaces and times without asking permission, without being framed by structures, the walls of the educational institutions have fallen and the learning is broken down between the inside and outside. Digital residents are the inhabitants of learning networks that are configured as maps with real nodes in virtuality, games, emotions, curiosity, and creativity become their luggage; and they walk through asking, experimenting, making mistakes and redoing as many times as necessary, such as the steps they take to reach their destination... It will be necessary, then, to think about these relationships in terms of social, educational, and digital tensions. Today, Bauman's metaphor could be resumed regarding "liquid society" and in the educational space, about the possibility of "liquid pedagogy" thinking that sustains epistemic distrust as unique, immovable and static paths, as opposed to the metaphor of water as an element of fluidity, dynamism, and the possibility of permanent change. From this framework, this notion of the "liquid" is linked to the concept of emerging pedagogies, such as those concepts and practices that are still unfinished, begin to be seen as necessary to get under way with the objective of making visible new practices and new categories in Teaching and learning processes. The impact of technological irruption on educational practices should be analyzed. We will seek to "redirect the trip" hack education and turn it upside down. How is the fall of the walls, seamLess learning and disruptive methodologies such as gamification and inverted class to cause learning? In short, does technological irruption promote educational disruptions?

A8

STRATEGIES TO IMPROVE TEACHING/LEARNING IN THE CLASSROOM

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Current times demand new pedagogical teaching and learning strategies in the classroom. Biological Sciences Students Teacher must be known that scientifically seizing the object of study not only requires disciplinary knowledge, other than learn to use them in different work contexts and with real problems. This requires an open teacher formation who can listen to the daily knowledge (that this learning subject carries out). In this sense, there are several academic stories that it is necessary to know in order to provide, not only a knowledge epistemology for his university career, but for the student himself challenges the old with the new knowledge for its professional training. The topic presentation goal reflects and discusses 10 years of pedagogical experience in an imperative search for teaching strategies to improve classroom education and learning in Courses of the Teaching Career in Biological Sciences.

Experience development begins in 2008 when starting the pedagogical task in the courses Human Body I and II of the Career of Biological Sciences Teachers of the Faculty of Exact Sciences of the National University of La Pampa (UNLPam). Some questions and challenges will be going through in these years like:

- 1- How to move from a model of traditional and memorial teaching towards a socio-constructivist model?
- 2-How to overcome the fragmentation of disciplines towards an interdisciplinary view?
- 3-What mediators for learning will be necessary to carry out the change?
- 4-What is the place of action research in these processes?

A9

ECO-EPIDEMIOLOGY: EXAMPLE LEISHMANIASIS

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Eco-epidemiology is a theoretical framework by which events of interest for collective health are approached from different disciplines and attending to multiple levels of time and space. It deals with the physical and biological properties and interactions studies by the ecology, but also with the social determination from the population to the individual that the term "ecological" refers to in epidemiology. We will present leishmaniasis as an example of this disciplinary approach, a parasitic vector-borne disease, so the possibility of their transmission is intensely modulated by the environment and the dynamics of vectors and reservoirs, but at the same time, precisely because of anthropogenic modifications of land use and climate, demographic and market changes, the probability of generating an epidemic outbreak is fundamentally socio-economic and cultural. In order to do this, the "driver" factors of epidemics must be discriminated at different scales, from the investigation of continental-regional models (macro-scale) to the study of epidemic focus (meso-scale) to the survey of the transmission site (micro-scale). Each of these levels corresponds to different objectives and consequently generates evidence for different strategic approaches to make political decisions (macro-scale), programmatic decisions (meso-scale), and operational decisions (micro-scale).

A10

REMOVAL OF TOXIC METALS USING AS BIOADSORBENTS NON-VIABLE BACTERIAL BIOMASS AND INDUSTRIAL WASTES

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Al^{3+} , Cu^{2+} and Zn^{2+} are emerging pollutants found in soils and industrial and domestic wastewater. Removal of these metals from soils would contribute to the better development of horticultural crops, while, the treatment of aqueous effluents, would lead to obtaining quality recycled water suitable for the irrigation of these crops. The removal of toxic metals can be carried out through the biosorption process, in both discontinuous (batch) and continuous (fixed-bed column) modes of operation, using biological materials capable of retaining metal ions on their surface. Non-viable biomass of *Pseudomonas putida* A (ATCC 12633) entrapped in agar-agar beads is able to attach Al^{3+} , Cu^{2+} , and Zn^{2+} binding them to -NH, -OH, -COOH, -CO and -PO groups on their cellular surface. With these beads, in batch mode, the adsorption equilibrium was reached at 45 min, 4 h and 6 h, for Al^{3+} , Cu^{2+} and Zn^{2+} , respectively, showing a maximum sorption capacity (q) of 0.09 mg Al^{3+} /g beads, 0.270 mg Cu^{2+} /g beads and 0.102 mg Zn^{2+} /g beads. Adsorbent efficiency was stable, at least, until 8 successive cycles of adsorption/desorption. For the continuous mode assays, 47 g of beads were packed in a glass tube with an inner diameter of 2 cm. Aqueous solutions at pH 4.3 with different Al^{3+} , Cu^{2+} and Zn^{2+} concentrations (6-210 mg/L) were filter through the fixed bed column at different flow rates (0.5-1.5 mL/min). The fixed-bed column showed q values between 0.15 and 0.20 mg metal/g beads and the removal percentage was close to 70% for the three evaluated metals. Industrial wastewater supplemented with 30 mg/L of Cu^{2+} and Zn^{2+} (simulated effluent) were filtered through the fixed-bed column at a flow rate of 0.5 mL/min. q value was 0.15 mg metal/g beads and the removal percentage was close to 66%. The fixed bed column was stable, at least, until 12 successive cycles of adsorption/desorption. In all conditions evaluated, complete desorption of metals was achieved with HCl 0.01 N. When was used as alternative adsorbent the barley bagasse, low cost waste from brewing industry, in batch mode the adsorption equilibrium was reached at 6 h for Al^{3+} and Zn^{2+} and at 16 h for Cu^{2+} . This adsorbent showed q values significantly higher respect to obtained with beads with biomass trapped (3.57 mg Al^{3+} /gr bagasse, 8.33 mg Cu^{2+} /gr bagasse, and 0.518 mg Zn^{2+} /gr bagasse). The results, as a whole, show the high efficiency of the beads with non-viable biomass of *Pseudomonas putida* A (ATCC 12633) trapped and of the barley bagasse to attach Al^{3+} , Cu^{2+} and Zn^{2+} , and support the notion of their potential use for the removal of these metals from soils and industrial and domestic wastewater containing them.

A11

EFFECT OF AMBIENT LIGHTING ON LIFE QUALITY IN DIFFERENT ECO-REGIONS OF JUJUY PROVINCE

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The population of Jujuy province is distributed in four eco-regions: Yungas, Valley, Quebrada, and Puna, resulting in a variety of ecosystems and ambient factors that could affect them. In this study we try to show a relationship between light-ambient-and life quality, considering the interaction of different weather variables that determine our performance and wellbeing. The main goal is to establish the relationship between natural lighting and life quality using the WHOQOL-BREF test. The photoperiod continuous data recompilation (twilight, relative heliophany) and light intensity, in different eco-regions of Jujuy province, show the highest values in summer solstice when compared to winter, without finding a latitudinal significant difference. The WHOQOL-BREF test, which considers four dimensions: physical and psychological health, social relationship, and ambient, showed psychometry resolutive power by seasons. Statistical analysis (variance, Tukey's test, correlation coefficient) did not show differences between spring-summer and winter-autumn, according to the data obtained in the measured photoperiod and maximum intensity of lighting. The trend indicates that the values of all studied variables decrease towards the winter solstice (with lower values at higher altitudes) and increase towards the summer solstice (higher values at higher altitudes), according to the expected chronobiological models.

A12

ENVIRONMENT AND EMERGING INFECTIOUS DISEASES

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Emerging infectious diseases are those that arise at a certain time and place and threaten to become a new epidemic. That is: new diseases, existing diseases but not yet recognized and known diseases that are expanding their area of geographic expansion. Among the factors that cause the emergency, are very important those related to the environment: climate change, land use, water quality, intensification of agriculture, loss of biodiversity, changes in social behaviors, mobility and transport, urbanization, etc. Among the best known emerging infectious diseases are Ebola, hantavirus, avian influenza, dengue, SARS, and bovine spongiform encephalopathy. So, not only changes or adaptations of infectious agents contribute to the emergence of new diseases, changes in society or the environment, many caused by the human being, can also be the origin of new diseases whose control requires understanding their genesis and their forms of transmission.

A13

RATIONAL DESIGN OF BIOCOMPATIBLE FORMULATIONS OF NEW ANTIFUNGALS FOR APPLICATION IN SUSTAINABLE AGRICULTURE

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(ICYTAC- CONICET)

Botrytis cinerea (*B. cinerea*) is a fungus that causes gray mold. This is a disease that induces the decay and deterioration of grape berries by altering its chemical composition. The organoleptic properties, such as the taste and color of wine, are affected. Its control is difficult because the fungus is genetically variable and has developed strains resistant to many of the chemicals in the last years. In addition, the indiscriminate use of fungicides has increased the development of microbial resistance in medicine and agriculture. Hence, it is essential to discover new low toxic substances. Based on this, a comprehensive study has been carried out in our working group. It includes the synthesis of new antifungals, the design of biocompatible formulations, the evaluation of their bioactivity against *B. cinerea*, and the study of the mechanism of action of new entities, through molecular modeling. A family of compounds was accomplished by the arylation of the commercial fungicides triadimefon (1) and cyproconazole (2), via two different methodologies ($S_{RN}1$ -Stille reaction or Suzuki reaction). The bioactivity was proved against *B. cinerea*, isolated from infected grapes in the province of San Juan, Argentina. The results showed that the monofluorinated compound (3) was the most active and turn levels of cytotoxicity were comparable to commercial fungicides. Furthermore, molecular modeling studies demonstrated that this derivative displayed interactions with the active site of the enzyme very similar to those observed with 2 and voriconazole. These results encouraged us to continue the study in search of the ideal, more effective, and less toxic agrochemical than commercial ones. For this purpose, two strategies were studied. One of them was the combination of synthetic antifungal (3) with citral, an essential oil of recognized activity against *B. cinerea*. The other one was the design of biocompatible formulations of derivative 3 using chitosan as a biodegradable polymer. Cytotoxicity studies are currently underway, with the aim of confirming whether these formulations are less toxic than their precursors. This work constitutes a contribution to the redesign of more effective and safer agrochemicals for their potential application in sustainable agriculture.

ABSTRACTS

GENERAL, CELULAR AND MOLECULAR BIOLOGY

A14

SORTILIN KNOCK-DOWN AFFECTS THE PROCESSING AND EXPRESSION OF CATHEPSIN D IN EPIDIDYMAL CELLS

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The intense secretory activity of epididymal cells contributes to sperm maturation. Lysosomal enzymes are highly secreted by the epithelium into the epididymal lumen, and they can participate in remodeling the sperm surface during their maturation process. In most eukaryotic cells, the intracellular transport of these enzymes is mostly regulated by mannose 6 phosphate receptors (MPRs). The lysosomal protease cathepsin D (CatD) is mostly transported by MPRs, although the receptor sortilin (Sort) has also been implicated in this transport since CatD is complexed with prosaposin (Psap), the natural ligand for Sort. In this study, we evaluated the incidence of the Sort expression on transport and processing of CatD in cultured epididymal cells. In a rat epididymal cell line (RCE-1), the sortilin knock-down was induced by transfection with a sortilin pSilencer. Then, we observed by western blot that this Sort silencing increases the expression of other proteins, such as the cation-dependent MPR (CD-MPR) and Psap. In turn, CatD is decreased under these conditions, where the immature form of the enzyme (proCathepsin D) prevailed. Meanwhile, a similar distribution of wild type RCE-1 was observed by IFI. These preliminary results suggest that proCathepsin D could be transported by sortilin and CD-MPR alternatively, but its processing is affected by the silencing of sortilin in this cell type.

A15

SALVIGENIN AFFECTS THE PROLIFERATION AND ULTRASTRUCTURE OF TRYPANOSOMA CRUZI EPIMASTIGOTES

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Trypanosoma cruzi is the causal etiologic agent of Chagas disease. In cultures, this parasite is mainly found in the epimastigote form and a low percentage in the infective form trypomastigote. The current chemotherapy against *T. cruzi* is insufficient because the available drugs, Nifurtimox and Benznidazole, have limited activity, and show toxic side effects in patients. Therefore, the "screening" of purified molecules from natural sources, mainly plant leaves has become an important tool for the fight against Chagas disease. Many natural compounds, extracted from plants native of Argentina, have been shown to be effective against the parasite. Among them, flavonoids are an important family of molecules that have been widely studied. In this work, we analyze the effect of the natural flavonoid Salvigenin (SVG) isolated from *Baccharis scandens* on the growth of *T. cruzi* epimastigotes (strain Dm28c). SVG showed an antiproliferative effect on epimastigotes, even at low concentrations. This effect was irreversible even in the short term of exposure to the compound. SVG significantly decreases the mitochondrial activity of the parasites, at all the concentrations tested (1, 5, and 10 µg/mL). This alteration is related to changes in ROS levels observed with the treatment. When we analyze the ultrastructure of the parasites, we observed disorganization of the cytoplasm at a general level, an increase in cytoplasmic vacuolization. Also, the presence of structures that appear to be like "membrane blisters" is highlighted. From these results, it is necessary to identify the molecular targets of the parasites for the action of this compound and to determine if SVG can affect the life cycle of *T. cruzi*.

A16

XANTOMICROL MAY AFFECT THE PROLIFERATION OF TRYPANOSOMA CRUZI EPIMASTIGOTES BY MULTIPLE MECHANISMS

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Trypanosoma cruzi is the causal etiologic agent of Chagas disease. In cultures, this parasite is mainly found in the epimastigote form and a low percentage in the infective form trypomastigote. The current chemotherapy against *T. cruzi* is insufficient because the available drugs, Nifurtimox and Benznidazole, have limited activity, and show toxic side effects in patients. Therefore, the "screening" of purified molecules from natural sources, mainly plant leaves has become an important tool for the fight against Chagas disease. Many natural compounds, extracted from plants native of Argentina, have been shown to be effective against the parasite. Among them, flavonoids are an important family of molecules that have been widely studied. In this work, we analyze the effect of the natural flavonoid Xantomicrool (XML) isolated from *Baccharis scandens*, on the growth of *T. cruzi* epimastigotes (strain Dm28c). XML showed an antiproliferative effect on epimastigotes, even at low concentrations. This effect was irreversible, but a minimum exposure of 6 h to the compound was necessary. XML affects the mitochondrial activity of the parasites, at all the concentrations tested (1, 5, and 10 µg/mL). Changes in ROS levels were observed with the treatment. When we analyzed the ultrastructure of the parasites, we observed disorganization of the cytoplasm and an increase in cytoplasmic vacuolization. Also, the presence of structures that appear to be like "membrane blisters" is highlighted. From these results, it is necessary to identify the molecular targets of the parasites for the action of this compound and to determine if XML can affect the life cycle of *T. cruzi*.

A17

CEREBELLUM-DEPENDENT MOTOR AND COGNITIVE ABILITIES ARE REDUCED IN AN AGING RAT MODEL

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As we age, movements become slower and inconsistent and require more attention. The ability to control movement is crucial. Therefore, in order to succeed in promoting longer functional independence, we need to understand the normal aging process of motion control. Such an aging engine is obviously multifactorial and complex. However, one of the specific structures that may have a predominant role in understanding this process is the cerebellum. Previously, we observed Bdnf expression oscillates on a circadian basis in the young rat cerebellum. Interestingly, aging reduces the rhythm's amplitude and phase shifts its acrophase. Here, we examine the evidence supporting the hypothesis that motor aging causes alterations in functions such as motor coordination, motor learning, and balance, at the time when the locomotor activity starts in nocturnal rats and following the maximal expression of Bdnf. We evaluated these parameters through the Single Reaching Pellet (SRP), which determines performance in motor coordination and motor learning, and Beam Walking (BW) Tests, which establishes the performance in balance. The tests were performed by young (3 month-old, n = 11), and old (22 month-old, n = 5), Holtzman rats maintained under 12 h light:12 h dark and constant temperature conditions as well as fed with a regular chow diet and water ad-libitum. The statistical tests used in this study were *t*-test in the case of the behavioral tests, and one-way ANOVA followed by Tukey's post-hoc test, ChronosFit and Cosinor, in the case of Bdnf expression. We observed that the percentage of total success rate and the quality of the first attempt during the performance are significantly lower in old rats compared to young rats ($P = 0.01$) in SRP. Better performance of cognitive and behavioral tests performed by and observed in young animals at the end-of-the-day-beginning-of-the-night, follows the maximal expression of the neurotrophic factor, Bdnf1 at CT 05:39 ± 00:09. In the case of the BW test, we observed that the old rats presented higher numbers of foot slip as well as falls than the young group ($P < 0.05$), probably, because the test was conducted at a time of the day following the nadir of Bdnf rhythmic expression. Understanding the cognitive role of the cerebellum during aging has the potential to offer insights into the origins of cognitive deficits in some motor functions, which are often underestimated, misunderstood, and not treated.

A18

ANALYSIS OF ANTI-HER2 DRUG INTERACTION IN HER-2 POSITIVE HUMAN BREAST CANCER CELLS: SK-BR-3 AND BT-474

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Breast cancer (BC) is the most common malignancy among women all over the world. Around 25% of all BC overexpress the human epidermal growth factor receptor 2 (HER2) and it is traditionally associated with worse prognosis, shortened disease-free survival, and overall survival. Trastuzumab (TZ) is the standard adjuvant treatment for this type of patient. It is well known that TZ has survival benefits when associated with chemotherapy in the treatment of patients with early operable and metastatic HER2 positive BC. Although TZ therapy has brought significant clinical benefits, not all patients respond. Moreover, the vast majorities of responders eventually relapse and suffer heart diseases too. So, TZ resistance and cardiotoxicity are clinically important problems. Thus, it is necessary to develop new therapeutic approaches based on the combination of different existing and FDA's approved drugs. In this study, we evaluated the effect of different concentrations (0.1–10 µg/mL) of anti-HER2 therapies such as TZ, Trastuzumab-emtansine (TD) and Lapatinib (LP), alone and in combination, to determinate their effect on the cell proliferation and to evaluate synergistic effects between drugs in two breast cancer cell lines: SKBR3 and BT-474. To study drug interactions, we

performed MTT assays and to evaluated synergism and antagonism between drugs, we used Computer Software Program CompuSyn. Our results indicate that 72 h-treatments with different doses of TZ, TD, LP, and their combinations are effective decreasing SKBR3 and BT-474 cell proliferation, and LP has the greatest effect decreasing it. The SKBR3 cells are more sensitive to treatments than BT-474 cells. The analysis of pharmacological interaction in SKBR3 cells showed synergism in the combination TZ+LP (10:1 µg/mL) and TZ+LP (10:10 µg/mL), and in these concentrations, is possible a 10-fold TZ reduction achieving the same therapeutic effect. Additionally, TD+LP exhibited synergism in all doses tested, and in TD+LP (10:1 µg/mL) and TD+LP (10:10 µg/mL), is possible a 10-fold TD reduction achieving the same therapeutic effect. On the other hand, in BT-474 cells, we observed synergism between TD+LP in all doses tested, and only in the lowest dose for TZ+LP (1:0.1 µg/mL). A favorable 10-fold TZ or TD dose reduction is possible for combination 10:1 µg/mL and 10:100 µg/mL in TZ+LP and TD+LP. In conclusion, the coadministration of anti-HER2 therapies with different mechanism action in patients with HER2 positive BC could contribute to improve their prognosis and reduce the adverse effects of therapy because the TZ or TD doses applied would be lower due to the adjuvant effect of LP.

A19

SYNERGISTIC ANTITUMOR ACTIVITY BY COMBINING RETINOIC ACID WITH FOCAL ADHESION KINASE INHIBITOR

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Breast cancer (BC) is a common malignant disease worldwide. Retinoids are currently being used in clinical trials to treat or prevent cancer progression and have been proposed as an adjuvant treatment of breast carcinoma because of its ability to inhibit cell growth. We have previously demonstrated that long retinoic acid (RA) treatment (10^{-6} M) reduced cellular adhesion and migration in BC cells. In addition, we verified that the administration of Trastuzumab (TZ) in combination with RA synergistically decreased cell survival, adhesion/migration/invasion in BC cells. TZ+RA strongly reduced Focal Adhesion Kinase (FAK) expression and induced nuclear FAK translocation. We speculate that RA+FAK inhibitor (FAKi) could reduce tumor growth and tumor formation by preventing tumor adhesion. We used the LM3 cell line, derived from a murine mammary adenocarcinoma, with tumorigenic and metastatic capacity in BALB/c mice treated or not with FAKi for 72 h. We performed an orthotopic assay evaluating LM3 tumor growth in the mammary gland of female BALB/c bearing or not a slow-release RA (10 mg)-containing subcutaneous silastic pellet or an empty pellet as control. RA and FAKi separately reduce the tumor growth, but the combined treatment induced a stronger inhibition in tumor volume. In addition, each drug seems to increase mice's survival, but only the combination of drugs is statistically significant. Furthermore, we also performed an experimental metastatic assay. Then LM3 cells pretreated or not with FAKi for 72 h were injected into the tail vein of mice bearing or not RA-containing pellet. RA significantly reduced lung metastatic dissemination. FAKi and the combination RA+FAKi presented a lower, but non-significant, number of lung nodules than the control group. In conclusion, the sensibility to RA therapies could be increased with FAKi coadministration in BC tumors.

A20

EFFECT OF BOTULINUM NEUROTOXIN OF MENDOZA *CLOSTRIDIUM BOTULINUM* STRAIN ON TUBULIN IN BREAST CARCINOMA CELLS (MCF 7)

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Botulinum neurotoxin serotype A (BoNT A), produced by *Clostridium botulinum*, causes botulism and is used to treat multiple diseases. Its potential action in cancer therapy is currently being evaluated. In precedent studies, we have shown that BoNT A from native soil strains (Su), they have different characteristics to that of the prototype A Hall strain such as; greater specific toxic activity (AE) and differences in molecular structure and enzymatic activity against SNARE proteins. In this work, we study the action of an autochthonous BoNT A compared to the A Hall on tubulin in the MCF-7 cell line of breast carcinoma. The autochthonous BoNTs of the strain Su 1935 (Tupungato) and A Hall were used in their native form, purified by saline precipitation. The values of AE (LD₅₀/mg protein) and electrophoretic characteristics under non-denaturing conditions were determined. MCF7 cells were cultured on coverslips and incubated with 250 and 500 LD₅₀ of the BoNTs for 10, 25, 45, and 90 min. Later, the cells were fixed and processed for immunodetection. As primary antibodies were used anti-tubulin or anti-Golgi 97 and as secondary anti-mouse-Alexa 488. The preparations visualized by fluorescence microscopy. At 90 min incubation with 250 LD₅₀, it was observed that ~90% of the cells were taken off and deformed by the action of the BoNT A 1935 and ~40% for the BoNT A Hall, while with 500 LD₅₀ both toxins were deleterious to the cells. When the cells were incubated with the toxins for 25 min, a disruption of microtubules with both toxins was observed, the effect being greater with the BoNT A1935. This effect was accompanied by a redistribution of the Golgi apparatus. Western blotting showed the shape of new tubulin bands, possibly due to protein degradation. This effect was also greater in the BoNT 1935. These results show a cytotoxic action of BoNT A with disorganization of cell microtubules, being observed with

greater intensity in the cells treated with the autochthonous BoNT A. The degradation of tubulin and its intracellular reorganization would be part of the deleterious action of this toxin on tumor cells, opening new perspectives for therapy against solid tumors.

A21

DIFFERENTIAL EXPRESSION AND LOCALIZATION OF BETA-CATENIN AND HSP27 AFTER CISPLATIN/DOXORUBICIN TREATMENT IN TRIPLE NEGATIVE BREAST CANCER CELLS

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The treatment of triple-negative breast cancers involves the administration of the conventional chemotherapeutic drug doxorubicin, given the lack of specific targeted agents. Novel therapeutic strategies, such as cisplatin, are currently being tested for these patients. Many studies have demonstrated that aberrant Wnt/ β -catenin signaling serves a role in the development of breast cancer, while others have concluded that abnormal regulation of the Wnt pathway induces tumor cell chemoresistance. The small heat shock protein 27 (HSP27) is overexpressed in human breast cancer cells. As a result, cancer cells may suppress apoptosis and develop resistance to antineoplastic agents, such as doxorubicin. The present study sought to examine the role of the Wnt/ β -catenin and HSP27 signaling pathway in response to cisplatin (CisPt)/doxorubicin (Doxo) treatment in human triple-negative (TN) breast cancer cell lines. Material and Methods: MDA-MB231 (TN) and MCF10A cell lines were used. Cell viability was measured using MTT assay and IC₅₀ values were obtained after 48 h of CisPt or Doxo exposition. β -catenin and Hsp27 gene expression were measured by qPCR. Total and active β -catenin, phospho ant total HSP27, phospho and GSK3 β , phospho and total p38 expressions were measured by western blot and immunofluorescence. 3D cell culture from MDA MB231 cells was treated with increasing concentrations of CisPt and Doxo for 48h. Results: MDA-MB231 cells showed higher IC₅₀ values for CisPt and Doxo than the MCF10A cell line. In MDA-MB231 cells, the expression of β -catenin, active β -catenin, total and phospho-GSK3 β , and total HSP27 significantly decreased in the CisPt group ($P < 0.05$). No changes were observed in the Doxo-treated group. In MCF10A cells, the expression levels of total and active β -catenin did not modify with CisPt treatment, but in the Doxo group, the proteins evaluated showed a tendency to increase. Also, in MCF10A Doxo treatment significantly decreased the expression of GSK3 β in comparison with control ($P < 0.05$). In contrast, CisPt administration significantly increased phospho-GSK3 β expression, respect to the control group ($P < 0.05$). Interestingly, in MDA-MB231 cells the nucleolus appeared disaggregated, and active β -catenin increased at this subcellular localization after CisPt and Doxo treatment. In contrast, total β -catenin was preferentially localized in the Golgi. On the other hand, 3D cell culture was more resistant to Doxo-treatment than 2D cell culture. CisPt induced a decrease in 3D cell culture growth. Conclusions: CisPt treatment was associated with decreased expression of β -catenin and HSP27. While, in Doxo-treated cells, as related to stable levels of β -catenin and increased expression of HSP27. The differential expression and localization of β -catenin and HSP27 could be related to a differential cellular response depending on the cytotoxicity mechanism of the chemotherapeutic agent used, which in turn affect the cell fate decision. Our preliminary data indicate that β -catenin and HSP27 may be potential therapeutic targets in TNBC.

A22

EFFECT OF A PPARGAMMA SYNTHETIC AGONIST ASSOCIATED WITH RETINOIC ACID ON THE 24-HOUR RHYTHMS OF BMAL1 AND ROR α PROTEINS IN AN EXPERIMENTAL MODEL OF ALZHEIMER'S DISEASE

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Alzheimer's disease (AD) pathogenesis is associated with cognitive deficit and alterations in the circadian rhythms. Recently, PPAR- γ agonists have shown neuroprotective effects in neurodegenerative disorders. Previous studies indicate a role of retinoic acid in cognitive activities and anti-amyloidogenic properties. Previously, we found that an intracerebroventricular (i.c.v) injection of A β (1-42) modified the daily rhythms of A β and BMAL1 in the rat temporal cortex (TC). Continuing with that study, the objectives of this study were: first, to analyze the effect of an i.c.v. injection of A β (1-42) peptide on the 24-h rhythms of ROR α protein levels in the rat TC; second, to evaluate the effect of the PPAR γ agonist, pioglitazone, along with the RXR ligand, retinoic acid, on those temporal patterns. Groups were defined as (1) control (saline solution), (2) A β -injected (A β aggregates-10 μ g), (3) A β -injected treated with Pio-RA (A β aggregates-10 μ g) and (Pio 10mg/kg, ip)/AR (1mg/kg, ip) by 15 days. Rats were injected into the lateral ventricle (coordinates: AP: -1 mm, L: 1.5 mm, and DV: -3.5 mm). TC samples were isolated every 4 h during a 24h period. A β , BMAL1 and ROR α proteins levels were determined by immunoblotting. To analyze the daily rhythmicity, 12 rats from each group were used. The data were analyzed by one-way ANOVA followed by Tukey's test, a $P < 0.05$ was considered to be significant. Daily rhythms were assessed by the Chronos-Fit software. We found that the injection of A β (1-42) modified the daily rhythms of ROR α protein level in the rat TC. The treatment of Pio-RA reestablished the daily rhythms of A β , BMAL1, and ROR α protein levels. These findings would emphasize the importance of Pio-RA in the modulation of the daily rhythmicity of clock genes in AD.

A23

SESQUITERPENE LACTONES AFFECT THE REDOX SYSTEM OF *TRYPANOSOMA CRUZI*

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Chagas disease is caused by *Trypanosoma cruzi* (*T. cruzi*) and affects millions of people worldwide, mostly in Latin America. Despite its sanitary importance, there are currently only two drugs available for its treatment: benznidazole and nifurtimox, both exhibiting serious adverse effects on patients. In order to complete its life cycle, *T. cruzi* faces extreme environmental conditions –i.e. oxidative stress– as it propagates from an insect vector to a mammalian host, driving the transition from non-infective epimastigote to the infective form trypomastigote. It is known that the antioxidant defense system in the trypanosomatids is different from that in mammalian cells since the parasites have exclusive molecules and reducing enzymes. Because of this, the parasite redox machinery is an attractive target for antiparasitic therapies. The sesquiterpene lactone dehydroleucodine (DhL), is a trypanocidal molecule – containing an alpha-methylene group that could react with sulfhydryl groups of key redox enzymes. This study was focused on elucidating the DhL mechanism of action and extended to ten DhL derivatives (DC-X1 to DC-X10) obtained by chemical substitutions on the methylene group. We firstly confirmed an antiproliferative effect of DhL and its chemical derivatives, being DC-X6 one of the most active. The effect of DhL and DC-X6 was blocked by reduced glutathione, suggesting that compounds are reactive to sulfhydryl groups of certain molecules. Moreover, parasites overexpressing reducing enzymes, such as Tc-CPX, showed a protective effect against these STLs. Consistent with these results, both STLs increased ROS concentration in the wild type parasites. These results indicate that STLs induce oxidative stress on the parasites, possibly by affecting some crucial enzymes of the redox system.

A24

A SEMI-SYNTHETIC MOLECULE DERIVED FROM DEHYDROLEUCODINE AFFECTS THE *TRYPANOSOMA CRUZI* CELL CYCLE

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Trypanosoma cruzi is a parasite causing Chagas disease, which is endemic in Latin America, but in the last 20 years, it has expanded worldwide. The current treatment is restricted to Nifurtimox and Benznidazole, but both are relatively toxic and have limited efficacy on the patients. The development of new effective therapeutic agents is urgently needed. The sesquiterpene lactones (STLs) are natural compounds purified from native plants of Argentina with multiple pharmacological applications. The STL dehydroleucodine (DhL), has an alpha-methylene group that could react with multiple sulfhydryl group-containing proteins, affecting cellular functions such as proliferation, the activity mitochondrial, leading to the cell death/apoptosis. This study is focused on elucidating the action mechanisms of DhL and its derivative DC-X11, obtained by chemical substitution, on *T. cruzi* epimastigotes (strain Dm28c). We observed that DhL and DC-X11 have antiproliferative and cytostatic effects on the parasites. By morphological and ultrastructural studies, we observed an increase of parasites with multiple cell nuclei, kinetoplasts, or flagella after the treatment with DC-X11, suggesting an effect on late steps of the cell cycle (i.e., cellular division). These results were confirmed with parasites synchronized with hydroxyurea (HU 20 mM) for 24 h, and then they were treated with the compound. We concluded that the derivative DC-X11 inhibits *T. cruzi* proliferation by delaying the progression of the cell division. Further studies are necessary to identify the molecular targets affected by DC-X11.

A25

THE NEUROTOXIN OF AN AUTOCHTHONOUS *CLOSTRIDIUM BOTULINUM* AFFECTS THE ACTIN CYTOSKELETON IN BREAST CANCER CELLS

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Botulism is a neuroparalytic disease caused by botulinum neurotoxins (NTBo, serotypes A-G) produced by *Clostridium botulinum*, whose main reservoir is the soil (Su). Infant botulism is a toxi-infection, caused by the ingestion of spores, subsequent colonization, and the production of toxins *in situ*. The autochthonous NTBo would correspond to subtype A2 and have higher toxicity than A1

(Botox®), so in the future, they could be used as a therapeutic agent. The NTBos mechanism of action on certain pathologies is still to be clarified. Previous results from our laboratory showed that autochthonous NTBo 1935 from Su, degrades actin of rat brain homogenates, suggesting this protein could be an active target of NTBos. In this work, the action of this NTBo on the actin cytoskeleton in mammary tumor cells was evaluated. The NTBos of Su from strain 1935 and strain A Hall (both serotype A) were purified by saline precipitation. MCF7 cells (breast cancer cells) were cultured in Petri dishes or coverslips with 250 LD₅₀ of the NTBos for 25, 45, or 90 min. After incubations, cells were processed for Western blot or immunofluorescence in order to evaluate the distribution and expression of actin. NTBo 1935 produced higher actin degradation and an increased location of this protein at the plasma membrane in comparison with A Hall in a time-dependent manner. However, at 90 min of treatment, we observed 90% of cytotoxicity, and further studies at this time were not evaluated. These results provide new insights about the NTBo mechanism of action and its possible use in the fight against breast cancer.

A26

A MELANOMA CELL LINE EXPOSED TO EXTREMELY LOW FREQUENCY MAGNETIC FIELDS: ASSESSMENT OF PROLIFERATION

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Extremely low-frequency magnetic fields (ELF-MFs) have been the axis of heated discussions for decades for their possible causal link to childhood leukemia. However, the ELF-MFs are interesting for the opposite reason: a possible therapeutic use. Indeed, there are several *in vitro* experiments reporting inhibition of cancer cell proliferation, and some *in vivo* studies also point in the same direction: significant reduction of tumor growth has been reported in mice with induced breast cancer tumors, sarcoma, and melanoma. In order to elucidate the effect of magnetic fields on the B-16 cell line (murine melanoma), we built two identical systems of coils of cubic geometry. Each one consisted of a triaxial system of 3 pairs of coils in an orthogonal arrangement. Then we proceeded to perform three experiments (each repeated three times). In all of them, cells were seeded in 96-well microplates (one “control” and one “exposed”), and cell viability was measured by the MTT assay at t = 72 h (beginning of exposure was considered time zero, t = 0 h). A negative control (or sham-exposure) was first conducted where both plates were subjected to the same field (static, vertical 50 microTeslas ‘ μ T’, “MF_{ref}”). In a second experiment, one of the plates was exposed to a 50 Hz 100 μ T_{peak} alternating current (AC) field plus MF_{ref} while the other one was kept at MF_{ref}. In the third experiment, a gradient of the direct current (DC) field was evaluated. No significant differences were found between both plates in any of the three experiments. In summary, the combinations of AC/DC magnetic fields that we tested, for an exposure time of 1h did not affect the viability in the B-16 cell line. Probably, different field parameters, exposure durations and intermittence, as well as cyclic exposure patterns are necessary to obtain results of biological relevance and a possible therapeutic effect.

A27

EFFECTS OF PIOGLITAZONE-RETINOIC ACID ON DAILY RHYTHMS OF APO E IN AN EXPERIMENTAL MODEL OF ALZHEIMER'S DISEASE

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Alzheimer's disease (AD) late onset, which constitutes 90% of cases, could be mainly attributable to deficiencies in the clearance of the A β Apolipoprotein E (Apo E) is associated with age-related risk for Alzheimer's disease and plays a key role in facilitating the proteolytic clearance of A β from the brain. ApoE expression is transcriptionally induced by PPAR γ in coordination with RXRs. Taking into account those observations, the objectives of this study were: first, to analyze the effect of an i.c.v. injection of A β (1-42) on the 24-h rhythms of A β , BMAL1, ROR α , and ApoE protein levels in the rat prefrontal cortex (PC); second, to evaluate the effect of pioglitazone-retinoic acid (Pio-RA) on those temporal patterns. Four-month-old male Holtzman rats were divided into three groups defined as: control, A β -injected (A β) and A β -injected treated with Pio-RA. Rats were maintained under 12 h-light:12 h-dark conditions before the sacrifice. A β , BMAL1, ROR α , and ApoE proteins levels were analyzed by immunoblotting in PC samples isolated every 6 h throughout a 24-h period. The regulatory region of Apo E was scanned for E-box, RORE, RXRE, and PPRE sites. We found that an i.c.v. injection of A β (1-42) modified the daily variation of ApoE, BMAL1, ROR α , and A β protein in the rat prefrontal cortex. Also, we found E-box, RXRE, and PPRE sites on the regulatory region of the Apo E gene. The treatment of Pio-RA reestablished the rhythmicity of those temporal patterns. These findings might constitute, at least in part, the molecular basis of the restoration of daily rhythmicity of Apo E by the administration of Pio-AR in AD.

A28

THE IN VIVO EFFECT OF NATURAL COMPOUNDS ON LEISHMANIA (L.) AMAZONENSIS

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The leishmaniasis is a spectrum of diseases caused by infection with protozoan pathogens of the *Leishmania* genus, with an estimated 2 million new cases per annum. *Leishmania* parasites are transmitted to a mammalian host via the bite of an infected sand fly. The clinical forms of the disease (cutaneous, mucocutaneous, and visceral leishmaniasis) depend on the species of *Leishmania* involved. In Argentina, affects the northern region of the country with an incidence that has increased over the last two decades. Current treatments for leishmaniasis are unsatisfactory due to high associated toxicity, cost, complex administration, and the emergence of resistant strains. Efforts have greatly increased over the last decade to identify novel compounds with anti-leishmanial properties. Thus, one strategy in the search for new compounds is the screening of molecules purified from plant sources. Terpenes appear as good candidates because they are abundant in the plant kingdom, and some of them have shown significant activity against trypanosomatids. The terpenes 5-epi-icetexone (ICTX) and Abietane (HABTO), isolated from *Salvia gilliessi*, are effective against *T. cruzi* and *Leishmania spp. in vitro*. We evaluated the effect of ICTX and HABTO in an *in vivo* model of cutaneous leishmaniasis. Male BALB/c mice were infected in the right footpad with 1×10^5 promastigotes of *L. (L.) amazonensis* and locally treated, once a week for 4 weeks, with 1 mg/animal/day of ICTX or HABTO. We observed that the treatment with the compounds decreases footpad swelling compared to the controls. This is related to the significant decrease in parasite load, splenic index, and IgG levels observed with every treatment. Although many more analyses should be made, these natural compounds could be effective to treat cutaneous leishmaniasis.

A29

EFFECT OF A PPAR γ SYNTHETIC AGONIST ASSOCIATED WITH RETINOIC ACID ON DAILY RHYTHMS OF ROR α AND REV-ERB β IN AN EXPERIMENTAL MODEL OF ALZHEIMER'S DISEASE

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Alzheimer's disease (AD) is the main cause of dementia in the aging population. It is characterized by a progressive cognitive decline and circadian rhythms alterations. At the molecular level, cellular oscillators consist of a network of interlocking transcriptional-translational feedback loops, a positive and a negative one. The positive loop is constituted by the heterodimer BMAL1: CLOCK and the negative loop by phosphorylated PER-CRY complexes. RevErba and ROR α transcription factors, members of the retinoic acid-related orphan receptor (ROR) family, complete the molecular clock machinery. Previously, we found that an i.c.v. injection of A β (1-42) modified the daily rhythms of ROR α and REV-ERB β expression in the rat hippocampus. Taking into account those observations, the objective of this work was to investigate the effects of pioglitazone-retinoic acid (Pio-RA) on the rhythms of ROR α and REV-ERB β expression, as well as BMAL1 and A β protein levels, throughout a 24-h period, in the rat hippocampus. Four-month-old male Holtzman rats were divided into three groups defined as (1) control, (2) A β -injected, (3) A β -injected treated with Pio-RA. Rats were maintained under 12 h-light:12 h-dark conditions and received water and food *ad libitum*. Tissues samples were isolated every 6 h for a 24-h period. ROR α and REV-ERB β mRNA levels were determined by RT-PCR and A β and BMAL1 protein levels were analyzed by immunoblotting. We found that Pio-RA reestablished rhythmicity of those temporal patterns indicating PPAR γ -RXR heterodimer might be a transcription factor involved in circadian regulation and a potential target for the restoration of temporal patterns of clock genes in AD.

A30

HSP27 AND BETA CATENIN ARE IMPORTANT KEY REGULATORS IN THE RESPONSE TO CHEMOTHERAPY

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Cellular senescence is a feature of cancer that can be induced by multiple mechanisms in tumors and can exert both beneficial and detrimental effects on tumor initiation, growth, therapeutic efficacy, and tumor recurrence. The treatment of triple-negative breast cancers (TNBC) involves the administration of the conventional chemotherapeutic drug doxorubicin, given the lack of specific targeted agents. Furthermore, doxorubicin enhances cellular senescence in other tumor cell lines which is considered as a tumor-suppressive mechanism. Recent evidence indicates however that senescent cells secrete various growth factors and cytokines, some of which may paradoxically promote cancer progression such as TGF β , EGF, Wnt ligands, IL8, and IL6. They are known for their ability to promote tumor progression through the inhibition of apoptosis, induction of epithelial-mesenchymal transition (EMT), and/or resistance to therapy. The present study sought to examine the role of the Wnt/ β -catenin and Hsp27 signaling pathway in response to cisplatin (CisPt)/doxorubicin (Doxo) treatment in human triple-negative (TN) breast cancer cell lines and their participation in chemoresistance. MDA MB231 (TN) and MCF10A cell lines were used. Cellular senescence was assayed by measuring SA- β -galactosidase (SA- β -Gal) activity. Apoptosis was evaluated by TUNEL assay and Annexin V-FITC Apoptosis Detection Kit. β -catenin and active β -catenin, Hsp27, phospho Hsp27, survivin, N-cadherin, vimentin, caspase 8, and cleaved PARP

expressions were measured by western blot. Increased numbers of senescent cells (larger and flatter) were observed in both MDA-MB231 and MCF10A cells exposed to the IC50 dose of doxo while CisPt treatment induced apoptosis in MDA-MB231 cells. After 48 h exposure to doxo and recovery for another 48 h, elicited high expression levels of beta-catenin, Hsp27, N-cadherin, vimentin, and survivin, with respect to control. With CisPt treatment an important decreased expression of these proteins was observed. On the other hand, HSP27 downregulation produced significantly decreased expression of HSP27 and beta-catenin. The simultaneous treatment with siRNA and doxorubicin for 48 h kept low expression levels of both proteins. Under these conditions, we observed fewer senescent cells and an increased percentage in apoptosis, measured through the expression of the cleaved PARP and caspase 8 by western blot. CisPt induced apoptosis and Doxo elicited senescence. Hsp27 and beta-catenin may be involved in the cell decision (apoptosis/senescence) after chemotherapy. Doxo treatment promoted high expression levels of EMT related proteins. The downregulation of HSP27 and β -catenin sensitizes the cells to Doxo treatment by decreasing senescence and leading to apoptosis.

A31

CHLOROQUINE ALTERS EXPRESSION AND DISTRIBUTION OF CATION-DEPENDENT MANNOSE 6-PHOSPHATE RECEPTOR OF BREAST CANCER CELLS

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Breast cancer is one of the most important causes of morbidity and mortality worldwide. Several tumoral cells have increased their lysosomal biogenesis in response to metabolic alterations, which also has an impact on the integrity and/or lysosomal functionality, showing increased levels of lysosomal proteases, such as cathepsin D (CatD). CatD was reported to induce apoptosis when it is released into the cytoplasm. Since the lysosomes could play a role either as initiators or executors of apoptotic processes when the membrane integrity is altered, this organelle could be taken as a potential therapeutic target against tumors. Lysosomal proteases are delivered by mannose-6-phosphate receptors (cation dependent, CD-MPR, and independent-, CI-MPR) from the trans-Golgi to late endosomes, where enzyme-receptor complexes are dissociated, and the receptor recycled. Chloroquine (CQ) is a lysosomotropic agent that prevents endosomal acidification. It accumulates inside the acidic compartments and it is used as a potent adjuvant when combined with antitumoral drugs. The aim of this study was to evaluate the effect of CQ on the lysosomal protease endocytic pathway through the CD-MPR behavior in breast cancer cells. Tumorigenic mammary cell line MCF-7 was incubated with CQ for 12 h. Cultures were subjected to immunoblot analysis and IFI. We observed an increased expression of CD-MPR by the treatment. Moreover, the receptor is redistributed from a perinuclear region to a punctuated cytoplasmic detection. In addition, the mature CatD form was decreased by the treatment. Our results suggest that CD-MPR-containing compartments are affected by CQ, possibly leading to impeded CatD trafficking and processing.

A32

NOREPINEPHRINE MODULATES DAILY RHYTHMS IN EX VIVO SPLENIC MACROPHAGES

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The splenic macrophages (M Φ) phagocytes and eliminates circulating pathogens and orchestrate the development of the specific acquired immune response. Even though circadian effects on the immune system have been documented, the circadian regulation in the spleen has not been completely elucidated yet. In mammals, the central clock in the suprachiasmatic nucleus (SCN) of the anterior hypothalamus synchronizes cell-autonomous clocks. Communication between SCN and spleen occurs by the sympathetic nervous system (SNS), through nerves that release norepinephrine (NE) in areas of T and M Φ cells. Previously, other authors reported the daily oscillation of NE in the spleen. Our focus was to determine whether rhythmic expression of the molecular clock in the splenic M Φ is regulated by SNS innervation. For this purpose, we disrupted the autonomic innervation to the spleen of 4-month old male Holtzman rats, by splenic injections of guanethidine. Guanethidine has several effects on peripheral sympathetic neurons including the blockade of neural transmission, depletion of neuronal NE stores, and blockade of the reuptake of NE into the neurons. Animals were maintained under 12 h-light: 12 h-dark conditions and *ad libitum* food/water intake until the experiment. First, we evaluated the effects of local guanethidine injection on spleen macroscopic appearance, size, weight, and lymphocyte numbers. Control animals received intrasplenic saline solution injection. The Student's *t*-test was used for comparison of data between both groups. Second, to study the NE temporal impact on the molecular clock of splenic M Φ , ten days after injection of saline solution or guanethidine, control (N = 4/ZT) and sympathectomized rats (N = 3/ZT) were euthanized at different times during a 24 h period (ZT2, ZT6, ZT10, ZT14, ZT18, and ZT22) and spleen was aseptically removed for ex vivo cultures. The clock transcription factor, BMAL1, and ACTIN were analyzed by Western blot from splenic adherent cells. Time-point data were computed by one-way analysis of variance (ANOVA) and followed by Tukey's post hoc test. Further, chronobiologic statistics were used for validating temporal changes as rhythms. Thus, each series of data were analyzed by the Cosinor method. We found no significant differences in the spleen macroscopic appearance, size, weight, and lymphocyte number between both treated groups, suggesting local guanethidine injections did not have any toxicity on spleen cells. The splenic M Φ from control rats showed a diurnal oscillation of BMAL1 (% rhythm: 71.8), with its acrophase occurring in the middle of the light period. In contrast, the ex vivo splenic M Φ from

guanethidine-treated animals lost the diurnal oscillation of BMAL1. Our results would indicate regulation of the molecular clock in splenic adherent cells by the SCN, through the NE sympathetic pathway.

A33

BYSTANDER EFFECT OF SENESCENT TUMOR CELLS INDUCED BY GAMMA RADIATION

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Premature senescence is a cell-autonomous tumor suppression mechanism actioned by the stimulus as ionizing radiation that affects cell genome integrity. However, senescence cells (SCs) secrete different factors known as senescence-associated secretory phenotype (SASP) able to promote or suppress tumor growth in a non-cell-autonomous manner. In this way, SASP may induce a kind of senescence in their surrounding environment cells via paracrine effects, known as bystander senescence. The aim of this study was to evaluate whether SASP from radiation-induced senescent cells (RIS) induced bystander senescence in proliferating tumor cells and consequently inhibits cell proliferation. Cell cultures of murine melanoma cell line B16F0 were seeded and 24 h later exposed or not to 10 Gy gamma radiation (iB16F0 and B16F0 cells respectively). Three days later senescence was evaluated by senescence-associated β -galactosidase activity (SA- β -gal). Conditioned media (CM) from B16F0 (control CM) and iB16F0 (iCM) were collected and used to evaluate the bystander effect of SASP in proliferating B16F0 cells. B16F0 cells were incubated with CMs and cell proliferation and percentage of senescent cells were measured. At three days after irradiation, a higher percentage of iB16F0 cells were found in senescence (SCs) (3 ± 2 % of B16F0 cells vs 46.9 ± 2 % of iB16F0 cells; $**P < 0.01$). When iCM was incubated with B16F0 cells, it suppressed cell growth. After three days incubation, were found $598.5 \pm 7.5 \times 10^3$ cells treated with iCM vs $764 \pm 5 \times 10^3$ cells treated with CM ($**P < 0.01$), and an increased SCs percentage in iCM treated cells was observed (6.3 ± 3 % CM vs 27.5 ± 5 % iCM; $**P < 0.01$). We conclude that SASP from iB16F0 cells would suppress tumor cell growth by inducing bystander senescence. Further studies should be done to understand the mechanisms involved and exploit its therapeutic consequences.

REPRODUCTIVE AND DEVELOPMENTAL BIOLOGY

A34

N-ACETYLGLUCOSAMINE CONTENT CHANGES IN BULL SPERM DURING EPIDIDYMAL MATURATION

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After leaving the testis, spermatozoa undergo several molecular changes up to reach the female genital tract. Most of these modifications occur during epididymal transit as a direct result of exposure to, and interaction with, epididymal luminal environment. Among these modifications, addition, removal, and/or modification of external sugars on glycolipids or glycoproteins have been described in different species. The aim of this study was to evaluate changes in N-acetyl-glucosamine (NAG) content of the plasma membrane glycoproteins in bull spermatozoa during their epididymal maturation. For this purpose, fresh epididymides from bulls (Aberdeen Angus) were used. Organs were dissected into caput, corpus, and cauda, and the sperm were obtained by slicing of the tissue and centrifugation. Ejaculated spermatozoa were obtained from cryopreserved samples. NAG and galactose (GAL) were evaluated in sperm glycoproteins by using respectively WGA and BS-I lectins conjugated with either FITC for flow cytometry or with biotin for blotting assay. By flow cytometry, we observed that NAG detection increased progressively from caput to ejaculated sperm. By blotting sperm glycoproteins onto nitrocellulose membranes, and subsequent detection with biotinylated lectins, we observed a decrease in the NAG content of some glycoproteins, while the signal increased in a ~60 kDa protein of cauda spermatozoa. No major changes were observed in the GAL content between sperm from caput to cauda. We also observed that β -N-acetyl-glucosaminidase activity increased from caput to cauda and the enzyme is redistributed to the epididymal fluid. These changes could provide new insights about carbohydrate rearrangement that can be used as parameters for sperm maturation.

A35

STEROIDOGENIC ABILITY OF MACROPHAGE SECRETIONS IS MODIFIED BY DEXAMETHASONE IN RATS WITH POLYCYSTIC OVARY

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Polycystic ovary syndrome (PCOS) is a chronic disease affecting reproductive function and whole-body metabolism. The syndrome is characterized by hyperandrogenism, ovarian dysfunction, obesity, and insulin resistance. Moreover, a higher exhibition of proinflammatory agents, such as nitric oxide (NO) and certain androgen-induced ovarian monocyte-derived cytokines participate in endocrinal and pathophysiological events of PCOS. Successful use of corticosteroids in the treatment of anovulatory infertility has been reported, although the theoretical basis for this application has not been fully elucidated. We have previously informed that secretion from spleen macrophages (MΦ) can induce hormone release from the ovary. Based on this evidence, now we investigated the possibility that dexamethasone (DEX) could affect the steroidogenic ability of MΦ secretions and consequently modifies the ovarian androstenedione (A2) release from rats with polycystic ovary (PCO). PCO condition was induced by injecting 60-day old rats with a single 2 mg estradiol valerate (Sigma, USA) dose dissolved in 0.1 mL of corn oil. After 2 months, the rats were sacrificed (N=6 per group in two different experiments). MΦ (1×10^6 cells) from PCO and no-PCO (control) rats were cultured for 24 h in RPMI medium in the absence or presence of 10^{-6} M DEX. Afterward, the medium was removed, the MΦ were washed twice, and finally, the cells were cultured for an additional 24 h period. The respective culture media were collected and used to stimulate ovaries from PCO and control rats for 3 h in metabolic bath. The ovarian A2 release was measured by electrochemiluminescence (Cobas e411), while NO (as nitrites) were quantified by Griess reaction. The mRNA expression of interleukin (IL)-1β and IL-6 (pro-inflammatory cytokines) were assessed in ovary by RT-PCR. PCO ovaries released less A2 and NO after stimulation with secretions of PCO MΦ+DEX, compared with PCO MΦ secretions ($P < 0.05$). The mRNA expression of IL-1 and -6 in PCO ovaries incubated with secretions from PCO MΦ+DEX was lower than that obtained with PCO MΦ secretions ($P < 0.05$). In PCO MΦ, the NO release was higher than control and PCO MΦ+DEX ($P < 0.01$ and 0.05 , respectively). The observed modulation of A2 release may suggest a link between excess androgen, chronic inflammation, and immune cells in PCO rats. Modification of steroidogenic ability of MΦ secretions by DEX might contribute to the beneficial effect on reproductive function in polycystic ovary syndrome patients.

A36

CROSSTALK AMONG SPLEEN MACROPHAGES AND ANTERIOR PITUITARY IN A POLYCYSTIC OVARY MODEL IN RAT

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Interactions between the neuroendocrine and immune systems are essential for the maintenance of homeostasis and overall health. The hypothalamic gonadotropin-releasing hormone (GnRH) plays a pivotal role in the neurohormonal control of reproduction. At the anterior pituitary level (AP), GnRH interacts with specific receptors located at the surface of gonadotrophs (GnRH-R) to regulate in an integrated manner the synthesis and release of gonadotropins (luteinizing hormone: LH and follicle-stimulating hormone: FSH), which, in turn, promote the development of gonadal functions. In addition, the immune cells express GnRH and GnRH-R, suggesting an autocrine or paracrine role for GnRH within the immune system. It has been shown that nitric oxide (NO) may act in both the AP and immune system as a neurotransmitter and, also, it is associated with reproductive pathogenesis. Polycystic ovary syndrome (PCOS) is a clinical disorder characterized by hyperandrogenism and chronic anovulation in women. A neuroendocrine hallmark of PCOS is persistently rapid LH and GnRH pulsatility, which favors the synthesis of LH over that of FSH. Previously, we showed in rats with polycystic ovary, that spleen macrophage secretions modified the LH release from AP. Now, the focus of this study was to determine in MΦ in culture whether GnRH as well as the secretions of AP influences the ON secretion. PCO condition was induced in adult rats with a single 2 mg estradiol valerate dissolved in 0.1 mL of corn oil. MΦ from PCO and Control rats (two rats/group in two independent experiments) were cultured (1×10^6 cells) for 24 h in RPMI medium (basal condition). Then, MΦ were treated with or within of GnRH 10^{-8} M, Cetrorelix 10^{-6} M (GnRH antagonist) for 3 h. The medium was removed, the MΦ were washed and the cells were cultured for 24 h more. Also, MΦ was incubated with PCO-AP secretions. In all incubation liquid, it was measured the NO release by Griess reaction. The mRNA levels of GnRH-R, in MΦ and AP, were assayed by RT-PCR. In basal conditions, PCO-MΦ released more NO than C-MΦ ($P < 0.05$). GnRH and PCO-AP secretions increased NO levels in relation to

PCO basal value ($P<0.01$). Cetrorelix significantly decreased NO response in AP and MΦ from C and PCO rats. In both, MΦ and PCO-AP, GnRH-R mRNA increased compared to C ($P<0.01$ and $P<0.05$, respectively). Our results indicate that GnRH and its receptor are a link between the two systems. Exogenous GnRH modulates NO release from PCO-MΦ. Likewise, PCO-AP secretions, which contain endogenous GnRH, affect in PCO-MΦ the NO response. This bidirectional communication could be contributed to abnormal gonadotrophins secretion in polycystic ovary syndrome.

A37

INFLUENCES OF GONADOTROPIN-RELEASING HORMONE AND NITRIC OXIDE ON LUTEAL REGRESSION THROUGH THE PERIPHERAL NEURAL PATHWAY

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Using the Coeliac Ganglion-Superior Ovarian Nerve-Ovary system (CG-SON-Ov) of late pregnant rats, we have physiologically demonstrated the presence of GnRH/GnRH-receptor system in CG and the influence of NO on the ganglionic GnRH favoring its release. In addition, taking into account that the peripheral neural system can act as a modulator of corpus luteum of pregnancy (CL), the objective of this work was to investigate the effect of cetrorelix (CTX) and L-NAME from CG, through the SON, on functional and structural luteal regression markers. The CG-SON-Ov system, extracted from rats with 21 days of pregnancy, was incubated in Krebs Ringer at 37 °C, keeping CG and Ov connected by the SON, in separate compartments, for 180 min. The experimental groups (N=6 in each experimental group) consisted in the addition in the ganglionic compartment of (a) CTX, GnRH receptor antagonist (10^{-6} M); (b) L-NAME (N^G-nitro-L-arginine methyl ester), an inhibitor of nitric oxide synthase (100 μM); and (c) L-NAME (100 μM) + CTX (10^{-6} M). The control group consisted of CG-SON-Ov systems that were untreated. Progesterone (P), the main steroid secreted by the CL, was determined by RIA in the ovarian compartment and the luteal mRNA expression of P synthesis and degradation enzymes, 3 beta-hydroxysteroid dehydrogenase (3β-HSD) and 20-alfa-hydroxysteroid dehydrogenase (20α-HSD) respectively, were assessed by RT-PCR. Apoptotic cells were detected by TUNEL in ovarian tissue sections. The data were statistically analyzed using one-way ANOVA and post hoc Tukey's test ($P<0.05$). CTX, L-NAME, and L-NAME + CTX added in CG increased the ovarian P release, significantly ($P<0.01$, $P<0.05$, and $P<0.001$, respectively). The comparative statistical analysis between the groups L-NAME and L-NAME + CTX indicates a prevalence of the effect of CTX ($P<0.05$). The expression of its synthesis enzyme, 3β-HSD in CL showed a tendency to increase in the three experimental groups, this increase was significant when L-NAME+CTX was added in CG ($P<0.05$). While the expression of P degradation enzyme, 20α-HSD, decreased significantly when CTX was added in CG ($P<0.05$). The morphometric study of luteal cells confirmed that the addition of CTX in CG significantly decreased ($P<0.001$) levels of apoptosis in CL with respect to the control group. In conclusion, GnRH may contribute as a regulator factor of the functional and structural luteal regression, through the peripheral neural pathway, necessary for delivery to occur.

BIOCHEMISTRY, PHYSIOLOGY AND NEUROCHEMISTRY

A38

MEMORY AND LEARNING DECLINE ARE RELATED WITH TEMPORAL CHANGES OF COGNITIVE FACTORS IN AGED RATS

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Aging brain undergoes several changes leading to a decline in cognitive functions. *Bdnf* and its receptor *TrkB*, are expressed in different brain regions, including the hippocampus, and regulate a wide range of functions such as synaptic plasticity, long-term potentiation, and neurogenesis, which are fundamental for memory and learning. Interestingly, previous results from our laboratory showed a loss of the circadian variation in the expression of *Bdnf* and *TrkB* mRNA in the hippocampus of older rats. The objective of this work was to evaluate the effect of aging on cognitive processes and compare it with the temporal expression of *Bdnf* and *TrkB* in the rat hippocampus. Holtzman male rats were separated into two experimental groups: young adult (3-months old) and old (22-months old) rats. Cognitive performance was assessed by the Barnes maze (BM) test for spatial learning and memory and by the Novel Object Recognition (NOR) test for contextual learning. In the BM test, older rats showed a significant lower exploratory frequency of the target region ($P<0.05$), longer total exploratory activity ($P<0.01$), greater numbers of errors in reaching around the

target hole ($P<0.05$), longer escape box latencies ($P<0.05$), higher distance traveled on the platform ($P<0.05$) and lower percentage (%) of exploration of the meta holes ($P<0.05$), compared to young adult rats. In the case of the NOR test, older rats showed a significantly shorter time for novel objects exploration ($P<0.01$), compared with the young adult rats. Taken together, the exposed evidence suggests that the loss of Bdnf and TrkB rhythmicity in the hippocampus could play a central role in the deterioration of memory and learning in aging. Thus, the alteration in the cognitive factors would be reflected in the low performance of the aged animals in both behavioral tests. Understanding the basis of cognitive impairment during normal aging is essential to develop strategies to prevent aging-related diseases and improve the quality of life of our older adults.

A39

FORCED SWIMMING REVERT THE METABOLIC ALTERATION PRODUCED BY CHRONIC PRENATAL STRESS

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Numerous evidences from animal and human studies suggest that prenatal stress is related to the fetus developing to produce metabolic syndrome in adult life. Metabolic syndrome is characterized by the following medical conditions: dyslipidemia, insulin resistance, glucose intolerance, hyperglycemia, and increased cellular oxidative stress. Forced swimming (FS) is a type of physical exercise, a stressor that its effects depend on stimuli intensity and duration. The aim of this work was to determine the effect of prenatal immobilization (IMO) stress and postnatal FS stress on blood glucose, parameters of lipid metabolism, and liver oxidative stress. For the study, we used three-months-old male offsprings from IMO stressed mothers (EP) and offsprings from no stressed mothers (CP). Half of the CP and EP animals were submitted to a forced swimming session for 30 min. Before blood extraction, corticosterone (COR), glycemia, total cholesterol, high-density lipoprotein (HDL), low-density lipoprotein (LDL), triacylglycerides (TAG), and insulin levels were measured. The atherogenic index was analyzed. Thus, the livers of all animals were removed for oxidative stress (MDA levels) analysis. The animals were maintained according to the Guide for Care and Use of Laboratory Animals and the experiments were approved by the local Institutional Animal Care Committee. Results showed that FS postnatal stress increased COR levels in prenatal stress animals compared to control. IMO prenatal stress increased glucose, insulin, HDL-cholesterol, LDL-cholesterol, TAG levels, and atherogenic index. Additionally, FS attenuates the alterations produced by prenatal IMO stress in all metabolic parameters evaluated while MDA concentration was higher in all stressed group compared to controls animals. In conclusion, postnatal FS reverts the effects of prenatal stress in some metabolic syndrome factors, but oxidative stress could not be normalized by the FS session.

A40

HAVE THE STRESS HORMONES AN OXIDATIVE EFFECT ON THE ERYTHROCYTE MEMBRANES?

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Stress may induce both beneficial and harmful effects. The beneficial effects involve preserving homeostasis of cells/species. However, the harmful effects may receive more attention due to their role in various pathological conditions and diseases. Stress hormones, cortisol (CORT) and epinephrine (EPI) can interact with different cells, erythrocytes between them, through their bound specific or nonspecific to the cell membranes, leading to profound structural changes. In this work both in vivo and in vitro oxidative effects of stress hormones on erythrocyte membranes and their effect in red blood cell (RBC) aggregation and elasticity were studied. Control and stressed (movement restriction, 1 h/day, 3 times/week /45 days) adult Wistar male rats were used. After the last stress, blood samples were obtained and the globular sedimentation rate (GSR) was measured. After blood centrifugation, plasmatic corticosterone and bilirubin, osmotic erythrocyte resistance (OER), and erythrocyte aggregation (EA) were determined. Erythrocyte ghosts and cytosol were obtained after their hemolysis and sequential centrifugation. Aliquots of erythrocyte ghosts of control rats were incubated one hour at 37 °C (control) or with the addition of dexamethasone (DEX, 10⁻⁶ M), epinephrine (EPI, 10⁻⁶ M), or DEX plus EPI. Carbonyls and MDA in ghosts and SOD and CAT in cytosolic fraction were measured. Stressed rats decreased OER ($P\leq 0.006$) and increased bilirubin ($P\leq 0.0007$), corticosterone ($P\leq 0.01$), EA ($P\leq 0.05$), GSR ($P\leq 0.05$), SOD ($P\leq 0.03$) and CAT activity ($P\leq 0.01$), MDA ($P\leq 0.04$) and carbonyls ($P\leq 0.000001$). Both DEX ($P\leq 0.0001$) and EPI ($P\leq 0.01$) produced lipid oxidation in ghosts, but the effect was greater with DEX ($P\leq 0.004$) than EPI. No significant effects on both hormones together were observed. Both hormones (DEX, $P\leq 0.01$ and EPI, $P\leq 0.03$) increased carbonyl levels in the same magnitude, and the addition of the two hormones together did not have a summative effect. MDA (200%) and carbonyls (3000%) increase in vitro were of the same magnitude of those observed in chronically stressed animals, being the oxidation process significantly higher in proteins than in lipids. SOD and CAT increases showed that RBCs use their high-capacity redox system to scavenge intracellular radicals. Oxidative changes induced by the stress hormones would lead to structural alterations in the RBC membrane, increasing RBC aggregation. The great protein oxidation would be responsible for the membrane rigidity increase and the increase in hemolysis rate. Several

investigations show that oxidized membrane lipids could be repaired, depending on the plasma antioxidant capacity and that protein oxidation are irreversible and are generally associated with loss of function; the great stress-induced carbonylation observed in this work may result in the deleterious role in the etiology and/or progression of various human diseases.

A41

CADMIUM EXPOSURE MODIFIES SERUM GLUCOSE AND THE EXPRESSION OF INTESTINAL HORMONE GENES

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Cadmium (Cd) has rapidly gained considerable attention due to its critical role in the development of multiple endocrine-related disorders. From the last few decades, their contamination has been raised dramatically owing to continuous discharge in sewage and untreated industrial effluents. In previous studies, we observed linked cadmium exposure to disturbances in lipid metabolism on the intestine. The purpose of this work is to determine if cadmium exposure affects the regulation exerted by the intestine on glucose metabolism. For this, plasma glucose and intestinal expression of incretins were evaluated, hormones that stimulate insulin secretion and enhance its effect. Adult male Wistar rats, 180 to 200g body weight, were exposed to CdCl₂ (15 ppm) in drinking water, controls drank drinking water. Feeding was ad libitum. After 15 days (15 d), 30 days (30 d), and 45 days (45 d) of treatment the animals were sacrificed and the intestine was extracted for further study and stored at -80 °C. Serum glucose was measured by Wiener kits. The mRNA expressions of GIP (glucose-dependent insulinotropic polypeptide), GLP-1 (glucagon-like peptide-1), and PC 1/3 (prohormone convertase) were determined by RT-PCR, using S28 as an internal control. Cadmium treatment caused a decrease of blood glucose in 30 d and 45 d with respect to control ($P<0.01$) and 15 d groups ($P<0.01$). Expression PC 1/3 increased in 15 d respect to the control ($P<0.05$), and in 30 d and 45 d respect to the control ($P<0.001$) and 15 d ($P<0.01$); GLP and GIP did not show change. In conclusion, treatment with cadmium would be activating GLP (insulinotropic hormone) as it increases PC1/3 levels; this could contribute to the decrease of glucose levels found in the serum. Possibly the changes found could help trigger the metabolic syndrome that is reported in other works.

A42

GLYPHOSATE-RESISTANT TRANSGENIC ROUNDUP READY® SOYBEAN (GTS-40-3-2) DIET EFFECT IN WISTAR RATS

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During the last two decades, the advent of genetically modified organisms (GMOs) has revolutionized agronomic practices. In Argentina, soybean is the main genetically modified (GM) crop being over 90% of the Roundup Ready® (RR) variety, resistant to glyphosate, which is the main herbicide used. Due to the concern about the safety of meals containing GMOs, several studies have been made, although there is no consensus among scientists about GMOs' safety. In this work, Wistar rats were divided into two groups: one was fed with a diet containing 13% RR transgenic soybean (RR group) and the other group with a similar diet containing non-transgenic soybean (NO RR group) for 170 days, in order to evaluate different physiological parameters. Daily body weight gain (DWG) was measured. At the end of the experimental time, the liver was removed, and the blood was collected. After centrifugation, plasma and erythrocytes were separated. Osmotic Erythrocytes resistance was determined and then membranes and cytosol from erythrocytes were separated by centrifugation. Malondialdehyde (MDA) and carbonyls (CAR) were determined in membranes. In plasma, bilirubin (BIL), GOT, GPT, total proteins (TP), and creatinine (CRE) were determined. Hepatic MDA and superoxide dismutase (SOD) were measured. While DWG was higher in NO RR group (1.6 ± 0.05 vs. 1.37 ± 0.05 g/day, $P\leq 0.007$), no changes were observed in TP levels. An increment in hepatic MDA (115 ± 6.45 vs. 79.8 ± 6 nmol/g tissue, $P\leq 0.0017$) and SOD (6.9 ± 1.57 vs. 2.75 ± 0.68 U SOD/mg protein, $P\leq 0.032$) levels was observed in the RR group, which could indicate an oxidative stress process in this tissue, although no difference was observed in GOT and GPT levels. On the other hand, while erythrocytes showed decreased resistance in RR group (21.5 ± 2.64 vs. 14.86 ± 1.73 percent of hemolysis, $P\leq 0.05$), which could be due to a modification in the cell membrane, no difference was observed in plasmatic BIL or MDA, and CAR levels in the erythrocyte membrane. This would indicate that the change observed in the membrane resistance is not caused by oxidation. On the other hand, CRE levels did not show differences between both groups. According to the results, while some changes were observed in physiological parameters, some of them did not show differences between animals fed with RR or NO RR soybean. This indicates that RR soybean chronic intake could

have any harmful effect, some studies should be carried out in order to clarify the effects observed, and long-term studies should be performed.

A43

DETERMINATION OF RESPONSE TIME ON THE LABORATORY OF PROVINCIAL MATERNITY “TERESITA BAIGORRIA” (MPTB)

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The response time (RT) is the time that passes since a sample is received in the lab until the results inform is delivered. It is considered as an indicator of the laboratory's efficiency; its systematic measurement is indispensable to guarantee extra analytic quality. The aim was to evaluate global RT of MPTB's laboratory in chemistry, hematology, serology, and endocrinology areas in a period of time comprehended between 2016 and the first trimester of 2019. The differences in response time were analyzed in each one of the periods whit ANOVA, using software GraphPad Prism 5.0. Determinants made on different sectors were evaluated during 13 trimesters. On hematology between the trimesters 1 and 13, the increase in samples was 3944, and the RT reduced by 8% ($P<0.001$). On clinical chemistry between the trimesters 1 and 13, the increase of samples was 5696, and the RT reduced by 64% ($P<0.02$). On endocrinology between the trimesters 1 and 13, the increase of samples was 1027, and the RT reduced by 96% ($P<0.001$). On serology, there was a reduction of 1160 samples between the 1 and 13 trimesters, and the RT reduced by 84% ($P<0.01$). It was concluded that despite the increase in the number of samples, the TR has decreased significantly improving laboratory performance.

A44

EFFECTS OF MEDICINAL CANNABIS OIL ON THE COGNITIVE PERFORMANCE OF AN ANIMAL MODEL OF AGING

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The endocannabinoid system participates in the regulation of the neuronal synapse in different areas of the brain, including the cognitive areas (cortex and hippocampus), and the system activity can be modified by the consumption of exogenous cannabinoids. Actually in Argentina, cannabis oil has acquired medicinal significance, due to its use in non-conventional medicine for the treatment of different pathologies such as refractory epilepsy, chronic pain, nausea and vomiting, appetite stimulation, among others; however, its effect on learning and memory processes during aging is poorly known. For this, we use wild-type Wistar rats of 12 months old that were treated with 100 µL of medicinal cannabis oil (MCO, 0.75 mg CDB/Kg.day) or corn oil (CO, placebo), orally, up to 18 months of age. The cognitive tests (Novel object recognition) showed a significant increase of recognition memory of the MCO group vs CO group (ANOVA, Tukey's post-test, $P<0.05$). Then, animals were sacrificed according to the protocol approved by the Bioethics Committee of our University, and the brain cortices we dissected analyze the REST levels (a genic regulator involved in neuroprotection and normal cognition during aging) and we observed a significant increase of REST levels in MCO vs CO group (Student's *t*-test, $P<0.05$). Finally, we measure glycemia and lipidic profile in serum to detect any change due to the daily consumption of oils, and although we observed in the MCO group a tendency to lower glycemia, total cholesterol, HDL cholesterol and triglycerides, these changes were not significant. Together, these data suggest that in our model, the daily consumption of low doses of medicinal cannabis oil, can improve cognitive ability and induce neuroprotection during aging, without altering glycemia and lipidic profile.

A45

EFFECT OF SELENIUM AS A PROTECTIVE AGENT DURING ISCHEMIA INJURY INDUCED IN TRANSPLANT ORGANS

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Selenium is an essential element in human and animal metabolism, which is found as a constituent of selenoproteins. Selenium in selenocysteine form is integrated into the catalytic site of glutathione peroxidase (GPx), an antioxidant enzyme that protects the cells from the damage caused by reactive oxygen species (ROS). Oxidative stress is the imbalance between ROS and antioxidant defense systems, generating alterations of DNA, proteins, and lipid peroxidation. The imbalance occurs particularly during ischemia and lack of *postmortem* perfusion. This mechanism is of relevance in organs transplant, it affects the outcome during surgeries. The aim of this research was to evaluate the effect of selenium as a protective agent during ischemia injury induced *postmortem* in transplant

organs. Wistar rats (N=20) were administered via oral 75 µg/kg/day of seleno-methionine (Se-Met) during 7, 14, and 21 days. After sacrificed, the liver, heart, and kidney samples were collected at different *postmortem* intervals PMI (0-1-3-6-12 h). Total selenium concentration in organs was determined by Inductively Coupled Plasma Mass Spectrometry (ICP-MS). The administration of Se-Met produced a significant increase of selenium concentration in the liver (65%, $P<0.001$), heart (40%, $P<0.01$), and kidney (45%, $P<0.05$) respect to control. At 21 days, selenium showed significantly higher levels in kidney than control animals (3.75 ± 0.24 vs. 2.02 ± 0.37 mg/kg, $P<0.01$). Oxidative stress was measured by malondialdehyde (MDA) liquid chromatography. The MDA decreased significantly in the heart (0.21 ± 0.04 vs. 0.12 ± 0.02 mmol/g) and the kidney (0.41 ± 0.02 vs. 0.24 ± 0.03 mmol/g) at one-hour PMI ($P<0.01$). Organs removed at different PMI intervals showed lower production of MDA compared to the control group ($P<0.05$). Se-Met decreased significantly oxidative stress in transplant organs from 1 to 12 h PMI. The results suggest that selenium is a protective agent that improves transplant organs' survival and the outcome of transplant surgeries. Future studies will be focused on the specific activity of GPx and selenium in transplant organs.

A46

LONG-TERM EFFECTS OF NEONATAL HYPOXIA ON ANXIETY-RELATED BEHAVIORS AND HORMONAL RESPONSE TO ACUTE STRESS

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Neonatal lesions in the brain have less severe effects than in adults due to the neuronal plasticity of the developing nervous system, although they can cause cognitive and behavioral sequelae. Previously, we found that neonatal hypoxia (NH) transiently affected the expression of proteins associated with synaptogenesis in certain brain areas. The intermingled neural circuits controlling both stress and anxiety suggest a strong relationship between stress experiences and anxiety in both healthy and pathological conditions. We evaluated the long-term effects of NH on anxiety parameters and in stress-induced hormone release in adult female (estrous day of rat cycle) and male rats. Sprague Dawley rats at 4 Post-Natal Day (PND) were exposed to an atmosphere of low oxygen level (6.5% O₂ and 93.5% N₂) for 70 min. 4PND control pups were exposed to normal oxygen levels (Co) for 70 min. The humidity and temperature conditions were controlled. Pups were then returned with their mother until weaned, and then they were allowed to grow. At 3 months of age, both groups of rats were subjected to two tests, Elevated Plus Maze (EPM) to measure anxiety parameters and a stressful challenge to determine hormone response to acute stress (exposure to ether vapors for 2 min). EPM reflected an unconditional aversion to heights and open spaces, an anxiogenic behavior. The hormonal response to stress included the release of pituitary prolactin (PRL), adrenal progesterone (P₄), and adrenal corticosterone (CORT). Blood samples were collected before and after 5 min of stress exposure for serum hormone determinations by RIA. In the EPM test, both female and male hypoxic rats increased the number of entries to the open arms (OA) and the time spent in the OA compared to Co ($P<0.05$). The results obtained indicated an anxiolytic-related behavior induced by NH, that was higher in female than in male hypoxic rats. Basal levels (unstressed) of PRL and P₄ in NH rats remained similar to Co ones in both sexes. Only in female rats, NH increased the basal levels of CORT compared Co rats ($P<0.05$). In female and male rats, the hormonal release of PRL, P₄, and CORT induced by stress, were differentially affected by NH. Hypoxia attenuated the stress-induced PRL secretion in female rats ($P<0.05$) while this response was blocked in males. The release of CORT by stress was blunted in both sexes by NH. The release of P₄ by stress was inhibited in NH female but it was preserved in male rats. In conclusion, the long-term effects of NH were influenced by sex. NH altered the anxiety levels and the hormonal response to stress in adulthood. The alterations caused by NH at the brain level could be influencing the appropriate response to situations of stress and anxiety in adulthood.

A47

AMPHETAMINE PRENATAL EXPOSURE INFLUENCES SEX-DEPENDENT PITUITARY RECEPTORS EXPRESSION IN RESPONSE TO STRESS IN ADULT RATS

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Sex steroids modulate brain dopaminergic systems and influence neuroadaptive changes induced by prenatal exposure to amphetamine (PEA). Pituitary dopamine D2 receptor (D2R) and prolactin receptor (PRLR) are involved in the modulation of prolactin synthesis and secretion, and the endocrine milieu regulates prolactin in basal and stress conditions. Our objective was to evaluate the sex differences in pituitary D2R and long PRLR isoform expression following stress in adult PEA and control rats. Female Wistar rats were treated daily with amphetamine 2.5 mg/kg i.p./saline during days 15 to 21 of pregnancy. Their offspring were sexed, and the females were OVX at day 60 and 15 days later treated with estrogen/oil (E2; 2 x 5 µg/rat/24 h). At 75 days, male, OVX, and OVX+E2 rats were exposed to immobilization stress for 30 min. Blood and tissue samples were collected for corticosterone by RIA, and the pituitaries were obtained to determine D2R, PRLR (long) expression, and PRL content by real-time PCR and Western blots (WB). The data were analyzed using a two-way ANOVA. Our results showed that, in basal male control rats, corticosterone levels were lower than OVX+E2, and PEA blunted this response ($*P<0.05$). No sex differences were observed in

stressed rats. In basal conditions, OVX and OVX+E2 rats showed higher mRNA levels of pituitary D2R than male rats, and E2 blunted this effect in PEA rats ($*P<0.05$). Since pituitary D2R expression increased in stressed male rats, no effect in female control and PEA rats was observed following stress. Moreover, pituitary D2R expression (protein) was diminished only in PEA OVX+E2 rats ($P<0.05$), and no effects of stress were observed. Basal PRLR expression was higher in female than male rats in control and PEA rats ($^{\#}P<0.05$) but decreased in stressed rats only in the control group. High PRL mRNA levels were observed in OVX+E2 basal and stressed rats ($^{\#\#}P<0.01$), and PEA diminished PRL content in both conditions ($*P<0.05$). No effect was observed in male and OVX rats. In conclusion, sex differences were observed in pituitary D2R and PRLR expression in PEA rats in response to stress that could be affecting prolactin synthesis and secretion.

A48

PROGESTERONE EFFECT ON FACTORS INVOLVED IN THE PROGRESSION OF ENDOMETRIOSIS

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Endometriosis (EDT) is a hormone-dependent inflammatory disease characterized by the implantation and growth of endometrium-like tissue outside the uterine cavity. It is known that estradiol has an unfavorable effect in the development of pathology, while progesterone role is less established. Therefore, the aim of this work was to study the influence of progesterone on factors related to apoptosis, invasion, and inflammation that are involved in the development of endometriotic lesions in a murine model. The EDT was surgically induced in C57BL/6 mice by the autotransplant of three pieces of the right uterine horn to the intestinal mesentery. From day 1 after the operation, the animals of the control group (N=7) received sterile corn oil, while the animals of the experimental group (N=7) received 2.5 mg progesterone/kg bw/day subcutaneously for a month. Then, the mice were sacrificed by cervical dislocation, and the lesions were removed and stored at $-80\text{ }^{\circ}\text{C}$ for subsequent gene expression analysis (RT-PCR) of markers of the intrinsic apoptotic pathway (Bax and Bcl-2), inflammation markers (IL-1 β and TNF- α), and TNF receptors (p55 and p75). Peritoneal fluid was also collected for the determination of the enzymatic activity of metalloproteinase 2 (MMP-2) and its zymogen by zymography. The data were statistically analyzed by the Student's *t*-test ($P<0.05$). The administration of progesterone did not change the expression of apoptosis markers or the activity of the MMP-2 enzyme and its zymogen. In relation to the inflammation factors, the expression of IL-1 β and TNFRp55 did not modify, while the expression of TNF- α and TNFRp75 decreased significantly in endometriotic lesions in comparison with the control group ($P<0.05$). In addition to promoting the inflammatory process, TNF- α cytokine contributes to the maintenance of functional nerves and associated pain in EDT. Therefore, the results obtained would indicate that progesterone, although it seems not to alter apoptosis and invasion, can have a beneficial effect on this pathology by modulating the expression of the TNF- α -TNFRp75 signaling system.

A49

EFFECTS OF PRENATAL STRESS AND POSTNATAL ACUTE FORCED SWIMMING ON SOME BIOCHEMICAL PARAMETERS OF LIVER AND SKELETAL MUSCLE

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The aim of this study was to analyze the physiological response of adult rats to prenatal stress and postnatal acute forced swimming on skeletal muscle and hepatic parameters. Males of three months of age were used, controls individuals (PC), and some prenatal stressed for immobilization (IMO) during pregnancy (PS). Half of the PS and PC animals were submitted to the postnatal forced swimming stress section for 30 min. Corticosterone (COR), glycemia, and malondialdehyde (MDA) levels were studied. Our results showed that in basal conditions COR and glycemia levels are increased in PS animals. Also, after an acute exposition of forced swimming, COR significantly increases in the PS group in the same way as in PC rats but with minor effects. The hepatic MDA levels, in the control condition, showed a rise in PS rats compared to PC animals, being the difference maintained after forced swimming stimuli. In skeletal muscle, MDA values of PS animals were increased in basal conditions, but this effect was observed in all groups after forced swimming treatment. In conclusion, IMO prenatal stress causes a change in the regulation of the HHA axis, reflected in high basal levels of plasmatic COR and hyperglycemia. Moreover, it produces a hypersensitivity to acute stressors during postnatal adult life. On the other hand, PS and forced swimming stress generate high levels of oxidative stress in the liver and skeletal muscle.

BIOTECHNOLOGY AND GENETICS

A50

UPGRADING GRAPE AND OLIVE POMACES BY PECTINOLYTIC ENZYMES PRODUCTION: OPTIMIZATION OF CULTURE CONDITIONS

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In the wine and olive Cuyo region industries, large masses of grape pomace (GP), grape stalks, alperujo and olive pomace (OP) are produced. These wastes have polluting characteristics such as low pH and high content of phytotoxic and antibacterial phenolic substances, which hinder their biological degradation. According to current legislation, they must be treated for safe final disposal. Solid-state fermentation (SSF) is a biotechnological option to revalue them, by the generation of value-added products (for example, enzymes). SSF performance depends on many variables, and then it is important to state the relevance, for each of them, when a certain bioproduct is an objective. In a previous report, for the production of a Pectinolytic Enzyme Complex (PEC) by *Aspergillus niger* SSF, relevant variables were found. The objective of this work was to optimize the value of these relevant variables, for maximum PEC production. The Box Behnken Design (BBD) was used for the variables: glucose (G), micronutrient solution (MS), and initial humidity (H); as responses, enzymatic activities (Exo-polygalacturonase (Exo-PG), Endo-polygalacturonase (Endo-PG) and Exo-polymethylgalacturonase (Exo-PMG) were analyzed. The BBD requires setting three levels: low, medium, and high. The solid substrate was placed in flasks (50% GP and 50% OP) corresponding to a BBD test. Fifteen SSF was carried out for 3.5 days at 27 °C with an inoculum of 1×10^7 spores/g_{dry}. Enzymatic extracts from SSF were prepared in Na-acetate buffer (0.1 M, pH 5). The Exo-PG and Exo-PMG activities were determined at 45 °C by spectrophotometric methods and Endo-PG, by viscosity monitoring via Ostwald viscometer. Responses obtained (enzyme activities) were used for a quadratic polynomial model, ANOVA and response-surfaces construction, using the Design-Expert 11-Stat-Ease^{MR} program. The optimal values of the variables for maximum PEC (simultaneous Exo-PG, Endo-PG and Exo-PMG, production) were: 1.5% G, 5% MS and 66% H. The maximum enzyme activity values predicted by the model were: Exo-PG: 0.87 U/g; Endo-PG: 0.82 U/g and Exo-PMG: 0.77 U/g. When validating these results with SSF trials, it was observed that the maximum activity was: Endo-PG 26% more than predicted, Exo-PMG 33% less, both at 3.5 days, while for Exo-PG obtained 34% more than predicted after 3 days.

A51

CLONING, OVEREXPRESSION AND PURIFICATION OF MUR ENZYMES OF *BRUCELLA ABORTUS* FOR STRUCTURAL RESOLUTION BY X-RAY CRYSTALLOGRAPHY

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Peptidoglycan's biosynthesis is one of the most studied mechanisms in microbiology. It consists of two stages, a cytoplasmic phase dependent on the Mur protein family and a periplasmic phase controlled by the Penicillin Binding Proteins (PBPs) responsible for the formation of the bacterial cell wall. The PBPs have been the target for the search and design of antibacterials since the discovery of penicillin. However, with the increasing acquisition of resistance mechanisms by bacteria, the introduction of novel classes of antibiotics is necessary. On this line, drugs have been clinically incorporated which alter significantly other elements of the biosynthetic machinery as the case of fosfomicin, an inhibitory antibiotic of MurA. This protein, along with the rest of the Mur family, is responsible for synthesizing the UDP-N-acetyl-D-muramate pentapeptide (or UDP-MurNAc-pentapéptide), the precursor of peptidoglycan. As Mur enzymes are highly conserved in bacteria and there has not been found a crossed reaction with the eukaryotic cells, it is expected that inhibitors of Mur proteins may be potentially broad-spectrum bactericidal compounds. On this work, we focus on Mur proteins of *Brucella abortus*, the etiological agent of Mediterranean Fever, a world-wide distribution zoonosis. We have amplified the MurF gene from *B. abortus* strain with PCR, cloned inside an expression vector pET28a, expressed and purified to homogeneity by QIAGEN® Ni-NTA affinity resin. Now, we are running crystallization assays in order to solve its 3D structure by X-ray diffraction. This would help the rational design of new inhibitors against brucellosis.

A52

LIGNINOLYTIC ACTIVITIES PRODUCTION IN SOLID STATE FERMENTATION USING OLIVE POMACE. SCREENING OF VARIABLES FOR ITS OPTIMIZATION

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Alperujo (AL) is a by-product after the two-phase extraction process of olive oil. AL is semi-solid, and its composition by weight is water 83–96 %, organic matter 3.5–15 %, and mineral substances 0.5–2 %. The organic fraction includes sugars, proteins, organic acids, lipids, pectin, and phenolic compounds (PC). Due to its PC content, AL is often considered as a toxic by-product. In addition, because of legal requirements, it is necessary to carry out a detoxifying treatment prior to its final disposal. Solid-state fermentation (SSF) is a treatment that provides simultaneously detoxification and valorization for AL. Through some preliminary SSF tests, using culture medium made of olive pomace (obtained from AL) and filamentous fungi isolated from AL, microorganism growth, PC consumption, and ligninolytic enzyme production were verified. In the present work, the aim was to study the effects of some

variables in Lignin-Peroxidase (LiP) and Laccasa (Lac) production in solid-state fermentation of olive pomace, obtained from AL. For this, a statistical test was made using Plackett-Burman Design (PBD) for four variables, at two levels (low (-) and high (+)). Variables were X_1 : Aqueous Extract from AL (0 and 1 % p/p), X_2 : Veratryl Alcohol (0 and 0.02 % p/p), X_3 : Initial water content (55 and 65 %), and, X_4 : Mixing (No and Yes). The responses were LiP and Lac activities. The eight SSF essays according to PBD, were performed in a bench-scale fixed bed reactor. This reactor has an internal tray of 28 cm diameter and 10 cm high, which was eight-portion fractioned, for each one of the essays. The reactor was set in a room for 15 days, at controlled 27 °C and humidified forced aeration was provided. The culture medium was prepared with olive pomace (70 % p/p) and grape stalks (30 % p/p); inoculum was 10^7 conidia/g_{dry}. Enzymatic activities were quantified by spectrophotometric methods, using Veratryl Alcohol and ABTS as substrates for LiP and Lac, respectively. Samples for response determination were taken at days 3, 5, 7, 10, and 15. Our results showed, at 7 days (where better enzyme activities were found): For Lac and LiP production, the increase of X_3 variable showed a positive effect, while X_1 , X_2 , and X_4 showed a negative effect when they went from the low level to the high one. In the case of LiP production, X_3 was significant at 80% confidence level, and for Lac, 95%. The rest of the variables showed significance with different confidence levels ranging from 90% to 95% for LiP, and from 80% to 99% for Lac. The main conclusion is that only Initial Humidity Content (X_3) positively affects both enzyme activities. A future essay would be aimed to optimize variables whose effects were positive and to search for new inducers for both enzymes.

A53

ANTIMICROBIAL ACTIVITY AGAINST *ESCHERICHIA COLI* OF NANOPARTICLES OBTAINED BY DIFFERENT SYNTHESIS METHODS

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Escherichia coli is a pathogenic bacterium that causes serious infections, whose therapeutic treatment is threatened by the emergence of multiple resistance to conventional antibiotics. In recent years, metal nanoparticles (NP) have been studied for their antimicrobial capacity and its possible applications as an alternative to antibiotics against different pathogens. The NPs also vary in synthesis techniques either by chemical, physical, and biological based methods. The objective of this work was to study the possible antimicrobial capacity of nanoparticles obtained by chemical and biological methods against *E. coli*. The NPs obtained by a method chemical called citrate-gel (Quim-NP CuNi B1-300; Quim-NP CuNi B1-500; Quim-NP CuNi B3-300) and NPs obtained by a method biological, silver nanoparticles (AgNPs), synthesized by biological mediators such as *Cryptococcus laurentii* (AgNPs-C.I) and *Rhodotorula glutinis* (AgNPs-R.g). The antimicrobial capacity of the aforementioned NPs was evaluated in vitro by means of the agar diffusion method; 200 µL of an *E. coli* (ATCC 8739) suspension, at a concentration of 3×10^8 CFU/mL, were seeded in 10 cm Petri dishes with selective medium and incubated for 1 h at 37 ± 1 °C. Subsequently, wells of 3 mm were made aseptically and were filled with 25 µL of the suspensions of AgNPs-R.g, AgNPs-C.I, CuNi B1-300; CuNi B1-500, CuNi B3-300, and the combination thereof. Distilled water was used with the negative control. Then they were incubated for 48 h at 37 ± 1 °C. After incubation, the zones of inhibition (cm) were measured. The assays were performed in triplicate. Most of the NP evaluated showed antibacterial activity against *E. coli*. When combining chemical and biological NPs, synergistic effects are observed with an increase in antibacterial activity in some cases. We can conclude that NPs derived from chemical and biological synthesis could be used as antimicrobials against *E. coli* and when these are combined, the antibacterial effects increase. In the future, these applications of nanomaterials could be used as an alternative to the use of antibiotics against pathogen-resistant.

A54

COPY NUMBER VARIATION PROFILE OF HEAT SHOCK PROTEINS IN BREAST CANCER SUBTYPES

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Breast cancer (BC) has the highest incidence and mortality rates in women worldwide. Our country is not the exception, with 21558 new cases and 6380 deaths each year. One of the most used criteria to classify BC is the PAM50, which discriminates five intrinsic subtypes: Luminal A, Luminal B, HER2-enriched, Basal-Like, and Normal-Like. The heat shock proteins (HSPs) are expressed at high levels in a wide range of tumors and are closely associated with poor prognosis and resistance to anticancer therapy. The ubiquitous HSPs were identified as proteins required in thermal and other proteotoxic stresses, but nowadays, we know that they participate in various types of events: protein quality control, cell death pathways, and cell-cycle regulation. The HSPs have been implicated in several processes related to cancer and its resistance to therapy. Copy number variations (CNVs) are DNA fragments (1 kb – 5 Mb), which are present in a variable copy number compared to a reference genome. CNVs have a critical role in cancer development and progression. The purpose of this work was the CNV analysis of the HSP family in BC subtypes. The data were retrieved from the publicly available “The Cancer Genome Atlas” (TCGA) database. The segmentation data for de CNV analysis

belong to the Affymetrix Genome-Wide Human SNP Array 6.0 platform, and the CNTools “R” package was used to figure out which HSPs presents amplifications and deletions. Taking all BC patients, we found that five HSPs present amplifications (“CCT3”, “DNAJC5B”, “HSPA6”, “HSPA7” and “ODF1”). On the other hand, after dividing the BC cohort by molecular subtype, we found that Luminal A presented two HSPs amplified (“DNAJA3” and “TRAP1”) and one HSP with deletions (“DNAJA2”). Although Luminal B subtype no present specific amplification, four HSPs present deletions (“CRYAB”, “HSPA8”, “HSPB2” and “HYOU1”). HER2-enriched and Basal-Like subtypes shared amplifications in three HSPs (“HSPA6”, “HSPA7” and “ODF1”) as well as in the other subtypes. Several HSPs present both amplifications and deletions, some of which were subtype-specific, and others were shared in all patients. This work provides useful information to understand the CNV profile of the HSPs genes and facilitates the search for new prognostic markers and therapeutic targets.

A55

STRUCTURAL STUDY OF *TRYPANOSOMA CRUZI* PROTEINS

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Trypanosoma cruzi is the protozoan parasite that causes of the Chagas’ disease in humans. This is an endemic disease in Latin America that affects 18–20 million people. No vaccines are available at present, and drugs used for treatment show undesirable side effects. The identification of new targets for chemotherapy is a major challenge in the control of the disease. X-ray crystallography and complementary methods, principally *in silico* homology modeling, are important tools for structural characterization. Proteins involved in important processes such as protein expression, energetic metabolism, and mRNA maturation are potential targets for the rational design of drugs to treat Chagas’ disease. The ribosomal stalk is a pentameric complex formed by the P proteins: TcP0 and four TcP small proteins (TcP1 α , TcP1 β , TcP2 α , and TcP2 β). Homology models for these proteins have been obtained in our laboratory using the Modeller program. The TcP0 model has an N-terminal globular domain, an alpha domain, a disordered region, and a C-terminal negative tail. The TcP small proteins have an N-terminal four-helix up-and-down bundle domain and a C-terminal disordered domain, which ends in an acidic bend. Docking assays using HDOCK and PISA interaction results for the protein-protein interfaces suggested the formation of the following stable dimers: TcP1 α -TcP2 β , TcP1 β -TcP2 α , TcP1 α -TcP2 α , TcP1 β -TcP2 β , TcP2 α -TcP2 α , and TcP2 α -TcP2 β . The H1 and the H2 helices were oriented in an antiparallel manner, forming a 25 ° angle to form strong H1-H1 and H2-H2 associations. A pentameric model proposed in this work shows TcP1 α -TcP2 β and TcP1 β -TcP2 α dimers bounded to the TcP0 α -domain. Nucleoside diphosphate kinases are important hexameric proteins involved in *T. cruzi* energetic metabolism. The X-ray crystallographic structure for TcNDPK1 shows a helical multi-hexameric oligomer. They suggest a model for enzyme regulation and storage. TcCPSF30 and TcFIP1-like are nuclear proteins that are involved in mRNA cleavage and polyadenylation processes. These proteins are associated *in vivo*. We have shown that TcCPSF30 and TcFIP1-like are intrinsically disordered proteins (IDPs). These types of proteins are very difficult to crystallize and, therefore, to be solved by x-ray crystallography. A homology model for the TcCPSF30-TcFIP1-like interface region was obtained, and structural interaction was characterized. The structural particularities proposed in this work are a potential target for drug design.

A56

PRODUCTION OF *KOSAKONIA RADICINCITANS* IN CULTURE MEDIA FORMULATED FROM BAGASSE OF BEER

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In previous works, we show the biotechnological applications of *Kosakonia radincitans*. That is why the need arises to develop an economic culture medium for its large-scale production. The increase in craft breweries currently allows for a by-product, such as bagasse. This residue of the production process is the filtering and pressing of malt grains. Currently, in our country, there is not much use that is given to this waste. For this reason, an alternative is sought for reuse and thus optimizing the production of beer and *K. radincitans* (Circular Economy). The objective of this work was to develop a culture medium with a novel substrate such as beer bagasse, for the production of *K. radincitans*. Weighed 250 g of bagasse 900 mL of distilled water was added. It was processed to release the remaining sugars and sterilized. Two different liquid culture media were tested from beer bagasse, the first formulated with the addition of 5 g/L yeast extract (a) and the second without the addition of any substrate (b). A base medium prepared from commercial components (glucose 10 g/L; yeast extract 5 g/L) was used as control (c). A suspension of *K. radincitans* was standardized to a concentration equivalent to the 0.5 Mc Farland turbidity scale. An inoculum of (0.1 mL of the bacteria suspension) was seeded in 100 mL of the respective culture media. It was incubated in an oven at 28 °C a stirring of 120 rpm for 5 h. Subsequently, to quantify the growth, serial dilutions were made and seeded on plates containing counting agar. The bacteria reached a growth of 7.8×10^6 CFU/mL in the bagasse-based medium with the addition of yeast extract (a); the medium containing only

bagasse the microbial density was 6.2×10^5 CFU/mL. In the control trial (c) development of 9.5×10^6 CFU/mL was obtained. They did not show significant differences between the culture medium made from commercial components and formulated from the bagasse of beer with yeast extract. With these results, we can affirm that an economic medium with bagasse can be formulated for the production of *K. radicincitans*. The possible alternatives to enrich this beer by-product and obtain a better production of the microorganism of interest constitute future research perspectives.

A57

STUDY OF A PSEUDOSTATIONARY PHASE BASED CARBON NANOTUBES FOR THE SEPARATION OF EMERGING POLLUTANTS

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Drugs are emerging contaminants that cause greater concern, due to the fact that their consumption is massive, they are poorly biodegradable and continuously discharged without the possibility of being eliminated by the purification systems. Some antibiotics, analgesics, and anti-inflammatories are included in priority lists of hazardous substances. It has been shown that its presence in aquatic environments can cause ecotoxicity. The development of analytical methods that alert the presence of drugs in the environment is fundamental. Capillary electrophoresis (CE) is a technique with a great projection that offers a very interesting alternative to liquid chromatography. Taking into account its global high consumption, the studied drugs were paracetamol, naproxen, piroxicam, ketorolac, trimethoprim, sulfamethoxazole, cephalixin, and amoxicillin. PSP consisted of 20 mM sodium tetraborate buffer, pH 9.4, with carboxylated carbon nanotubes (c-MWNTs) was studied. A voltage of 12 kV was applied, and the detector was set at 190 nm. The results show the interactions between the analytes and the c-MWNTs resulted in an increase in migration time. On the other hand, as expected, we could not achieve a good separation of the analytes with a fused silica capillary without PSP. Under the optimal conditions, we were able to separate and determine the studied drugs in less than 20 min. The CEC procedure developed in this work was precise, reproducible, and sensitive. The advantages of this method include simplicity with high selectivity, as well as low cost and efficiency through the use of c-MWNTs as a pseudo-stationary phase, which demonstrates a great potential for application to the analysis of emerging contaminants in environmental samples.

A58

GREEN FLUORESCENT PROTEIN TRANSFORMATION OF *YERSINIA ENTEROCOLITICA* SEROTYPE O:3

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Yersinia enterocolitica (Ye) are Gram-negative bacteria that cause foodborne acute or chronic gastrointestinal and systemic diseases. Ye serotype O:3 is the most frequent cause of human yersiniosis and the most associated with postinfectious extraintestinal sequelae such as reactive arthritis. To analyze the pathogenic mechanism of these sequelae, identification of the Ye persistence requires the tracking of the bacteria by an easily detectable marker. Although Green fluorescent protein (GFP) transformation systems are well established for many enterobacteria, there are difficulties in establishing such transformation in the genus *Yersinia*, particularly in the serotype O:3. In this study, we labeled Ye O:3 with a GFP vector, containing chloramphenicol as a selection marker, using an electroporation protocol. Competent bacteria were prepared, and different electroporation parameters were tested: voltage (2 or 2.5 kV) and plasmid concentration (0.5 or 2 µg). The bacterial growth rate was studied in the presence of chloramphenicol at 25 or 37 °C. The *in vitro* GFP expression was analyzed in UV-transilluminator and by flow cytometry. In addition, the stability of the resident *Yersinia* virulence plasmid (pYV) was checked by autoagglutination and calcium-dependence assays. Furthermore, *in vivo* GFP expression was investigated by an oral infection in C57BL/6 wild-type and *TNFRp55*^{-/-} mice. We found that the highest electric-field strength (2.5 kV) and the lowest plasmid concentration (0.5 µg) resulted in greater yields of transformants. No adverse effects were observed in the growth behavior of the labeled strain compared to wild type (parental) strain. Transformed bacteria expressed GFP at both 25 and 37 °C. The foreign GFP plasmid did not alter the expression of pYV-encoded virulence factors. At day 5 after oral infection with GFP-Ye O:3, fluorescent colony-forming units were detected in Peyer patches, mesenteric lymph nodes, and spleen of mice. We conclude that GFP-Ye O:3 maintains the characteristics of the parental strain required for its use in *in vivo* tracking.

A59

KINETICS OF BIOGAS PRODUCTION IN HIGHLY CONCENTRATED ALPEORUJO, AT LAB AND BENCH SCALE

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“Alperujo” (AL) is the waste produced in two-phase decanter olive-oil mills. This waste is a potential raw material for biofuel production. Biogas production, by dry anaerobic digestion (DAD), is an environmentally friendly way. The aim of the present work was to compare the kinetics of biogas production at two different scales (A: 40 mL and B: 160 L), in dry anaerobic fermentation, using highly concentrated AL. For A-DAD, 60 mL syringes were used, containing 40 mL of AL (60% water content), inoculated with 4 mL of anaerobic isolation (volatile solids 0.225 g/mL, pH 7.64), and incubated at 32 °C for 15 days. Biogas and methane produced were quantified by pressure enhancement. For B-DAD, 200 Liters bioreactor was set-up, charged with 160 liters of AL, inoculated, at 10% (v/v) using the same inoculum explained before. Samples were taken periodically from A-DAD and B-DAD, and pH, volatile solids, total solids, and methane produced, were measured. Results presented methane production (L/Kg_{TS}) as a function of time for A-DAD and B-DAD were adjusted to a Sigmoidal modified Gompertz model ($R^2 = 0.9847$ and 0.8415 , respectively), using the SigmaplotTM software. The kinetic parameters found by the software are for A-DAD: Maximum methane accumulated $P_{max} = 0.0688$ L/Kg_{TS}, maximum production rate: $R_{max} = 26.86$ L/Kg_{TS}h, and lag time, $\lambda = 75.95$ h; for B-DAD: $P_{max} = 26.8$ L/Kg_{TS}, $R_{max} = 291.46$ L/Kg_{TS}h, and lag time, $\lambda = 559$ h. It can be concluded that the behavior of methane production is adjusted in an appropriate way to the Modified Gompertz Model for both scales. Differences in these constants may be given due to rheological changes and mass transference problems. When methane produced at B-DAD is affected by scale factor $f = 1/1000$, the kinetic shows clearly two periods. The first stage was similar for the methane obtained at A-DAD; the second stage presented a pick shape. The main conclusions are: it was possible to obtain kinetics for biogas production in high concentration AL, at lab and bench scales; at 200 L bioreactor (B-DAD), kinetics presented two-stage shape, and methane production and time consumed reached three and four times more than A-DAD, respectively.

CLINICAL MEDICINE AND ODONTOLOGY

A60

DEMINERALIZATION OF THE ENAMEL FOR A DRINK MORPHOLOGICAL AND MECHANICAL CHARACTERIZATION

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The mineral content of tooth enamel gives mechanical properties, although it makes it susceptible to demineralization against acidic agents. The consumption of non-alcoholic commercial drinks is a frequent eating habit in the population. It has aroused interest in the dental scientific community for the increased risk of erosion injuries. Despite the vast information on the action of different drinks on tooth enamel, there are not enough studies describing the variations in the microstructure of tooth enamel according to its organization in the external and internal zone. Our purpose was to characterize the morphological and mechanical aspects of radial enamel with Hunter-Schreger Bands exposed to *in vitro* flavored water. Longitudinal sections of dental crowns were obtained and were included in polymer, worn and polished. The observations were made by the ESEM FEI QUANTA 200 (SeMFi-LIMF. FI-UNLP) and nanodurty tests (INTEMA-CONICET-UNMdP) in the radial enamel and with bands, before and after the treatment. The immersion in flavored water was for 12 min. In the morphological description, the acid etching patterns of the enamel were considered. The hardness, elastic modulus, and contact depth of the indenter were recorded. At ESEM, the prisms presented different engraving patterns. The surface hardness decreased being enamel with more vulnerable bands. The flavored water used contains natural mineral water, citric acid and EDTA. Citric acid is a weak organic acid, and EDTA (ethylene-diamino tetraacetic acid) also has a chelating action and forms stable compounds with calcium. We conclude that flavored water, despite being natural mineral water, produces demineralization of the enamel compatible with an erosion lesion. The inner enamel zone with Hunter Schreger bands is more vulnerable to mineral loss.

A61

MUCINOUS CYSTADENOCARCINOMA OF THE APPENDIX: CASE REPORT AND REVIEW

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Mucinous cystadenocarcinoma of the appendix is an infrequent entity, which comprises only 4–6 % appendix neoplasm and 25% of appendix carcinomas. It is usually well-differentiated and presents a slow progression. Although the incidence of this neoplasm is extremely low, an accurate preoperative diagnosis is essential for proper surgical treatment. The tumor presents a slightly higher incidence in females (2:1) and usually occurs during the sixth decade of life. Frequently, patients with malignant appendiceal tumors develop symptoms and signs indicative of acute appendicitis. We present the case of a 34-year-old male patient who manifested pain

localized in the lower right abdomen. After complementary studies, laparoscopic appendectomy was performed, and the operative findings showed an appendicular mucocele (13 x 5 cm) with the presence of mucus in the peritoneal cavity. The anatomopathological examination described a mucinous cystadenocarcinoma of the cecal appendix, associated with peritoneal metastasis of mucinous cystadenocarcinoma. Therefore, in a second surgery, right hemicolectomy and intraperitoneal chemotherapy were performed. In addition to the case report, we expose a brief update of the current knowledge about the histopathological features and the molecular biology of this neoplasm.

A62

ANGIOTENSINOGEN M235T POLYMORPHISM AND HYPERTENSION IN A SAN LUIS POPULATION

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Hypertension (HTA) is a polygenic disorder resulting from the interaction of several genetic and environmental factors. Renin-angiotensin system (RAS) gene polymorphisms have relevant participation to develop HTA. The study of RAS polymorphisms is very controversial, and its identification in HTA patients is required. The aim was to investigate a possible association between the M235T polymorphism of the angiotensinogen (AGT) gene and hypertension in a San Luis-Argentina population. A case-control study was performed in 107 hypertensive patients (51% women) and 58 control subjects (67% women) from Juana Koslay Hospital (2014–2015). Blood pressure and body measurements were recorded. A blood sample was obtained and polymorphism AGT M235T was performed by Polymerase Chain Reaction combined with Restriction Fragment Length Polymorphism (PCR-RFLP). Mean age (years): 54.2 ± 9.3 HTA and 39.2 ± 13.7 control ($P < 0.0001$), Body Mass Index (kg/m^2): 31.7 ± 5.3 HTA and 27.1 ± 4.8 control ($P < 0.0001$). Systolic and diastolic blood pressure (mm Hg): 152.4 ± 15.3 / 90.1 ± 9.9 HTA and 118.0 ± 11.2 / 71.0 ± 9.4 control subjects ($P < 0.0001$). We found Hardy-Weinberg equilibrium in all groups studied ($P > 0.05$). The genotype frequency of M235T was: MM 12.1%, MT 48.5%, and TT 39.2% in HTA patients, and MM 15.5%, MT 51.7%, and TT 32.5% in control individuals; no significant difference was found in the patients studied. The allele frequency was M 0.36 and T 0.63 in hypertensive patients and M 0.41 and T 0.58 in control subjects. Chi-square analysis showed the T allele was statistically significant in HTA patients ($P < 0.0002$). The carriers of the T allele had a significantly increased risk of hypertension (OR = 2.47, 95%CI: 1.55–3.92; $P < 0.0002$) in the population studied. The AGT M235T could be a relevant role in the genetic predisposition to develop essential hypertension in the San Luis population.

A63

PHOSPHORYLATED HSP90 α AS PREDICTIVE AND PROGNOSTIC BIOMARKER IN TUMORS FROM PATIENTS TREATED WITH PLATINUM ANALOGS

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The heat shock protein HSP90 α is a ubiquitous molecular chaperone especially required for cancer cells as it chaperones proteins involved in oncogenesis, making it an attractive target for anticancer therapy. Phosphorylated HSP90 α (P-HSP90 α) on the threonine 7 (Thr-7) accumulates at the sites of DNA damage, involving this protein in DNA damage response. In addition, the basal level of P-HSP90 α has been proposed as a surrogate biomarker for genetic instability in tumor cells. Platinum analogs (cisplatin, carboplatin) that are widely used for the treatment of many solid tumors damage DNA by forming covalent adducts. Platinum-DNA adducts are bulky lesions that interfere with DNA replication machinery, resulting in the formation of double-strand DNA breaks. These lesions can lead to genomic instability and cell death. Unfortunately, the development of resistance to platinum-based agents may limit the efficacy of the chemotherapy. Thus, it remains a need for biomarkers of cisplatin-response and prognosis for cancer patients. Our aim was to determine the predictive and prognostic ability of P-HSP90 α in primary tumors from cancer patients who received platinum-based chemotherapy (cisplatin/carboplatin). P-HSP90 α expression was determined by immunohistochemistry in paraffin-embedded tumor tissues from 51 cancer patients before chemotherapy, with a mean follow-up of 19.2 months. The expression of the protein was evaluated according to a staining intensity score and the proportion of positive tumor cells. Clinical response was assessed after the third cycle of chemotherapy. Disease-free (DFS) and overall survival (OS) were periodically determined. Patients with a complete clinical response or partial response to chemotherapy showed nuclear expression of P-HSP90 α in contrast with tumors from patients with stable disease or progressive disease ($P < 0.01$). In addition, patients with a high cytoplasmic proportion of P-HSP90 α had a significantly worse OS ($P < 0.05$). No statistically significant relationship was found between P-HSP90 α expression and DFS. Our preliminary results provide evidence that P-HSP90 α could be a potentially valuable biomarker in predicting response to platinum-based chemotherapy and, may also be useful for defining the prognosis of the disease.

A64

RISK ASSESSMENT OF DIABETES MELLITUS IN THE RURAL POPULATION OF THE DEPARTMENT JUAN M. DE PUEYRREDÓN OF THE PROVINCE OF SAN LUIS

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Type 2 Diabetes Mellitus (DM2) is one of the chronic diseases with greater socio-sanitary relevance because its incidence is increasing in a worrying way. The screening of DM2 by performing analysis in the general population is not fully justified by the wide variability of glycemia and its low cost-effectiveness. Several simple scales have been developed to identify subjects with undiagnosed DM or at risk of developing it in the next 10 years; all offer similar results in sensitivity than oral glucose overload, although worse specificity. A questionnaire used is FINDRISC (FINnish Diabetes Risk SCore), developed by the Diabetes Society of Finland. Our objective was to identify the risk of DM2 development in a population aged 18 or over in a rural area of the Juan M. de Pueyrredón department of the province of San Luis. A descriptive, cross-sectional, observational study was carried out by means of a survey in the rural population grouped from the towns of Zanjitas, Cazador, Alto Pelado, and Beazley. A survey, physical examination and laboratory tests were performed. Weight, height for the calculation of body mass index (BMI), waist and hip circumference, and blood pressure were measured, and blood samples were drawn. Results (%): The physical examination and laboratory tests were carried out in 246 people, 23 were excluded because they were diabetic (9.34%). Sex: Female (F) (68.14), Male (M) (31.86). Age <45 years F (66.88) M (38.89), 45 to 54 F (9.09) M (13.89), 55 to 64 F (8.45) M (18.05) and > 64 F (15.58) M (29.17) Normal BMI <25 F (26.63) M (26.39), overweight between 25 - 29.9 F (30.52) M (31.34), obesity > 30 F (42.85) M (41.67.) Waist perimeter (cm) F <80 (21.42), between 80-88 (12.35) and > 88 (66.23); M <94 (43.06), between 94-102 (18.06) and > 102 (38.88). Physical Activity Yes F (81.17) M (54.17); Consumption of fruits and vegetables every day Yes F (54.55) M (56.95). Take medications for high blood pressure Yes; F (25.97) M (36.11). Physical Activity Yes F (81.17) M (54.17); consumption of fruits and vegetables every day Yes F (54.55) M (56.95). Take medications for high blood pressure Yes F (25.97) M (36.11). High glucose value Yes F (9.9) M (11.11). Family history of DM2 Direct family F (25.97) M (23.62), far family member F (14.29) M (6.94). Risk of suffering from DM2 in the next 10 years according to the test: Low F (20.13) M 31.95, slightly moderate F (35.06) M (25), moderate F (23.38) M (22.22), high F (20.78) M (18.05), very high F (0.65) M (2.78). The population studied was predominantly composed of young adults and females; two-thirds are overweight and obese. Almost half do not consume vegetables and fruits daily. A quarter of women and more than a third of men consume medications for blood pressure. A quarter of the population of both sexes has direct family members with a history of DM2. According to these results, a fifth of our population is at high and very high risk of suffering from DM2 in the next 10 years. The significant proportion of patients at risk of DM2 found in our study is a call for attention to health professionals, in the sense that it is possible to use simple questionnaires such as FINDRISC to identify the population at risk, its application being of special interest to the subgroup of patients with lower sociocultural level.

A65

BIOCHEMICAL EVALUATION OF THYROID FUNCTION IN PEDIATRIC PATIENTS OF SAN LUIS (ARGENTINA)

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Thyroid hormones play a major role in children's metabolism, growth, and pubertal maturation. Thyroid disorders are among the most common youth endocrine diseases. Its prevalence depends on gender, age, ethnic and geographical background, and iodine intake. Therefore, data of thyroid disorders from one population cannot be extrapolated to another. This study was hence conducted to identify the types of thyroid alterations in children and adolescents in our area. It was carried out from 2017 to 2019, including a total of 105 girls and 87 boys (age range 1–15 years, mean 8 ± 4.7 years) that were referred to a clinical laboratory for thyroid function evaluation. Patients under treatment with immunosuppressive drugs or patients suffering from nephrotic syndrome, protein-losing enteropathy, or severe malnutrition were excluded. Thyroid-stimulating hormone [TSH; RV: 0.7–5.97 (1–6 years), 0.6–4.84 (7–11 years) and 0.27–4.2 (>12 years) μ IU/mL], thyroxine (T4; RV: 5.52–12.6 μ g/dL) and thyroid peroxidase antibodies (TPOs; RV: <34 IU/mL) were performed by chemiluminescent immunoassay (Cobas e411-Roche). TSH above the reference range but <10 μ IU/mL with thyroid hormones (TH) remained within the normal range identifies subclinical hypothyroidism (sHT). Overt hypothyroidism (HT) was diagnosed if TSH was >10 μ IU/mL with low levels of TH. Autoimmune thyroid disease (AITD) was defined as the presence of TPOs with concomitant sHT or HT. Reduced TSH and increased HT levels indicate primary hyperthyroidism. Children were also tested for anemia and vitamin D status. Out of the 192 children tested, 17.7% (N=34; 22 girls and 12 boys) showed altered TSH values, being the sHT the most prevalent (N=24; 70.6%) followed by HT (N=7; 20.6%) and hyperthyroidism (N=3; 8.8%). Regarding TPOs, 4.7% of patients (N=9) had positive results (40.6 to 380 IU/mL), of whom: four had AITD, one had hyperthyroidism and four had TSH levels within the reference range. None children with thyroid abnormalities were found to be anemic. The most common associated disease in the children evaluated for thyroid-related disorders was vitamin D insufficiency. The present study serves as a basis for increasing the interest in the study of thyroid function in San Luis children and adolescents. Regular assessment of the thyroid function in pediatric age would help to avoid the serious effects of TH imbalance during a critical period in children's development.

A66

PHYSICAL THERAPY FOR CELLULITE TREATMENT

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Cellulite is a common phenomenon that particularly affects the thighs and buttocks of women. Little scientific evidence exists to support any of the many advertised treatments for it. This study was performed in order to assess the effects produced after the application of a protocol for physical therapy for “compact cellulitis” on twenty women aged 20 to 40 who were attended in San Luis city. Ultracavitator, massage therapy, endermology, and radiofrequency were used ten times, one for each time combined with infrared, for one hour, twice a week for five weeks, during 2016. Using a dermato-functional kinesics sheet to collect the necessary information and evaluating relevant data such as weigh, trochanteric circumferences, and pain. Photos were also taken (in two instances: at the beginning and end of protocol implementation) and observed afterward in order to collect all the information. The media age was 35.10 ± 4.12 years. The results showed that 100% of leg pain in patients decreased. All patients reduced the cellulite grade, improving its orange-peel appearance, in photos. All of them showed a loss in thighs and buttocks circumference measurements. The media of buttocks circumference reduction was 1.4 ± 1.36 cm, and the media of thighs circumference reduction was 2.00 ± 1.91 , both were statically significant ($P = 0.0002 < 0.05$). No statistical difference existed in weight lost ($P = 0.1392 > 0.05$). After analyzing and comparing the data obtained, we conclude that statistically significant improvements were observed in the appearance, pain remission, and measurements obtained after the implementation of the physical therapy protocol for compact cellulitis.

A67

EXPRESSION AND SUBCELLULAR LOCALIZATION OF P73 AS PROGNOSIS IN BREAST CANCER SUBTYPES

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The incidence of breast cancer (BC) is high in Argentina. The optimal treatment is based on proteomic and histological prognostic factors. Based on variations in gene expression, BC patients are currently classified into five major subtypes: luminal A, luminal B, ErbB2+, basal-like, and normal-like. We previously reported that the methylation of TP73 differed significantly between molecular subtypes. Luminal A tumors frequently exhibited unmethylated TP73; in contrast, all basal-like tumors exhibited methylated TP73. The P73 protein has numerous isoforms with different biological and antagonistic functions. The goals of our study were to determine the expression and subcellular localization of the P73 isoforms, its relationship with TP73 methylation, and their clinical prognostic significance in BC subtypes. For this, we evaluated the expression and subcellular localization of TA-p73 and Δ Np73 isoforms in 137 invasive breast tumors and three BC cell lines by immunohistochemistry and immunofluorescence, respectively. The results showed an exclusive nuclear localization of TA-p73 isoform in 88.9% of luminal BC tumors expressing this isoform. In contrast, basal-like and ErbB2+ tumors mainly exhibited cytoplasmatic expression of TA-p73 isoform in 69.9% and 72%, respectively. The Δ Np73 isoform was mainly expressed in the cytoplasm of all breast tumors with different molecular subtypes. Concomitant, TA-p73 isoform was localized almost exclusively at the nucleus of luminal BC cell lines, while it was mainly expressed in the cytoplasm of the basal-like cell line. Altogether our data suggest that expression and subcellular localization of p73 isoforms are useful factors as prognosis in breast cancer subtypes.

A68

EVALUATION OF SELECTIVE IGA DEFICIENCY IN CHILDREN AND ADOLESCENTS WITH GASTROINTESTINAL MANIFESTATIONS OF GLUTEN SENSITIVITY

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Selective immunoglobulin A deficiency (SIgAD) is the most common primary antibody deficiency. It is defined as a serum IgA level below or equal to 0.07 g/L in presence of normal levels of other immunoglobulin isotypes in children over 4 years of age. IgA deficiency appears to be a risk factor for infections, allergic diseases, autoimmune conditions, and malignancy. Most of the patients are still undiagnosed. Since gastrointestinal tract dysfunction is one of the most common immunodeficiency manifestations, we aimed to know the incidence of SIgAD in children and adolescents who come to our laboratory to evaluate malabsorption gastrointestinal symptoms. The study included 123 male and 134 female aged 4 to 18 years (mean 11 ± 4.3 years), who attended a private laboratory between June 2018–April 2019. Patients less than 4 years of age, with hypogammaglobulinemia or treated with immunosuppressants or cytostatics were excluded. Serum IgA and IgM levels were measured by immunoturbidimetry (Cobasc-311, Roche), IgG by single radial immunodiffusion (Biocientífica) and IgE by electrochemiluminescence (Cobas e-411, Roche). The results are interpreted according to the age of patients. Total SIgAD (undetectable levels of IgA) was found in 5 (1.94%; 1 child, 4

adolescents) and partial SIgAD (detectable levels of IgA, but below the lower limit of normal in 17 (6.6%; 11 children, 6 adolescents) of evaluated immunological patients. Regarding the coincidence of SIgAD with autoimmune diseases, one case of celiac disease and two cases of autoimmune thyroiditis were detected. Evaluation of SIgAD deserves consideration in the differential diagnosis in patients presenting an autoimmune disorder. Besides, they should be monitored over time because some of them may progress to common variable immune deficiency, associated with poorer prognosis. The implication of this work is to draw attention to the importance of the diagnostic approach to IgA deficient pediatric patient and relevance of individual diagnostic methods knowledge as well as to the proper interpretation of the results.

A69

METHODOLOGICAL ASPECTS OF THE STUDY ON CARDIOVASCULAR AND METABOLIC PROFILE IN RURAL POPULATION OF THE PROVINCE OF SAN LUIS

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Type 2 Diabetes Mellitus (DM2) and cardiovascular diseases integrate the Chronic Non-communicable Diseases (NCDs), which account for more than 60% of the disease burden globally and nationally. There is a lack of knowledge about the behavior of morbidity due to NCDs in rural areas of Argentina. The objective of this type of work was to develop a cross-sectional study in a grouped rural population of San Luis to describe the morbidity profile due to NCDs. The Population of reference constitutes persons of both sexes of 18 years and more of the localities of Beazley, Zanjitas, Alto Pelado, and Cazador of the department Juan Martín de Pueyrredón of the province of San Luis. The work was carried out in two stages: a validated survey questionnaire was applied at home in the first one, the second one included anthropometric measurements, FINDIRSC questionnaire (FINnish Diabetes Risk SCore), and blood collection in health centers or schools. Sociodemographic, psychosocial, and risk factors variables of NCDs were obtained by self-report. Physical measurements included body mass index (BMI) and waist circumference, the laboratory tests were baseline glycemia, glycosylated hemoglobin, and lipid profile. Data collection covered the months from September to November 2017. Summary measures were calculated for quantitative variables, univariate and bivariate analyses were performed, 95% CI (Confidence Interval) were calculated. The risk of developing DM2 for the next 10 years was estimated according to test FINDIRSC. Epi Info and R software were used. Participants signed informed consent and they were explained that their participation is voluntary by verbal informed consent. The study was approved by a research ethics committee. The following exclusion criteria were established to be under 18, have a residence time in the place less than 6 months, take systemic corticosteroids in chronic form, and be pregnant. 378 people agreed to participate in the study, after data cleansing, 375 participants met the inclusion criteria. 252 performed laboratory analysis (67.20%) and 250 physical examinations (66.67%). The performance of the respondents was 46.41%. The research pioneered Argentina in providing information on morbidity and risk factors for NCDs in a rural area of the country, a primary requirement for planning specific policies for the rural population.

A70

PROGNOSTIC VALUE OF BRCA1 IN BREAST CANCER PATIENTS TREATED WITH NEOADJUVANT ANTHRACYCLINE-BASED CHEMOTHERAPY

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Breast cancer is the leading cause of cancer death in women in Argentina. Anthracycline-based regimens represent the major chemotherapeutic agents used in breast cancer treatment. As DNA-damaging agents, anthracyclines induce DNA single and double strands breaks which are repaired by homologous recombination (HR) mechanism. The expression of DNA repair proteins varies among breast tumors and the DNA repair status can affect the chemotherapy response. The aim of our work was to determine the predictive/prognostic value of HR proteins CtIP and BRCA1 in breast cancer patients treated with neoadjuvant doxorubicin (DOX) or epirubicin (EPI) monochemotherapy. BRCA1 and CtIP expression were evaluated by immunohistochemistry in pre- and post-chemotherapy tumor biopsies (N=32) and compared with the tumor response and the patients' follow-up. BRCA1 expression increased in tumor cells after DOX/EPI administration whereas CtIP expression significantly decreased after chemotherapy. No associations were found between DNA repair proteins expression and the clinical and pathological response to neoadjuvant chemotherapy. However, BRCA1 expression (>10% of tumor cells) in the post-chemotherapy biopsies was associated with better DFS and OS ($P < 0.001$ and $P < 0.05$, respectively). Our results indicate that BRCA1 expression may be a useful prognostic marker in breast cancer patients treated with neoadjuvant anthracycline-based chemotherapy followed by cyclophosphamide, methotrexate, and 5-fluorouracil.

A71

HSP27 AFFECTS DNA DAMAGE RESPONSE IN HUMAN COLON CANCER CELLS TREATED WITH CISPLATIN

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HSP27 (HSPB1) belongs to the heat shock protein family with fundamental roles in protein homeostasis, transport processes, and signal transduction. HSP27 as an anti-apoptotic protein is over-expressed in many cancer cells. It has been also involved in cancer progression, resistance to cancer therapy and prognosis. Accordingly, HSP27 has become an attractive therapeutic target. Previously, we reported that HSP27 interacts with DNA mismatch repair proteins especially after cisplatin (cPt) treatment. However, the role of HSP27 in the response to cPt-induced DNA damage through the ATR-CHK1 pathway in mismatch repair (MMR) proficient/deficient tumor cells remains unknown. Here, using cultured human colon cancer cell lines HCT116+ch2 (MMR deficient, MMR-) and HCT116+ch3 (MMR proficient, MMR+) exposed to 10 μ M of cPt for 24 h, we explored the DNA damage by comet assay and the expression of phosphorylated CHK1 (pCHK1, Ser345), γ H2AX and pHSP27 (Ser78) by western blot. Cells were collected at T0 (immediately after cPt treatment), T3, T9, and T24 (3, 9, and 24 h post-cPt). Downregulation of HSP27 was obtained using the antisense oligonucleotide (OGX427) in a group of cells before cPt exposure. HSP27 expression was reduced by 50% by OGX427. After cPt treatment, HSP27 and γ H2AX expression increased progressively from T0 to T24 in MMR- cells; but MMR+ cells showed constant expression levels of HSP27 and increased levels of γ H2AX with a significant peak at T3 ($P < 0.001$), which decreased to basal levels at T24. pCHK1 showed similar levels from T0 to T24 in MMR- cells and followed a similar behavior than γ H2AX in MMR+ cells. Downregulation of HSP27 significantly reduced the expression of γ H2AX and pCHK1 ($P < 0.001$) and cPt-induced DNA damage in MMR+ tumor cells. Interestingly, pHSP27 increased at T3 and T9 after OGX427+cPt in both cell lines (MMR-/+). In addition, in cPt-treated MMR-/+ tumor cells, we verified by confocal microscopy the nuclear colocalization of HSP27 with p-CHK1 and CHK1 at T3 and T9. The Pearson correlation coefficient (PCC) was > 0.6 and Mander's overlap coefficient (MOC) was: MOC1 > 0.6 at T3 and > 0.8 at T9; and MOC2 > 0.7 at T3 and T9. No significant differences in the colocalization coefficients were found between both cell lines. Our preliminary data indicate that HSP27 might be involved in the DNA damage response associated with cPt treatment through the CHK1 pathway. Downregulation of HSP27 by OGX427 affects the levels of DNA damage and pCHK1, especially in MMR proficient tumor cells. Further studies will be needed to establish HSP27 effects on cPt sensitivity upstream or downstream the kinase CHK1.

PHARMACOLOGY AND TOXICOLOGY

A72

ARISTOLOCHIA ARGENTINA AND TRIPODANTHUS FLAGELLARIS: SPERM ABNORMALITY TEST

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Aristolochia argentina (Aristolochiaceae family) and *Tripodanthus flagellaris* (Cham. & Schldt.) Tiegh. (Loranthaceae family) are medicinal plants popularly known as “charrúa”, “charruga”, “patito”, “buche de pavo” and “liga blanca”, “liguilla”, respectively. The aim of the study was to evaluate the effect of *A. argentina* lyophilized extract (AALE) and *T. flagellaris* lyophilized extract (TFLE) on sperm parameters in mice. Evaluation of damage happened in the germinal cells is the assay based on the Wyrobek and Bruce morphology approaches. Infusion of the plant material (10%, Del Vitto LA 9258 and Del Vitto LA & EM Petenatti 8553, UNSL) was prepared, separated by filtration and the aqueous extract was concentrated and lyophilized to preserve it. Spermatogenesis is an important process for the species continuous; one of its characteristics is that apparent great resistance to the damage. The proposed method used adult male mice with a body weight range from 25 to 30 g in a randomized manner. Distilled water was given to the mice for five days as a negative control. AALE and TFLE were dissolved in distilled water and administered orally, daily for 5 days, at a dose of 250 mg/kg, respectively. Thereafter, all mice were maintained on a basal diet for 34 days. The animals were euthanized by cervical dislocation on the 35th day after the first dose. The epididymis were excised and placed in physiological saline and minced with ophthalmic scissors. Smears were prepared on clean slides, fixed with methanol, and stained with 2% eosin. The morphological evaluation for sperm was carried out using a high magnification microscope. The number of abnormal sperms in 1000 sperms was recorded and abnormalities (abnormal heads as banana form morphology, amorphous, without hook and two tails) were counted. The frequencies of sperm abnormalities for AALE and TFLE were $8.2 \pm 0.80\%$ and $7.8 \pm 1.11\%$, respectively. There were no significant differences between dose 250 mg/kg each extract and the negative group ($7.2 \pm 0.81\%$, $P > 0.05$). A Chi-square test was used. The sperm abnormality test was a method of evaluating genetic injury to male reproductive cells of the mammals *in vivo* tests. The present test showed that the frequencies of sperm abnormalities were not significantly affected by *A. argentina* and *T. flagellaris*. The plants studied were found to be non-genotoxic in the mouse sperm abnormality test when orally administered to mice.

A73

DETERMINATION OF QUANTITATIVE MICROGRAPHY PARAMETERS IN *EUCALYPTUS GLOBULUS* LABILL. AND *EUCALYPTUS CINEREA* F. MUELL. EX BENTH

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During the routine pharmacobotanical quality control performed on samples of medicinal herbs labeled "Eucalyptus", marketed in the local market, the substitution of the official drug *Eucalyptus globulus* Labill. encoded by the National Pharmacopoeia Argentina 7th edition (FNA) was detected, with another Myrtaceae, *Eucalyptus cinerea* F. Muell. ex Benth. Its essential oils, with balsamic properties, contained in leaves, bark, and fruits, are used as antiseptics of the respiratory tract, expectorants and mucolytics, and administered in various pharmaceutical forms. It is used in the form of pills, inhaling candies, infusions, syrups, or in vaporizations. The oil is used topically as a treatment for muscle and joint pain, as well as to treat cold sores. The most frequently cultivated species in San Luis are *Eucalyptus globulus* Labill. "eucalipto medicinal" and *Eucalyptus cinerea*. "Eucalipto medicinal" is used in medicine for the treatment of respiratory conditions, as decongestant and expectorant. Given the importance that these species have acquired and lack pharmacobotany information, macro-, micromorphological, and quantitative micrographic studies are being carried out. This work was carried out to contribute to the botanical characterization and quality control of the commercial drugs. Plant material was obtained from the local markets of San Luis. Voucher specimens are preserved at Herbarium UNSL (*Eucalyptus globulus*: #533 and *Eucalyptus cinerea* #534). Techniques were performed by cut, paraffin embedding, staining, diaphanization, and dissociation. For counting the micrographic parameters, the samples were diaphanized according to the Dizeo technique. Photomicrographs were obtained with a Leitz DMRB microscope with a digital camera. Micrographic studies were performed by obtaining photomicrographs, aimed at the reliable identification of the medicinal plants involved obtaining significant differences ($P < 0.001$). The species can be distinguished between them both by their exomorphological and anatomical characters, as well as by their quantitative micrographic characteristics. This study contributes to effective quality control mainly in drugs crushed, milled, or reduced to powder.

A74

GASTROINTESTINAL ACTIVITY OF *CYCLELEPIS GENISTOIDES* IN RATS AND MICE

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Cyclelepis genistoides D. Don (Asteraceae) is an Argentinean endemic shrub, known by the vernacular name "palo azul" or "matorro negro". It is widely used in folk medicine as a diuretic and antirheumatic agent. The aim of the study was to evaluate the activity of *C. genistoides* on small intestinal transit in mice and the gastroprotective effect of this plant in rats. The plant was identified by Dr. L. A. Del Vitto, Universidad Nacional de San Luis, Argentina, and the voucher specimen (Del Vitto et al. 8950-UNSL) has been previously deposited at the UNSL Herbarium. The infusion of *C. genistoides* 10% was prepared according to Argentinean Pharmacopoeia. Adult albino Wistar rats (200–250 g) and mice (20–25 g) were used. They were housed in standard environmental conditions and fed with rodent diet and water *ad libitum*. The animals were randomly assigned to different groups. The experiments were carried out following Provision ANMAT No. 6344/96. Experimental protocols were approved by the Institutional Committee for the Care and Use of Animals (CICUA) of the Chemistry, Biochemistry and Pharmacy Faculty, National University of San Luis, Argentina. Gastric lesions were produced according to the method by Robert et al., (1979). All rats were housed in wire mesh-bottomed cages throughout the study to prevent coprophagy. The necrotizing agent absolute ethanol was administered orally (1 mL/animal), and 1 h later the animals were euthanized by inhalation of carbon dioxide. The stomachs were removed, opened along the greater curvature, and washed gently with ice-cold saline solution. The scanned images of the stomachs were analyzed using a program developed by NIH. Absolute ethanol produced gastric ulcers in all the animals treated. *C. genistoides* (500, 750, and 1000 mg/kg) prevents the formation of gastric lesions induced by absolute ethanol ($P < 0.001$ vs. absolute ethanol). In another experiment, the effect of *C. genistoides* on small intestinal transit in mice was tested using the charcoal method. Mice were fasted for 18 h prior to starting the experiments and had free access to water. The animals were pretreated orally with *C. genistoides* at a dose of 125 and 250 mg/kg. The charcoal meal was administered 30 min after the administration of *C. genistoides*. A control group was established, which was administered the same volume of vehicle. Mice were euthanized by cervical dislocation and the small intestine was rapidly removed and laid out on white filter paper for inspection and measurement of distances traversed by the charcoal. The length traversed by the charcoal marker was calculated as a percentage of the intestine length. At the doses studied no significant change was observed on small intestinal transit. The present study showed that the infusion of *C. genistoides* possesses a gastroprotective effect in an *in vivo* study, whereas intestinal transit did not change in this experimental model.

A75

SCAVENGING OF INTRACELLULARLY PRODUCED HOCl WITH RESVERATROL PROTECTS INSULIN SIGNALING IN ADIPOCYTES

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Resveratrol is a cell-permeable *trans*-stilbene derivative that can scavenge intracellularly produced HOCl (J. Biol. Chem, 285:20062/71). Our diet-induced obesity B6-mouse model showed that epididymal AT expresses myeloperoxidase (MPO) in adipose tissue macrophages (ATM) that also express M1-pro-inflammatory markers (e.g., IL-6, iNOS). MPO expression was observed in ATM located in the typical crown-like structures; and also inside adipocytes. Tissue fractionation showed MPO mRNA in the ATM, but not in the stromal-vascular fraction. Treatment of these isolated adipocytes with H₂O₂ blocked glucose uptake. Based on these findings, we hypothesized that MPO interferes with AT insulin signaling by producing HOCl and causing oxidation of intracellular components involved in insulin-triggered signaling. To test this hypothesis, we differentiated human adipocytes and loaded them with human MPO. Treatment of MPO-loaded adipocytes with H₂O₂ caused intracellular production of HOCl, reduced insulin-triggered GLUT-4 translocation to the membrane, and glucose uptake. Furthermore, 5,5-dimethyl-1-pyrroline *N*-oxide (DMPO)-based immuno-spin trapping (Biochim Biophys Acta. 2014;1840(2):722-9) and MS-tandem showed radicalization of specific components of the insulin signaling (e.g., IRS-1/2, SOCS3, GLUT-4). These effects were prevented by resveratrol, but not by taurine or methionine because these cannot pass across the membrane. Together our findings suggest that resveratrol can protect insulin sensitivity in the obese AT by scavenging intracellularly produced HOCl. These findings are also in agreement with the role of resveratrol in protecting genomic DNA against intracellularly produced HOCl in MPO-loaded epithelial cells. Supported by: PICT-3435, PIP-916, PROICO-100218/023418, and PUE-013.

A76

VALIDATIONS AND ESTABLISHMENT OF USES OF *HEDEOMA MULTIFLORUM*, *LIPPIA INTEGRIFOLIA* Y *SATUREJA PARVIFOLIA* IN SAN LUIS

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The Argentine flora includes numerous valuable species for human and animal therapy. The applications of native medicinal plants, consecrated by centuries of employment and popular tradition, are of paramount importance to guide pharmacological studies. These studies must be based on solid pharmacobotanical bases, particularly in terms of taxonomic identity and anatomical characteristics, which allow differences to be established between nearby species, even with substituents or adulterants of genuine drugs. The objective of the work was to conduct ethnobotanical studies of native medicinal aromatic species: *Hedeoma multiflora*, *Lippia integrifolia*, and *Satureja parvifolia*. The plant material that was collected in various areas of the province of San Luis, conversations were also held with residents of the community through semi-structured interviews to collect qualitative and quantitative data about the main plants used, who uses them, attributed use, forms of use and for what diseases they are used. The herbalization process was carried out using techniques for botanical samples and the species was characterized. Data were analyzed using ANOVA (quantitative data) or a Chi-squared test (qualitative data). The form of consumption is greater in infusion according to what is mentioned by the Argentine pharmacopeia. The way of obtaining medicinal plants is varied but common among the species studied (field, pharmacies, herbalists, etc.). The recommendation for the use of medicinal species is from a relative, that is, the tradition of passing from generation to generation is conclusive with the ethnobotanical study. The educational level goes hand in hand with the use of *C. odorum* and *L. integrifolia*. The collection of popular knowledge and the rescue of the traditional use that is transmitted from generation to generation of the sampled species associated with applied research studies will allow the sustainable use and conservation that these species require.

A77

SOY BASED DIET INFLUENCES IN HEMATOLOGICAL MODIFICATIONS INDUCED BY CADMIUM INTOXICATION IN RATS

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Cadmium (Cd) is a toxic metal and it is considered an important environmental contaminant. Furthermore, it is widely known that consumption of soy protein has beneficial health effects. We studied cadmium effects on hematological parameters under different

diets in 4 lots of female Wistar rats: 2 lots received casein (Cas) and 2 lots received soybean flour (So) as protein sources. Within each group: 1 lot received regular water (control-Co) and the other 15ppm of Cd in drinking water for 60 days. Blood samples were collected in heparinized tubes, and they were analyzed using a hemocytometer ADVIA 360 (SIEMENS) and an autoanalyzer A15 (BIOSYSTEMS). ANOVA test was used for statistical analysis. Red Blood Cell Count (RBC) decreased in SoCo vs. CasCo ($P<0.01$), and Medium Corpuscular Volume (MCV) diminished in SoCd vs. CasCd ($P<0.01$). The mean corpuscular hemoglobin concentration (MCHC) was significantly increased in SoCd vs. CasCd ($P<0.05$). Red blood cell distribution width (RDW) decreased in SoCo vs. CasCo ($P<0.05$). Hemoglobin decreased in SoCo vs. CasCo ($P<0.05$). Hematocrit was lower in Soy vs. Cas groups ($P<0.05$). White Blood Cells Count (WBC) decreased in both intoxicated groups ($P<0.05$). No significant differences were found among lymphocytes (LYM), monocytes (MID), and granulocytes (GRA). Nevertheless, MID showed a trend to increase in the CasCd group, and GRA showed the same trend in the SoCd group. Blood glucose and uric acid increased in CasCd vs. CasCo ($P<0.05$), without changes between Soy groups. Alanine aminotransferase (ALT) activity showed no differences among the groups. Aspartate aminotransferase (AST) activity decreased in SoCd vs. SoCo ($P<0.01$). Total cholesterol did not change; nevertheless, HDL-cholesterol showed a decrease in SoCd vs. CasCd ($P<0.05$). These results showed that cadmium intoxication modifies hematological parameters, such as WBC, along with both diets; however, RBC showed changes only in casein-fed animals, while AST only changed in soy groups. Keeping this in mind, we can conclude that the source of dietary protein influences the metabolic and hematological changes induced by cadmium intoxication. In addition, these results provide a wider background to fully understand the alterations that were previously described in the lungs.

A78

QUALITY ASSESSMENT OF TAP WATER FROM AN EDUCATIONAL INSTITUTION OF SAN LUIS CITY BY MEANS OF TOXICITY, CYTOTOXICITY AND GENOTOXICITY ASSAYS

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Safe water is that which is consumed without any bacteriological or chemical risk and must be acceptable in terms of smell, color, transparency, and taste. The quality of the water that enters the distribution system must be preserved until it is consumed. Some of the main causes that contribute to water quality deterioration in the distribution network are the sanitary state of the water in the storage places (cisterns or high tanks), and the conditions of pipelines (pollution, quality, age, materials (lead pipes)); these may yield on microbiological or chemical (inorganic and organic) contamination. The objective of this research was to study the toxicity, cytotoxicity, and genotoxicity of tap water from an educational establishment in the city of San Luis, through the *Allium cepa* test. Seven water samples were taken from faucets located in seven bathrooms of the educational institution. The experimental design included eight treatments, a control group (distilled water), and seven tap water samples, all of them with three replicates. Apical meristems were obtained from bulbs of *A. cepa* exposed to control and treatment solutions for three days. Length of roots was measured and then the classic methods of fixing, staining, and squashing, were performed to obtain the mitotic slides (4 for each treatment). A total of 4000 cells were analyzed for each treatment. Root length was significantly different between treatments ($F_{7,158} = 21.32, P<0.0001$), and was significantly lower in roots exposed to tap water samples 5 and 7 than in control and the other tap water samples. Mitotic Index (MI) was significantly different between treatments ($F_{7,31} = 31.92, P<0.0001$), being also significantly lower in roots exposed to tap water samples 5 and 7 than in control and the other samples. MI in roots exposed to water sample 4 was significantly higher than in control roots. Significant differences were observed between treatments in the percent of chromosomal aberrations (CA) ($F_{7,31} = 2.76, P<0.05$). CA was significantly higher in roots from water sample 2 than in the control. The percent of nuclear aberrations (NA) was also significantly different between treatments ($F_{7,31} = 62.99, P<0.0001$), being significantly higher in roots from water samples 1, 2, 3, 6, and 7 than in all other treatments and control, except treatment 6. The results indicate abnormalities in the growth of roots, cell division, chromosomes, nuclei, and nucleoli. Root growth decreased when cell division and nucleoli were severely affected. AC observed were chromosomal bridges, loose chromosomes, broken chromosomes, C-mitotic effect, sticky chromosomes; and among the AN, micronuclei, nuclear bulbs, irregularly shaped nucleoli, nucleolar material release from the nucleus to the cytoplasm, were observed. Taking into account the literature, the observed effects may be compliant with those caused by metal ions; consequently, it may be appropriate to survey the presence of them in the distribution network of the tap water studied.

A79

USE OF GREEN ALGAE *OOCYSTIS* SP. LIKE A BIOLOGICAL MODEL FOR TOXICOLOGICAL EVALUATIONS OF BDE-209

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Microalgae are photoautotrophic oxygen-evolving microorganisms which include: Chlorophyceae, Cyanophyceae, and Bacillariophyceae. They are morphologically diverse and present extensive distribution. Microalgae reflect a wide range of physiological properties and are tolerant of environmental stress. Therefore, they have been widely used as model species to evaluate different environmental stressors in ecotoxicological tests. The microalgae *Oocystis* sp. is one of the numerous planktonic green algae, usually of reservoir waters. It has characteristics of a model species: rapid growth and it is easy to handle in laboratory conditions. Our goal is to propose to *Oocystis* sp. as a model species for ecotoxicological tests. The samples of the microalgae were

collected with a phytoplankton net from temporary puddles in “El Volcán” town in San Luis province. In the laboratory, the organisms were kept in Bold δ Basal Medium (BBM). The initial biomass (6×10^6 cells/mL) was exposed to contaminants such as polybrominated biphenyls (BDE-209) which toxicity is known, for 96 h. Cell count examination was performed using a Neubauer chamber under Optic Microscope (400X). Chlorophyll stress index and oxidative stress enzymes (ROS) were determined with spectrometry. *Oocystis* sp. showed sensitivity with a 50% growth inhibition with respect to the control from 100 $\mu\text{g/L}$ of BDE-209. Catalase activity increased with values greater than 100 $\mu\text{g/L}$ of BDE-209; while TBARS showed differences only in algae exposed at 100 $\mu\text{g/L}$ of BDE-209. Chlorophyll showed signs of stress and senescence from 100 $\mu\text{g/L}$ BDE-209 (value 0.03 mg/g). *Oocystis* sp. showed toxicity response, which provides relevant information about sensitivity, and they can be used as a model in environmental risk studies to BDE-209.

A80

DISPENSED MEDICINES EVALUATION IN ELDERLY PATIENTS OF PAMI SOCIAL SECURITY IN A SAN LUIS CITY PHARMACY

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Medications consumption evaluation, especially in the elderly, is indispensable to contribute to improving pharmacological therapy in this type of patient. Association between age and drug use, due to the higher prevalence of chronic diseases and comorbidities, is common in the elderly. Our objective was to analyze medicines dispensations in PAMI Social Security patient's in a pharmacy in San Luis. An observational and cross-sectional study of the dispensations in PAMI Social Security patients from March to May 2019 was carried out in a San Luis city pharmacy. Drugs (D) were coded according to the ATC Classification System of the World Health Organization. 3340 D present in 2420 prescriptions were analyzed, corresponding to the following groups: C (35%), A (22%), N (18%), H and M (6%), B (4%). The percentages in descending order, within each group were: C: ACE inhibitors (C09), lipid-lowering agents (C10), and beta-blockers (C07). The most dispensed of the C09 group were losartan and enalapril, followed by valsartan; in the C10 group, rosuvastatin and atorvastatin, finally in the C07 group was carvedilol. A: the most dispensed were: hypoglycemic agents (A10) and antacids (A2); then vitamins (A11) and those of group A03. Within the A10, metformin was the most dispensed; A2 group: pantoprazole, esomeprazole and omeprazole, and ranitidine reached 17%. Vitamin D3 is the most indicated within A11. N: 32% were antiepileptic (N03), 25% Psycholeptics (N05) and 20% Psychoanaleptic (N06), and 14% analgesics (N02). Within N03, clonazepam (54%) and pregabalin (22%) were the most dispensed; in N05, alprazolam (51%); and in N06 memantine and sertraline were the most dispensed. In N02 paracetamol reached a higher percentage. The most prescribed drugs are from the cardiovascular, digestive, and nervous systems, indicating the main pathologies that are prevalent in this age group, such as hypertension, dyslipidemia, diabetes mellitus, and probably insomnia and anxiety. Excessive prescription of proton pump inhibitors in polymedicated and more vulnerable elderly was probably realized by the risk of gastric ulcer appearance, although in most cases this is not justified by the possibility of serious adverse reactions' occurrence. Vitamins are not necessary if the diet is adequate. High dispensing of benzodiazepines is alarming because these drugs are not necessary if is maintained good sleep hygiene. Moreover, these drugs can produce dizziness, and as a consequence, falls and possible hip fractures that can lead to death in the elderly. It is important to reduce its prescription because they implicate an elevated risk in the elderly and a high cost for the health systems.

A81

A NOVEL METHOD FOR HEAVY METALS DETERMINATION IN AEROBIOLOGICAL SAMPLES

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Atmospheric air carries a big number of particles and environmental pollutants coming from anthropogenic activities, to which people are exposed. Metal pollution has been increasing rapidly over the past century in relation to the growth of the human population's contaminants production. Thus, aero particles such as pollen grains may contain various trace elements, and their amounts vary from one species to another. They can present traces of metals such as lead (Pb), barium (Ba), and manganese (Mn), which are among the most common air and soil contaminants, and they are considered toxic to living beings. In the present work, we propose a simple and fast method for the multielement determination of traces in aerobiological samples. The sample of aeroparticles was obtained with a volumetric Lanzoni sensor, which is located on the terrace of the National University of San Luis. This device allows the capture of particles suspended in the air with a week periodicity, and with a suction flow of 10 L of air/min, similar to the volume of air inhaled by the human lung. The pollen content was analyzed for three months with continuous aerobiological sampling, in the atmosphere of the San Luis city, Argentina. Aerobiological samples were observed and readied with an optical microscope at 400X and they were identified through palynological atlases and with the palynothèque belonging to Aerobiology δ laboratory. For multielement determination, a mass spectrometer with inductively coupled plasma (ICP-MS) was used. The aeroparticle samples were subjected to acid digestion at 90 °C for 1 h. The conditions of analytical determination were optimized. The nebulizer gas flow used was 0.85 L/min and the RF power was 1200 W. The pollination period studied was from February to April of 2019. Compared

with other pollen types, Chenopodiaceae and *Artemisia* were the most abundant in the atmosphere of San Luis during the analysis period. Linear regression models were used to analyze the metals vs. pollen content. Positive associations were found between Mn and Chenopodiaceae ($r=0.86$, $P=0.0015$). In the same sense, it was observed positive associations between Pb and Chenopodiaceae ($r=0.78$, $P=0.0026$). The maximum concentration values for Chenopodiaceae (30 pollen grains/m³ of air) were recorded in March, while the maximum values for *Artemisia* (11 pollen grains/m³ of air) were recorded in February. In this study, during the pollination period, an increase in the elemental content was observed. Therefore, the metals could be transported by aero particles such as pollen grains increasing respiratory disorders.

A82

EFFECT OF THE JOINT ADMINISTRATION OF KETAMINE AND FLUOXETINE IN POSITIVE CALBINDINE INTERNEURONS OF THE BASILATERAL NUCLEUS OF AMYGDALA IN *RATTUS NORVEGICUS*: PRELIMINARY RESULTS

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There are different pharmacological treatments for depression, limited in their effectiveness and many have very long latency periods. Current research points to NMDA antagonists as possible therapeutic targets of this disorder. The objective of this work was to assess at a behavioral level the synergistic interactions between the antidepressant of the SSRIs group fluoxetine, and ketamine, a non-competitive NMDA receptor antagonist, and correlate the findings with structural changes in the basolateral nucleus of the tonsil with respect to positive calbindin interneurons. Holtzman rats that were treated with fluoxetine were used. 10, 15, and 20 mg/kg and ketamine. 2.5, 5, and 10 mg/kg and subsequently were evaluated in the forced swim test each group consisted of n=20. Fluoxetine in doses of 10 and 15 mg/kg and ketamine in doses of 2.5 and 5 mg/kg did not produce a significant decrease in immobility time. On the contrary, if significant decreases were observed with fluoxetine 20 mg/kg ($P<0.01$) and ketamine 10 mg/kg ($P<0.05$). Likewise, there was a significant decrease in immobility time when fluoxetine 10 mg/kg + ketamine 2.5 mg/kg ($P<0.01$) and fluoxetine 15 mg/kg + ketamine 5 mg/kg ($P<0.0001$). Twenty-four hours after the test, positive calbindin interneurons of the basolateral nucleus of the tonsil were analyzed by immunohistochemistry. Each group consisted of n=3. It was observed that the saline group presented significant differences with the group treated with 20 mg/kg of fluoxetine ($P=0.001$), with the group treated with 5 mg/kg of fluoxetine ($P=0.0001$) and with the group treated with 5 mg/kg of fluoxetine + 2.5 mg/kg of ketamine ($P=0.0001$), the latter being the one with the least positive calbindin interneurons. Ketamine groups have not yet been analyzed. These results indicate that co-administration of fluoxetine and ketamine can induce a more potent antidepressant activity than when used alone. In addition, the decrease in the density of positive calbindin interneurons in the basolateral nucleus of the amygdala could be directly related to the action of drugs on these neuronal populations, thereby increasing neuronal plasticity and being able to restore excitatory and inhibitory balance.

A83

CENTRAL, PERIPHERAL, AND HEMODYNAMIC EFFECTS OF NANOFORMULATED ANANDAMIDE DURING HYPERTENSION

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Hypertensive lesions –dependent on inflammatory processes– would cause alterations at the central nervous system level with peripheral and hemodynamic consequences. Anandamide (AEA), an endocannabinoid, protects neurons from inflammatory damage. However, the administration of free AEA may produce central side effects, but its controlled release by nanoformulations would reduce or eliminate them. We evaluated possible effects of nanoformulated AEA on blood pressure, central and systemic inflammatory markers, and behavioral alterations in SHR and WKY. We used male rats of 250–300 g (N=7 per group) normotensive (WKY) and hypertensive (SHR) with and without treatment with nanoformulated AEA at a weekly dose of 5 mg/kg IP, for four weeks. We determined systolic blood pressure, expression of WT-1, Hsp-70, AT1, and iNOS in brain tissue, behavior, and blood levels of ultrasensitive PCR, IL-1, IL-6, TNF-alpha, Hsp-70, NADPH oxidase, and nitrites. We use ANOVA II and the Bonferroni post-test ($P<0.05$). The SHR denoted anxiety, overactive locomotor activity, and elevation of inflammatory and oxidative stress markers compared to WKY. At the cortical brain level, the SHR showed greater expression of WT-1, AT1, and iNOS and lower expression of Hsp70. The treatment with nanoformulated AEA significantly reduced systolic blood pressure and expression of WT-1, AT1, and iNOS, and Hsp70 increased within the cerebral cortex ($P<0.01$) in SHR rats. Additionally, nanoformulated AEA reversed

the abnormal behaviors of SHR, suggesting that this type of nanoformulation could contribute to the development of new therapies for the treatment of hypertension and behavioral disorders associated with neuroinflammation.

A84

AMARANTH GUMMIES: A NUTRITIOUS OPTION

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Amaranth was a crop and food of the native peoples of Latin America, such as the Maya, Aztecs, and Incas. It was appreciated for its high nutritional value and healing power, besides being part of religious mysticism. At present, the consumption of amaranth has been revalued due to its high nutritional value and gluten-free, FAO (Food and Agriculture Organization) and USDA (United States Department of Agriculture), declared the amaranth as the plant food with the highest nutritional value, and with enough potential to become the food of the future for humanity. Based on this, the science of food and nutrition, strive to discover, evaluate and use new food sources to help preserve health. Calories from sweets are empty calories, i.e., those that provide little or no nutritional benefit, are also of high consumption especially in children and adolescents, who constitute the age group that develops as future consumers. In this work, amaranth-seed gummies with improved nutritional value were developed. The comparison of the centesimal composition and the caloric value of gummies product marketed in gondolas and the proposed amaranth-seed gummies were made. The content of carbohydrates, total proteins, total fats, dietary fiber, ashes, and the total energy value were evaluated. The results obtained for commercial gummies were: the caloric value of 330 kcal, 81% of carbohydrates of which 51% were sugars, 1.8% protein, while for ash, fats, and fibers the value was zero. On the other hand, for the amaranth-seed gummies, the caloric value was 207 kcal, 28.4% carbohydrates (amaranth starch), 20.2% protein, 1.4% fat (high content of squalene), 1.7% of dietary fiber and 0.7% of ashes. From these results it is clear that the proposed amaranth-seed gummies could be highly beneficial since the attractiveness of this products would be combined, in terms of its consistency and flavor with the biophysiological properties of amaranth, thus promoting better feeding and allowing the inclusion in the Argentine market a novel gluten-free candy with potential health benefits, thus promoting the insertion of this pseudocereal into the diet.

A85

COMPARATIVE PROTECTIVE AND ANTIINFLAMMATORY EFFECTS OF FLAVONOIDS ON DIFFERENT ANIMAL MODELS: STRUCTURE-ACTIVITY RELATIONSHIPS

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Flavonoids are naturally content plant polyphenols found universally in all fruits, vegetables, tea, and red wine and medicinal plants, and are consumed regularly as part of the human diet. A wide variety of biological functions are attributed to these compounds including antioxidant, anti-inflammatory, antibacterial, anti-angiogenic, anti-allergic, and enzyme modulation. They have also been described as anticancer and/or chemopreventive agents. In this study, we compared the activity of eight flavonoids on different animal models like gastric ulcer induced by absolute ethanol (Method of Robert *et al.*, 1979), ulcerative colitis induced by acetic acid 10% (Method of Le Duc *et al.*, 1993) in rats, edema induced by carrageenan, acute inflammation model (Method of Sugishita *et al.*, 1981) in mice and cotton pellet granuloma, chronic inflammation model (Meier *et al.*, 1950) in rats. Flavonoids compared were the flavone 7-O-methyleriodictyol (1); flavones, 7-O-methylsudachitin (2), nevadensin (3), genkwanin (4); and flavones nepetin (5), quercetin (6), santin (7), morin (8). Adult albino Wistar rats (200–250 g) and mice (20–25 g) were used. All experiments were in compliance with the ANMAT No. 6344/96 for animal care guidelines. The protective effect of flavones (5), (3), (2) and flavanol (6) in the gastrointestinal tract was similar in both models ($P < 0.05$ vs. respective controls). These compounds showed good antiulcerogenic and gastroprotective effects and ameliorated the severity of the inflammatory lesions and reduced the damaged area in colitis. On the other hand, on acute inflammation model, compounds (6) and (8) showed significant anti-inflammatory activity at 3 h (51%, $P < 0.05$) and at 3 h (43%, $P < 0.05$) and 5 h (54%, $P < 0.05$), respectively; while, on chronic inflammation model, flavonoids (1), (2), (4), (5), (6) and (8) showed significant anti-inflammatory activity between 21 at 43% respect to control ($P < 0.01$). Several structural requirements for cytoprotective and anti-inflammatory effects include 3',4'-hydroxyl groups (B ring) observed in (1), (5), (6); 3'-hydroxyl and 4'-methoxyl groups (B ring) observed in (1). The evaluation of compounds with antioxidant properties, such as the flavonoids, may be interesting in the development of new strategies for the treatment of these gastrointestinal and inflammatory disorders.

A86

AN ALTERNATIVE COMPOUND FOR PRE-TREATMENT IN CHROMOSOMIC STUDIES IN PLANTS

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The first step in any technic applied for the study of the morphology and behavior of mitotic chromosomes in plants is the pre-treatment, the aim of which is to obtain chromosomes in metaphase, to get a good separation between chromosomes, and to obtain a higher contraction of them with a clear delineation of constrictions. The most used pre-treatments are cold, 8-hydroxyquinoline, paradichlorobenzene, and colchicine. The aim of this study was to investigate if a saturated solution of naphthalene ($C_{10}H_8$) is suitable as pre-treatment for the study of somatic chromosomes in plants. In order to analyze the effectiveness of a saturated solution of naphthalene as pre-treatment, it was compared with a saturated solution of paradichlorobenzene (PDB). The saturated solution of naphthalene (melting point = 80 °C) was obtained placing 20 g of mothballs in 500 mL distilled water, in a water bath until complete dissolution. Three treatments were included: (A) saturated solution of naphthalene, (B) saturated solution of PDB, and (C) without pre-treatment. Ten *Allium cepa* bulbs were germinated for 3 days on distilled water. Next, 10 germinated roots were randomly cut from bulbs and exposed to pretreatments for 3 h (5 roots on each pretreatment). Afterward, the usual procedures for fixing, staining, and squashing were performed, for all the roots under assay (treatments A, B, and C). An average of 2500 cells was analyzed in the root slides from each treatment, in order to calculate Mitotic Index (MI), and the percent of cells with chromosomes in metaphase. The comparison of MI and percent of cells with chromosomes in metaphase between treatments was performed by ANOVA, testing homoscedasticity by Levene test and normality by Kolmogorov-Smirnov test. The percent of cells with chromosomes in metaphase was significantly higher in *A. cepa* roots pre-treated with naphthalene (treatment A) and PDB (treatment B) compared with no pre-treated roots (treatment C) ($F_{2,11} = 16.71$; $P=0.001$). The MI was significantly lower in *A. cepa* roots pre-treated with naphthalene (treatment A) and PDB (treatment B), compared with no pre-treated roots ($F_{2,11} = 319.11$; $P<0.001$). In both pre-treatments (A and B) cells shown a strong effect on mitotic spindle disorganization, good chromosomal contraction and constriction visualization. Endomitosis and polyploidy induced cells were not observed. Cells in different phases of cellular division were detected on treatment C. The results obtained show that the saturated solution of naphthalene produced effects that are similar to those obtained with PDB, proving to be suitable for pre-treatment in cytogenetic assays in plants.

A87

QSAR STUDY FOR THE ANTIBACTERIAL ACTIVITY OF SUBSTITUTED CHALCONES AGAINST METHICILLIN RESISTANT *STAPHYLOCOCCUS AUREUS* STRAINS

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Methicillin-resistant *Staphylococcus aureus* (MRSA), identified in the early 1960s coincident with the introduction of methicillin, is now one of the most common causes of bacterial nosocomial infections, mainly in intensive care units. In this context, the chemistry of flavonoids is of particular interest since these compounds display a wide variety of biological activities, such as anti-inflammatory, antioxidant, antiallergic, hepatoprotective, antithrombotic, antiviral, and anticarcinogenic activities. Several mini and comprehensive reviews are available offering differing perspectives on the wide bioactivity spectrum displayed by these compounds. In our working group, more than two decades of research on the antimicrobial action of pure flavonoids, natural or synthetic, against strains of *S. aureus* and *Escherichia coli* have been carried out. Further, binary combinations of active chalcones with conventional antibiotics used against *S. aureus* strains have been also tested. The quantitative structure-activity relationships (QSAR) represent one of the most effective computational approaches in the drug design process. It helps to identify the molecular and physicochemical properties which govern the biological activities. In the present work, the antibacterial activities exhibited by a set of 25 substituted chalcones and derivatives on methicillin-resistant *S. aureus* strains (MN8 and NRS70) were analyzed by means of QSAR modeling. Thus, the minimum inhibitory concentrations (MICs) reported by Zhang M., *et. al.* (ACS Omega 2018, 3, 18343–18360) were modeled by means of linear discriminant analysis (LDA), and the structures of the compounds under study were characterized by means of properties calculated physicochemical and several nonempirical descriptors, such as topological, geometrical and quantum chemical indexes. Taking into account the information from the reported MICs data, the compounds were separated into three groups, namely highly active (MIC = 3.125 µg/mL), active and weakly active (MIC = >100 µg/mL) and weakly active or inactive (MIC = >200 µg/mL). The discriminant ability of the model was assessed by the correct classifications percentage, and the discriminant function quality was evaluated using the Wilks parameter (λ). The developed LDA model was highly significant statistically ($P<0.00001$) and showed a very good data-fitting ability. Thus, among the 25 observations used to fit the model, only one compound was incorrectly classified, that is, 24 (or 96.0%) were correctly classified. The predictive ability of the derived model was evaluated by using cross-validation and a rate of 84% correct classification was obtained (21 to 25). The linear discriminant functions for groups were constructed by using five molecular descriptors: $D1-3 = E3e (-2864, -2774, -2688) + TE2 (503, 488, 473) + Tu (-43, -42, -40) + SPAM (26290, 25732, 25158) + nR06 (224, 219, 212) + \text{constant} (-7113, -6814, -6514)$. From the relative magnitude of the obtained coefficients in these equations, it can be appreciated the contribution of a single variable to discriminate among three levels of activity or groups. From the analysis of the obtained results, it may be concluded that the antibacterial activities exhibited for the chalcones under the study is dependent on the molecular size as expressed by the non-directional WHIM descriptor (Tu), and especially dependent on the geometric factors and of molecular shape mainly accounted for the directional and three-dimensional WHIM descriptor (E3e) and the SPAM/ nR06 descriptors. However, these considerations do not imply that bulkiness/shape parameters are the sole factors in determining the antibacterial activity of the chalcones under study. Thus, electronic features as expressed by the TE2 topographic electronic descriptor used in the developed QSAR model must also be taken into account, and consequently, the overall activity will be determined by the detailed balance of these effects.

A88

**ANTIULCEROGENIC EFFECT FROM *JODINA RHOMBIFOLIA* LEAVES LYOPHILIZED
AQUEOUS EXTRACT ON GASTRIC MUCOSE OF MICE**

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The leaves of *Jodina rhombifolia* (Hook. & Arn.) Reissek (Santalaceae) are mentioned in some bibliographic quotes for its use as antiulcer in the Argentina and Brazil folk medicine. The aim of this study was to evaluate the antiulcerogenic effect of leaves lyophilized aqueous extract in mice. Rockland mice of either sex (25–30 g) were employed. The absolute ethanol-induced gastric lesions model was used for evaluating the antiulcer activity. The extract antiulcer effect was assayed by its oral (doses: 500 and 1000 mg/kg) and intraperitoneal (doses: 250 and 500 mg/kg) administration. At the same time was established a negative control group (saline solution). One hour later, the ulceration was induced experimentally by oral administration of absolute ethanol (0.1 mL/10 g body weight) and after one hour, the animals were euthanized by cervical dislocation. The stomachs were removed and opened along the greater curvature. The results of macroscopic inspection were confirmed by means use of the software developed by National Institutes of Health (NIH) for measuring the lesioned mucosa area. Orally pretreatment with extract at doses 500 and 1000 mg/kg produced a significant decrease in the intensity of gastric mucosal damages with an inhibition percentage of $49.33 \pm 12.81\%$ ($P < 0.05$) and $64.68 \pm 6.36\%$ ($P < 0.01$), respectively. For intraperitoneal administration, were recorded the inhibition percentages of $35.45 \pm 8.55\%$ ($P = ns$) and $54.27 \pm 11.49\%$ ($P < 0.01$), respectively for the doses of 250 and 500 mg/kg. The oral administration of the two extract doses significantly protected the gastric mucosa against the ethanol-induced lesions. This protective action could be due to a mechanical effect on the gastric mucosa, by prevention the direct aggression of ulcerogenic agent. So, we assayed the effect of its intraperitoneal administration with the same experimental model. In this case, the higher extract dose significantly decreased the lesion area promoted by ethanol, therefore suggesting that the gastroprotective action of vegetal extract not only depends on a simple mechanical action.

A89

**ANTISPASMODIC AND ANTIDIARRHOEAL ACTIVITY OF *JODINA RHOMBIFOLIA* BARK
AQUEOUS EXTRACT IN RATS AND MICE**

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Jodina rhombifolia (Hook. & Arn.) Reissek (Santalaceae), known commonly as “peje” or “sombra de toro”, is a small native perennial hemiparasitic tree that grows exclusively in South America. In several bibliographic quotes are reported on the use of bark for various gastrointestinal system affections in Argentine and Brazilian folk medicine. The aim of this work was to evaluate the effect of *J. rhombifolia* bark lyophilized aqueous extract on the intestinal function by means of *in vivo* and *ex vivo* experimental assays for determinate the antidiarrheal and antispasmodic capacity. In *ex vivo* assay was evaluated its effect on contractile concentration-response curves (CRC) induced by Carbachol and CaCl₂ in isolated rat thin intestine segments that were suspended in tissue baths to measure the antispasmodic activity. In the *in vivo* experimentation was determined the extract antidiarrheal capacity by means to use of the techniques: intestinal transit in mice, intestinal fluid accumulation in rats (enteropooling) and Castor oil-induced diarrhea in mice. The aqueous extract was administered orally and intraperitoneally at doses 125, 250, and 500 mg/kg. All data were expressed as the mean \pm SEM (standard error of the mean). A probability of $P < 0.05$ was considered significant. The bark extract inhibited the Carbachol CRC with $IC_{50} = 16.20 \pm 1.21$ mg/mL and the CaCl₂ CRC with $IC_{50} = 8.58 \pm 1.52$ mg/mL. The oral administration of the extract did not modify statistically the small intestine transit nor the severity of provoked diarrhea by castor oil, but all doses significantly reduced the intestinal fluid accumulation. Whereas, its intraperitoneal administration reduced significantly the small intestine transit (Inhibition percentage = 22.90, 40.57, and 56.41%, respectively, for the 125, 250, and 500 mg/kg doses) and severity of experimentally induced diarrhea by castor oil (Protection percentage = 73.33, 93.33, and 93.33%, respectively, for same doses). However, none three doses counteracted the hypersecretory activity of castor oil in the enteropooling technique. The results obtained in the *ex vivo* assay provide scientific data on the intestinal antispasmodic activity of bark extract. The inhibitory effect on CaCl₂ CRC suggests that the non-competitive interference with Ca²⁺ influx to the smooth muscle is the cause of the antispasmodic effect. This makes us assume that the anti-hypersecretory activity observed orally, possibly is due to the tannins present, whereas, in the effect about intestinal motility would participate in a more active manner the C-glycosilflavonoids, due to that these it have a low biodisponibility when are orally administered.

A90

RECOVERY OF THE WT1 RENAL PHENOTYPE AND ATHEROGENESIS REDUCTION: A NOVEL PROTECTIVE MECHANISM BY ORGANOSULFUR COMPOUNDS TREATMENT DURING CARDIOVASCULAR DISEASE

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Atherosclerosis is one of the leading causes of death from cardiovascular disease (CVD). Of interest, alterations in the transcription factor WT1 precede the development of CVD. The organogenic process -regulated by WT1- is modulated by oxidative/inflammatory (OI) mediators, and in this sense, it has been suggested to implement functional foods carrying organosulfur compounds (AF-OSCs) as a preventive anti-oxidative/anti-inflammatory strategy in CVD. So we decided to evaluate in a model of atherosclerosis, the possible renal changes of WT1, its correlation with OI markers, the impact on the development of atherosclerotic plaques, as well as the protection by the use of certain AF-OSCs. We used female mice deficient in apolipoprotein E (ApoE-KO) (N=8 per group) which received (A) standard feed, (B) standard food + sunflower oil, or (C) standard food + oil mashed with crushed garlic (380 ppm 2-vinylthiine, 30 ppm garlic, and 40 ppm diallyl sulfide). After two months of the food protocol, blood was taken to evaluate biochemical parameters, renal tissue for microscopy, ultrastructure, IHQ, PCR, OI markers, and thoracic aortas to quantify atheroma plaques by staining with Oil-red O. Group A evidenced a lipid, histological and functional profile consistent with the atherogenic process of the ApoE-KO hypercholesterolemic model. However, mice in group C showed a decrease in triglyceridemia after 2 months of treatment compared to group B (36.9 mg% ± 6.3 vs. 60.8 mg% ± 5.9; $P < 0.05$). No changes were observed in blood glucose and cholesterolemia. In group B, the expression of OI markers (IL-6, TNF- α , NADPH activity, and decreased nitric oxide availability) was exacerbated. Significant changes in renal WT1 expression/localization and consequent apoptosis were also evidenced. On the other hand, group C showed a reduction of OI markers with the recovery of the adult renal WT1 phenotype and lower apoptosis. The atherogenesis in the aortic arch was quantified, and it was found that garlic macerated oil reduces the formation of atherosclerotic lesions by 40%, compared to group B. We demonstrated unpublished phenotypic changes of WT1 at the renal level consistent with OI markers and cardiovascular correlate (development of atheroma plaques). The incorporation of AF-OSCs in the diet managed to reverse this phenotype, reduce OI markers, and reduce plaque size. These findings suggest WT1 as an unprecedented factor whose alteration could be critical in the natural history of atherosclerotic CVD. The confirmation of the implication of WT1 in the atherogenic process and its modulation by AF-OSCs would allow us to propose new therapeutic interventions.

NUTRITION AND HEALTH

A91

LOW BACK PAIN PREVALENCE IN CELIAC PEOPLE FROM 18 TO 60 YEARS OLD FROM THE CITY OF SAN LUIS

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Inflammatory bowel diseases (IBD) can be considered as systemic diseases since they often associate with extra-intestinal displays. Celiac disease (CD) is a chronic enteropathy arbitrated by the small intestine immune system, because of gluten exposure in the diet of genetically predisposed individuals. On its typical form, frequent on children, it shows itself as chronic diarrhea, weight loss, malnutrition, abdominal distension, and stunted growth. The most usual way in the grown-ups it's atypical, showing itself with anemia, abdominal pain, constipation, headaches, arthralgia, low size, osteopenia, among others. CD has been valued through years for its classic way of clinical presentation; however, this perspective has changed over the years, due to the CD still being able to be present even without digestive symptoms. Extraintestinal symptoms may appear before the intestinal, shaped as low back pain, sacroiliitis, hepatic disorders, etc. Our objective was to identify the low back pain prevalence in celiac people from 18 to 60 years old from the city of San Luis. An observational, descriptive, and cross-sectional research was performed through surveys of celiac people from both genders, from 18 to 60 years old, with confirmed diagnosis by serology and duodenal biopsy. Results (%): 60 adults were interviewed, female 91.67, male 8.33. The average age was 34,63 years old (SD 11.7) and the average age of diagnosis was 26 years old (SD 12,8); adherence to a gluten-free diet 80. Prevalent symptoms before and after diagnosis and both respectively: abdominal distention 53.3, 5 and 20; diarrhea 48.3, 3,3 and 6,7; fatigue 45, 1,7 and 18,3; abdominal pain 38,3, 1,7 and 20; hair loss 38,3, 5 and 16,7; weight loss 36,7, 3,3 and 3,3; flatulence 31,7, 5 and 15; vomit 31,7, 3,3 and 6,7; constipation 28,3, 5 and 8,3; sickness 23,3, 1,7 and 5; back pain 20, 20 and 43,3. Back pain area: low (low back pain) 39,62, upper and lower 24,63. All areas 11,32, half and lower 7,54. Pain intensity: moderate and intense 54,71, severe and unbearable 24,52. Abdominal inflammation and low back pain 75,4 ($P < 0,05$). Conclusion: prevalent gender was female; the diagnosis was performed mostly in early adulthood and adhesion to a gluten-free diet was high. The disease showed itself with intestinal symptoms such as diarrhea, vomiting, constipation, distension, and abdominal pain, and extraintestinal such as fatigue, hair loss, weight loss, and back pain. All symptoms, except back pain, decreased after a gluten-free diet. The back area with more pain was lumbar, and the connection between the abdominal

inflammation and the low back pain was meaningful. It's important to recognize early the low back pain associated with celiac disease, in order to prevent it from becoming a chronic process with substantial social repercussion, occupational and economical that adds up to the base disease.

A92

SHELF-LIFE TIME DETERMINATION OF ENZYMATIC EXTRACTS OBTAINED FROM GLABROUS CANARY SEEDS (CDC MARIA VARIETY)

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Hairless (glabrous) canary seed is a novel food and promising source of starch, protein, and oil that recently received food approval from Health Canada and GRAS status from US - FDA. Previous studies demonstrated that Glabrous canary seed (CDC-Maria variety) have a unique protein composition of high catalytic activity that can be used for the formulation of different food products. For this reason, its extraction and subsequent enzymatic fractionation were performed. The enzymatic extraction process was carried out with a specific protein phosphate buffer solution and subsequent centrifugation. The supernatant was called crude extract (EC). Subsequently, a fractionation using an isoelectric point precipitation process between pH 4.5 and pH 5.5 with citric acid was performed. Finally, the enzymatic fractions were preserved by lyophilization. The effect of the addition of the EC and the enzymatic fractions obtained as a food additive in gluten-free bread were studied. The EC and enzymatic fractions are innocuous from human consumption. This study allowed select as optimum sample the enzymatic fraction obtained at pH 5.3. Maillard reactions have a great influence on the quality degradation of enzymatic products. This process includes a series of reactions that generate the formation of the 5-hydroxymethylfurfural intermediate (HMF), a compound with activity in the UV spectrum-visible. In this work, the stability of the EC and the enzymatic fraction obtained at pH 5.3 were studied. For that, a determination of the deterioration kinetics was made from a spectrophotometric measurement of HMF at 280 nm using the Carrez I (potassium ferrocyanide at 15%) and Carrez II (30% zinc acetate) reagents. Subsequently, the shelf life was calculated at a temperature of 8 °C. The samples were stored in flexible Pet/Al/Pe containers at different temperatures: EC at 20, 30, and 40 °C and the fraction obtained at pH 5.3 at 20 °C and 30 °C for 50 days, and at 7 °C per one year. The results showed that all samples presented consistent multivariate kinetics of zero order. In the first instance, the production of the measuring compound (HMF) is generated from various chemical reactions. Subsequently, it is consumed when reacting with an amine forming undesirable browns pigments. The reaction rate constant of the HMF production no presented statistically significant difference between the studied samples (EC: 0.0057/day; pH 5.3: 0.0094/day). This may be due, that the lower pH of the sample obtained at pH 5.3 decreases the reactivity of the amino groups and increases the reactivity of the carbonyl groups of the reducing carbohydrates, increasing the HMF formation. However, the rate constants obtained for the HMF consumption reaction presented a statistical difference between studied samples. A lower value of the reaction rate of the fraction obtained at pH 5.3 was obtained (EC: 0.231/day; pH 5.3: 0.034/day). This may be due to its lower pH value (isoelectric point pH) that decreases the reactivity of the amino groups by a reduction of the net protein's surface charge. This demonstrated the strong influence of pH in the last phases of the reaction. Therefore, the freeze-dried fraction obtained at pH 5.3 at a storage temperature of 8 °C presented a higher shelf life (EC: 21 months and pH 5.3: 25.4 months).

A93

RISK FOR CARDIOVASCULAR DISEASE IN FEMALE PHYSICAL THERAPIST PATIENTS

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World Health Organization (WHO, 2000) says the measurement of waist circumference is an indirect indicator used for identification of risk for cardiovascular disease. Actually, is used by different associations to diagnostic, which limits are more than 88 cm for female and more than 102 cm for male. According to the cutting points, in 1997 were proposed three categories to identify risk in women: lower risk ≤ 79 cm, moderate risk 80 to 87 cm, and high risk ≥ 88 cm, but it can be different according to ethnics. The objective of the present study was to describe the characteristics of women who went to a center of physical therapist, between 2010 and 2019, according to the risk level of suffering cardiovascular diseases. For that, waist circumference was taken with a meter in 281 women in the first date. Also were taken the age, weight, and height to calculate Body Mass Index (BMI). The results suggest that the average age was 39.3 ± 11.1 years; average IBM was 24.6 ± 3.3 . About the population waist circumference, 19.2% was lower risk, 59.4% was moderate risk and 21.4% was high risk. The waist circumference categories depend on BMI ($P=0.000 < 0.05$). We conclude that most women had moderate risk for cardiovascular disease, and their average IBM was in the limit between normal and overweight. The risk for noncommunicable diseases increases, with increases in BMI, so is very important from physical therapist prevent them through the practice of regular physical activity and dermato-functional kinesiology.

A94

DERMATO FUNCTIONAL KINESIOLOGY TO REDUCE THE WAIST CIRCUMFERENCE

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An increased number of people are consulting for non-invasive aesthetic treatments that reduce abdominal obesity with the purpose of treating unaesthetic treatments and prevent non-transmissible chronic diseases associated (Type II diabetes mellitus, high blood pressure, vascular disorders) and others such as arthrosis, low back pain, sleep apnea, etc. Local researchers conducted a nutritional study in adults of downtown San Luis in 2016–2017, which determined that 52% of the population was overweight, and 22% had obesity, without significant differences between sexes. Dermato functional kinesiology, which consists of different reducing treatments, could be a safe and effective alternative in reducing abdominal circumference in order to prevent cardiovascular and other risks. The purpose of this research is to determine the effects of the application of this kinesics in the abdominal area. We selected 281 women who assisted to dermato-functional kinesiology center, during 2010–2019. The average age was 39 ± 11.17 years. After an initial assessment with a tape measure, different kinesics reducing treatments were applied. Conventional treatment was applied to the first group for ten sessions (a combination of ultracavitator, massage therapy, endermology, and thermotherapy). Cryolipolysis was applied only once to the second group, while 6 sessions of High-Intensity Multi Focalized Ultrasound or monopolar Cryoradio frequency were applied to the third group of patients. After concluded their treatments, the mid-waist perimeter of all women was evaluated again to compare. A statistically significant perimeter decrease (P -value: 0.000, 0.000, and 0.016 < 0.05 , respectively) was obtained in each of the groups analyzed. Although all the treatments were statistically significant, it would be necessary to analyze the cost-benefit relationship in each one, because if with the cryolipolysis the same results are obtained only once, it prevents the patient from having to attend treatment several times. It is concluded that dermato-functional kinesiology offers an effective and safe alternative to reduce the circumference of the waist and thus promote the prevention of diseases associated with android obesity.

A95

LIFESTYLE AND COGNITIVE RESERVE IN OLDER ADULTS FROM THE PROVINCE OF SAN LUIS

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An active and healthy aging process is affected by a person's lifestyle, which is related to aspects that are evident in attitudes, values, and behaviors. Disease, problems, or ailments that manifest in old age are originated in the lifestyle that we have previously adopted and they depend on whether it has included protective factors that maintain health and promote longevity or risk behaviors that are harmful and reduce life expectancy. Cognitive Reserve (CR) is a protective factor of cognitive aging and neurodegenerative disease associated with age. Several factors are involved in its formation: innate ones, like brain size and synaptic density; and acquired ones, which depend on the environment, such as educational level, occupation, knowledge of different languages, playing intellectual games, etc. This topic is especially important since human beings' average lifespan is currently longer. The objective of this study was to know the relationship between lifestyle and cognitive reserve of older adults, of both genders, who attend senior centers in the province of San Luis. The sample was intentional, non-randomized, and it was made up of 65 older adults (38 women and 27 men) aged between 66 and 79 years. The Fantastic questionnaire by Ramirez V dez and Agredo was applied to the sample, as it allowed us to identify and evaluate participants' lifestyles through a multidimensional method (evaluation of physical, psychological and social factors). Similarly, the Cognitive Reserve Questionnaire by Rami, Valls-Pedrest, Batr s-Faz and Molinuevo was used to assess individuals' schooling level, training courses, occupation throughout life and language skills, among others. The results indicate that 24% of the participants lead an excellent lifestyle, 21% a good one, 19% a regular one, and 1% a bad one. We conclude that 69% of the sample lead a healthy lifestyle, which includes: regular physical activity, low consumption of alcohol, tobacco, medications and stimulant drinks, a diet low in processed foods, fats and sugar, healthy rest habits, accident prevention behaviors, emotional well-being related to self-esteem, healthy interpersonal relationships, stress and anxiety. Most individuals' CR (78%) is within the lower range (0–7 points), 5% in a medium-range (7–9 points), 11% in a medium-high range (10–14 points), and 6% in a higher CR category. Considering the data obtained, we can conclude that only 22% of the studied older adults have protective resources against possible brain aging. The deficits could be related to low schooling levels, work activity, low bilingualism, few training courses, and little musical and foreign language training. Regarding the relationship between the variables, a positive correlation in the total sample was found: the healthier the lifestyle, the higher the CR.

A96

USE OF SOURDOUGHS FOR THE ELABORATION OF HEALTHY COOKIES

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Topinambur (*Helianthus tuberosus*) is a tuber with high inulin content, a prebiotic polysaccharide used as an ingredient in functional food. The aim of this paper was to prepare healthy cookies with low caloric and high fiber content. Cookies were prepared from two sourdoughs with different compositions in order to study their characteristics and behavior on the final product. Two Type I sourdoughs were prepared, with a dough yield (dough weight/flour weight x 100) of 200. The fermentation procedure was performed for 5 days, maintaining the temperature between 25 and 30  C. In both cases, the initial fermentation was carried out with Whole

Wheat Flour (Pureza Integral, Molino Cañuelas S.A.C.I.F.I.A., Buenos Aires, Argentina). Both sourdoughs differed in their flour composition, using either 100% Whole Wheat Flour (100WWF) or combining in equal proportions m/m, Whole Wheat Flour and Wheat Flour 000 (Morixe, Prepared by Province, Argentina) (50WWF). Topinambur flour (TF) used for elaborating cookies was obtained from our own crops (on the FICA-UNSL site) and was characterized using official methods of analysis (AOAC, 1990). For cookie preparation, the following ingredients were mixed: sourdough 25 g, TF 25 g, salt 0.5 g, (Celusal, I.Q. y M. Timbo S.A., Buenos Aires, Argentina), tap water 3 g, extra virgin olive oil 4 g (Natura Extra Virgen clásico, Aceitera General Deheza S.A., Córdoba, Argentina) and thyme 0.5 g (Alicante, Caf   La Virginia S.A., Santa Fe, Argentina). The obtained dough was then sheeted with a roller kneader until 2 mm height and cut into circular pieces of 2.7 cm diameter. The resulting cookies were placed on an oven tray and baked in an electric oven ATMA (HG5010AE, China) at 150   C for 7 min. Finally, the cookies were allowed to rest for 30 min before storing them into airtight polyethylene bags. In order to observe the possible differences between cookies according to their sourdough, texture tests (Texturometer Brookfield Textire Pro CT), color tests (Chroma meter CR300), and humidity and ashes determinations (AOAC, 1990) were performed. There were no significant differences ($P>0.05$) between the resulting cookies regarding deformation percentage, fracturability, chewiness (textural properties), color, humidity, and ashes. It can be concluded that the flour type used in sourdoughs did not affect these parameters on cookies. On this basis, it is possible to increase cookies benefits by using 100% Whole Wheat Flour sourdough (100WWF), which provides a higher amount of fiber than Wheat Flour sourdough (50WWF) without altering cookies properties.

A97

ROTATOR CUFF INJURY: COMPARATIVE STUDY OF THE EFFECTIVENESS BETWEEN TREATMENT WITH MANUAL THERAPY AND TREATMENT WITH PHYSIOTHERAPY ASSOCIATED WITH STRETCHING EXERCISES

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The objective of this study was to identify the joint mobility degree differences, the scale of Shoulder Pain and Disability Index (SPADI) after applying a treatment with manual therapy in relation to another with physiotherapy and stretch exercise, in patients between 25 to 60 years with rotator cuff injury. They were divided into two random groups, group A is treated with manual therapy and group B it was applied physiotherapy associated with stretching exercises. In the first session, the entire population made the kinesics card, the evaluation of the affected shoulder through the SPADI scale, and the measurement of mobility degrees through a goniometer. Once treatment finished, the affected shoulder was evaluated with the same instruments. The data were analyzed through the use of Microsoft Excel and SPSS program version 22. The treatment produced statistically significant changes in the reduction of pain ($P<0.05$), in the reduction of the level of difficulty in performing daily living activities ($P<0.05$) and in joint mobility degrees increase, in abduction, adduction, external rotation and internal rotation ($P<0.05$). Treatment B achieve also significative statistically changes in the reduction of pain ($P<0.05$), in the reduction of difficulty level in performing daily living activities ($P<0.05$) and in increase joint mobility degrees in abduction, adduction, external rotation and internal rotation ($P<0.05$). In conclusion, both treatments produced changes statistically satisfactory in the reduction of pain and the level of difficulty in performing the daily life activities, as well as in the increase of the joint mobility degrees. There were no statistically significant differences between both treatments; however, treatment A is more recommendable, since it is not necessary to acquire physiotherapy equipment which is expensive and also time extend is minor, approximately 30 min with respect to treatment B that takes approximately 60 min.

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EVALUATION OF ANTIFUNGAL ACTIVITY OF NANOFORMULATES FROM VEGETABLE EXTRACTS AGAINST *FUSARIUM GRAMINEARUM*

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In recent years, the search for natural sources of compounds with antifungal and repellent activity has acquired great importance since they are today a promising alternative to counteract the negative effect of some phytopathogenic microorganisms, due to their low cost, and because they are friendly to the environment and health in general. Products of plant origin have been mostly studied in their chemical composition, with emphasis on secondary metabolites, which are involved in biological control against pathogens or pests. The objective of this work was to evaluate the antifungal activity of silver nanoparticles synthesized from plant extracts, such as rosemary (*Rosmarinus officinalis*), "pichana" (*Baccharis spartioides*), and false sunflower (*Heliantus petiolaris*), using ascorbic or citric acid as a control. Silver nanoparticles (AgNPs) were synthesized from silver nitrate and ammonium hydroxide at 80   C. Subsequently, they were characterized by UV-visible spectroscopy and transmission electron microscopy (TEM). The evaluation of the fungicidal activity was carried out by measuring the halo of the fungal growth with respect to the negative control. Potato dextrose agar (PDA) was used, which was added 50% of the different concentrations of the extract. For sowing the plates, 3 mm diameter slices of phytopathogenic fungus colonies were used, which were placed on the center of Petri dishes, which were then sealed and incubated at 26   C. The results obtained were evaluated considering as effective those extracts that showed a percentage of growth less than or equal to 80% and a percentage of inhibition greater than or equal to 20%. The growth values found for the control and the species of rosemary, pichana, false sunflower, were 20%, 95%, 70%, and 75%, respectively. Regarding inhibition, the values

found for the plant species of rosemary, pichana, false sunflower and control were (without inhibition) 30%, 30%, and 70%, respectively. From the analysis of the results, it is concluded that the AgNPs of the false sunflower and pichana species are potential agents against the fungus *Fusarium graminearum*.

A99

EFFECT OF LYOPHILIZATION PROCESS ON THE PROPERTIES OF THE ENZYMATIC FRACTIONS OBTAINED FROM GLABROUS CANARY SEED (CDC MARÍA)

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Enzymatic technology is of primordial interest for food biotechnology, because, among other reasons, their properties as quality improvers of products like gluten-free bread. Until now, a product that can mimic gluten properties has not been found. Therefore, in this work, the extraction and fractionation of enzymes from Glabrous canary seed (CDC-María variety) were performed. The enzymatic extracts were used to replace gluten. The enzymatic extraction process was carried out with a specific protein buffer solution, followed by fractionation using an isoelectric point precipitation process. For this, the solution pH was adjusted at 4.5, 5, and 5.5. Subsequently, lyophilization was used as a preservation method. The effects of the drying process on the effectivity of enzymatic fraction (EF) as an improver of gluten-free bread was evaluated. Lyophilized EF was added at concentrations between 0.5 to 1% (w/w). The results were compared with a control sample without EF addition. The volume increase percentage and aeration percentage of the baked product were calculated from the images taken from nine slices of bread for each formula. Texture profile was studied with a TMS-TOUCH (Food Technology Corporation, DASTEC) texture analyzer during a period of three days. Shelf life and hardening speed of bread were calculated. The addition of EF obtained at pH 4.5, pH 5 and pH 5.5 without lyophilization generated a significant statistically decrease in the firmness and hardening speed with respect to the control sample. The highest reduction of these parameters was obtained for the sample with the addition of the EF obtained at pH 5.5 (4.65 ± 0.32 N/day) with respect to the control sample (8.34 ± 0.48 N/day). The addition of EF at pH 5.5 showed the highest values of volume increase and aeration with respect to the control sample (33.88% and 21.2%, respectively). Therefore, the EF obtained at pH 5.5 was selected as the optimal formula. The addition of this lyophilized fraction at a concentration of 0.75% (w/w) allowed to obtain the lowest firmness value and the highest volume increase and aeration with respect to the control sample. However, from the second day of storage, a progressive decrease in the firmness was observed. This may be due to a loss of integrity of the matrix. Besides, the addition of this EF generated a statistically significant increase in the hardening speed with respect to the control sample (19.62 ± 0.98 N/day). The behavior of the samples with EF with lyophilization or without it presented important differences. This may be due to the lyophilization process that probably caused the denaturation or loss of activity of some enzymes of the EF, generating this negative effect on the sample. Therefore, the use of the EF obtained at pH 5.5 without lyophilization, generated significant improvements in the final characteristics of the gluten-free bread. In subsequent studies, the effects of different lyoprotectant agents will be investigated. This will allow obtaining adequate preservation of its enzymatic activity.

A100

ENZYME EXTRACTION FROM GLABROUS CANARY SEED (CDC MARIA) AND ITS POTENTIAL USE AS FOOD ADDITIVE IN A GLUTEN FREE PRODUCT

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The development of gluten-free products represents a challenge for the food industry since the lack of gluten impact negatively on its structural and sensory qualities. The bakery industry uses different additives, like enzymes, for mimicking gluten properties. In previous studies was demonstrated that Glabrous canary seed (CDC María) has a high protein content with catalytic activity and lacks gluten proteins. Therefore, we performed its extraction, characterization, and then its application as a food additive. For that, a crude extract (EC) was obtained using a specific buffer solution for proteins. The EC was conserved through a lyophilization process and then was used in the elaboration of gluten-free bread. The influence of lyoprotectant agents, addition (inulin and maltodextrin) to the EC and its influence on the baked product's final properties were studied. Lyophilized EC was added in different concentrations (w/w): 0.25%, 0.5% and 0.75%. The results were compared against a control sample without the addition of enzymes. The lamella thickness was determined from the analysis of images captured using an electron microscope. Texture profile was studied with a TMS-TOUCH (Food Technology Corporation, DASTEC) texture analyzer during a period of three days. Shelf life and hardening speed of bread were calculated. On the first day of storage, the control sample showed a firmness of 30.2 N and a hardening speed of 8.34 ± 0.48 N/day. The addition of lyophilized EC without the addition of lyoprotectant agents, at the different concentrations studied, allowed statistically reduce the hardening speed and the firmness of the samples throughout the storage period compared to the control ($P < 0.001$). The sample with the incorporation of lyophilized EC at 1% (w/w) presented the lowest hardening speed (5.72 ± 0.27 N/day). The formula with the addition of lyophilized EC at 0.75% (w/w) allowed the greatest decrease in firmness on the first day of storage (12.1 N) with respect to the other tested concentrations. The addition of the lyophilized EC with lyoprotectant agents, maltodextrin and inulin, allowed to decrease the firmness and lamella thickness with respect to the control sample. However, these samples showed an increase in the parameters of firmness and lamella thickness in comparison to the samples with the aggregate of

lyophilized EC without lyoprotectant agents. This may be due to the higher hardening speed values founded for the samples formulated using EC with lyoprotectant agents. Therefore, the use of lyophilized EC obtained from Glabrous canary seed without the use of protective agents allowed to improve the properties of gluten-free breads. This procedure improved their shelf life time and reduced its firmness throughout the studied storage period.

A101
ASSESSMENT OF THE ANTIOXIDANT CAPACITY OF FRUITS OF
SCHINUS MOLLE (FALSE PEPPER)

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Around the world, medicinal plants are used in diverse cultures to treat and prevent many diseases that affect humans. Our country is characterized by diversification of these plants, which were used by our ancestors to cure diseases, through essences, plant patches, and massages, among others; these were complemented with rituals, prayers, and sacred supplications to the gods of nature. This is the case of the plant species *Schinus molle* L., recognized for its antibacterial, antimicrobial, and other healing properties. Its medicinal properties are located mainly in leaves and fruits of the "molle". Leaves have an essential oil of a bitter principle, known as gomoresin oxidase. The objective of the present work was to evaluate the antioxidant capacity of the fruits of *S. molle*. To do this, phytochemical screening was carried out by means of test-tube reactions and the determination of the antioxidant capacity through a quantitative test with the DPPH method, where absorbance was read at 517 nm. Total phenolic content was determined by the Folin-Ciocalteu method; the absorbance at 760 nm was measured. The fruits were collected and then extracts were made by maceration in water and a 1/1 water/ethyl alcohol mixture, both at room temperature. Phytochemical screening allowed identifying the presence of flavonoids, tannins, phenols, reducing sugars, saponins, triterpenes, and steroids. The anti-radical activity, expressed as a percentage of free-radical trapping capacity (% FRTC) of the extracts of *S. molle*, was 86.12% in water and 94.20% in water/ethyl alcohol. These values were deemed acceptable. The polyphenol content in the fruits was 0.689 mg of gallic acid/L in water and 0.420 mg of gallic acid/L in water/ethyl alcohol. From these results, it is concluded that the fruits of *S. molle* have high antioxidant capacity values, with a direct correlation between the total phenolic values and the radical trapping capacity values. The antioxidant activity found in the studied species suggests its application in the food industry as a possible substitute for synthetic antioxidants.

A102
POSTURAL CAPTORS IN UNIVERSITY STUDENTS

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Posturology studies balance between the postural system of the human body and how it aligns itself in relationship to the surrounding environment, during static and dynamic alignment as it stabilizes itself in space against gravity and other forces, and any treatments. Novo (2008), Barrata (2006), Fernandez Pinto (2016), have said there is a relationship between postural captors, which are in the muscles and ligaments of the feet, joints of the jaw, spine, eyes and inner ear, and the people posture. The aim of this study is to describe the prevalence of dysfunctional postural captors in University students of physical-therapist from San Luis. 39 students, who gave informed consent, were evaluated during 2018. Front, back and both sides were observed. The dysfunctional captors are determinate from Chaitow postural scales, proposed in 2006. The population was 19 to 28 years old, but the average age was 21.3 ± 1.95 . 59% were female and 41% were male. The first reason for consultation was low back pain (30.8%), then thoracic and neck pain 17.9%. The 33.3% had another pain different from vertebral spine. Downloaded postural alterations (jaw and eye) were 59%, but most common were jaw (38%), in men. Eyes captors were dysfunctional about 21%, most in men. On the front side, upward postural alterations were in 41% of the population, 24.6% in the foot, and 16.4% in sacral. Foot types were most common in men (16.8%) and sacral type in women (13.6 %). The 23.1% had two or more dysfunctional captors, but the most had jaw-eye (12.8%). These results were similar to those of Loroño (2012) and Villoria (2003) about low back pain, and it shows the influences from the captor's jaw-eye and trigeminal nerve. For future investigations, we suggest increasing the population and to make the relationship between dysfunctional captors and myofascial chain. We conclude that download postural alterations are more common, the first dysfunctional captors is jaw-eye, then only eye, most in male; but in women, there are more upward postural alterations of sacral type.

A103
HYPOTHYROIDISM AND NECK DISORDERS

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Hypothyroidism is a condition in which the thyroid gland is not able to produce enough thyroid hormone (TSH). Most of the time, it happens when there is a dysfunctional gland, but in endemic zones, it could be produced by lower levels of iodine at the environment. On the other hand, alterations in the alignment of the head and neck are one of the most common postural alterations which could explain the high prevalence of neck pain and Myofascial pain syndrome overpopulation. Yap (2007) and Loreto D áz (2014) state that one of precipitating and perpetuating factors for that are endocrine and metabolic deficiencies like TSH insufficiencies. The aim of this study is to observe the relationship between neck disorders in patients with medical diagnoses of hypothyroidism. It was an observational, descriptive of a transversal study. In 2018, twelve women were evaluated in a physical therapy center from Rio Cuarto City, with neck pain and medical diagnostic of hypothyroidism. They were interrogated and physically examined afterward. Laboratory, neck radiographs, and gland ultrasound were also included. The average age was 30 ± 11.5 , ranging from 15 to 51 years. The media level of TSH was 4.31 ± 3.14 , although the highest was 13.28. The media level of T4 was 1.07 ± 0.13 , all normal, and about T3 there is a lot of lost data. 75% of patients had normal levels of TSH, while in the rest it was highest. Nevertheless, when ultrasound images of the gland were analyzed, most of them had normal images, and those patients who had nodules, had also normal levels of TSH. 83.3 % of patients had negative thyroid antibodies, while the rest were positive. From ten radiographs, 80% were decreased neck curvature, 10% increased and 10% normal. In the neck mobility test, only four had dysfunctional vertebrae, which only one time for each one (C1, C3, C5, and C6). Nevertheless, 9 types of dysfunctional were found, with 78% in flexion, 11% extension, and 11% neutral position. From 9 ultrasound images, 66.7% had normal gland and 33.3% had nodules. From twelve patients, 91.7% showed at least one associated symptom (all neck pain) and only 8.3% had three other symptoms. None of them had a goiter, but 58.3% had decreased mobility of the gland and 100% had tension fascial in neck zone. Even in this case, all patients Hypothyroidism had any disorder on their necks. We could not conclude that endocrine and deficiencies produce cervical dysfunction. We suggest studying more people and the blood circulation of the gland to complete conclusions. Maybe the high prevalence or dysfunctional cervical could explain that muscle and fascial retraction can affect the blood circulation of the gland and disturb its function.

A104

PREVALENCE OF RESPIRATORY DISEASES IN PEDIATRICS PATIENTS WHICH REQUIRE OXYGENATION BY HIGH-FLOW NASAL CANNULA

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The use of other than conventional oxygen therapy systems is required to face the challenge of respiratory disorders in children with moderate hypoxemia. A high-flow nasal cannula has several advantages, such an inspiratory flow exceeding the patient's demand, gas heating and humidification and a higher inspired oxygen fraction (FiO_2), providing an alternative treatment to avoid the worsening of the underlying condition. The prevalence of disorders that required the use of high-flow cannula in children from 1 month to 14 years of age who were admitted to the pediatric clinic or pediatric intensive care unit at Hospital San Luis from September 1, 2018, to August 31, 2019, is reported. First and last name, age, gender, medical diagnosis, day and time of Admission to high-flow cannula, Clinical Evaluation Scale as appropriate: Wood Downes Score or modified Tal Score were recorded for all children. A total of 511 patients were derived from respiratory kinesiology; 27 of them required a high-flow cannula, most (70%) with bronchiolitis (BL), 11% with community-acquired pneumonia (CAP), 11% for brief resolved and unexplained events (BRUE), 4% for infant botulism. The remaining 4 % were recurrent wheezing infants (RWI). Most children (81%) started with a high-flow cannula in the pediatric clinic, while 19% did at pediatric intensive care. 37.03% of the total of these children required higher escalation to a ventilation system, while only 31.8% of those with a bronchiolitis diagnosis required escalating to a ventilation system. The average age with a BL diagnosis was 3.7 months, with CAP of 9 months, and with BRUE of 2.7 months. The use of high-flow cannula according to the diagnosis required an average of 3.1 days for BL, 4.3 days for NAC, and 1.3 days for BRUE. As a conclusion, in most children requiring high-flow cannula the shortness of breath subsided within days, with an improvement of the clinical scores, leading to the decrease of supplemental oxygenation and flow until they were discharged home. Prevalence of the need for high-flow cannula was observed in predominantly obstructive disorders, such as BL and RWI, with predominance in children under 6 months of age. We consider it necessary to continue the evaluation of patients selected for the high-flow nasal cannula, as well as knowing the existence of continuous positive pressure on the airway related to the flow applied during the use of a high-flow cannula. In this way, we would try to define the indications to respiratory conditions that require the application of high-flow nasal cannula.

VETERINARY, ANATOMY, HISTOLOGY AND ANIMAL FISIOLGY

A105

PCNA EXPRESSION IN THE PINEAL GLAND OF FOETUS AND SEXUAL IMMATURE VISCACHAS (*LAGOSTOMUS MAXIMUS MAXIMUS*): AN IMMUNOHISTOCHEMICAL AND MORPHOMETRIC STUDY

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The non-histone nuclear protein PCNA (proliferating cell nuclear antigen) plays a key role in the initiation of cell proliferation. It works as a cofactor of DNA polymerase-delta and its use as a marker of proliferating cells has been validated by several studies in different species. The pineal gland acts as a neuroendocrine interface, transforming ambient light changes into hormonal variations. Our experimental model, the viscacha (*Lagostomus maximus maximus*), is a hystricomorph rodent with a seasonal reproductive pattern synchronized by the environmental photoperiod and modulated by the pineal melatonin. In this study, we performed an immunohistochemical and morphometrical study to evaluate PCNA expression in the pineal gland of fetuses and sexual immature viscachas. Four pineal glands (N=4) per group were analyzed. The fetal glands were collected from fetuses of late pregnant females classified on the basis of fetal weight and crown-heel length. The sexual immature animals (2–3 kg) were classified according to body weight and light microscopy observations of testes. The glands were processed for light microscopy and the PCNA was identified immunohistochemically. The percentage of immunoreactive cells was determined using the formula $(A/A + B) \times 100$, where A is the number of immunoreactive cells and B is the number of nuclei in immunonegative cells. Statistical differences were evaluated by means of the Mann–Whitney's test. A value of $P < 0.05$ was considered significant. Immunohistochemical results revealed that PCNA-immunoreactivity (PCNA-ir) was detected mainly in the nuclei. Positive PCNA nuclei were oval or spherical shaped, according to the pinealocytes. The percentage of PCNA-ir cells was higher in the fetal glands (1.72 ± 0.07) compared to the sexual immature ones (1.29 ± 0.12). In the fetal pineal, a higher cell proliferation was observed, which indicates that this moment of pregnancy is fundamental for the gland development. Increased serum levels of estradiol and progesterone during pregnancy might be related to the increased number of PCNA-ir cells. In the post-natal life, the percentage of PCNA-ir cells decreased, according to the low levels of melatonin and androgens reported during the sexual maturation. Further studies are needed to confirm this relationship.

A106

EXOGENOUS MELATONIN EFFECT ON THE CELLULAR PROLIFERATION OF THE VISCACHA EPIDIDYMAL CORPUS

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In several studies has been observed that melatonin could act such as a stimulant or inhibitor of the cellular proliferation in the male reproductive system according to the established conditions. The proliferating cell nuclear antigen (PCNA) plays an essential role in nucleic acid metabolism as a component of the mechanism of replication and repair in mammals and has been used as a marker of proliferating cells in several studies. The viscacha (*Lagostomus maximus maximus*) is a wild South American rodent with nocturnal habits and seasonal reproduction. The objective of the present work was to study the effect of melatonin administration on the proliferative activity of the epididymal corpus through the analysis of PCNA expression by immunohistochemistry (antibody AM 252-5M Biogenex). The adult animals were captured during the period of maximal gonadal activity in their habitat near San Luis city. The animals were divided into two groups: (1) the experimental group (N=4) received two daily subcutaneous injections of melatonin (Sigma, 100 µg/kg body weight in aqueous solution) at 09:00 h and 17:00 h for 9 weeks, and (2) the control group (N=4) received only the diluent. The blood samples were obtained from anesthetized animals. The epididymal samples were surgically removed and processed for optical microscopy. The serum testosterone levels were determined by a solid-phase competitive chemiluminescent enzyme immunoassay. The serum testosterone levels were significantly lower in melatonin-administrated animals (159 ± 36.43) than control animals (487.50 ± 45.35). The PCNA expression was observed principally in epithelial cells being scarce in stromal cells. The lower percentages of PCNA-ir epithelial cells were observed in melatonin-administrated animals (17.09 ± 1.23) in relation to that observed in the control group (81.71 ± 1.22). Our results showed that the PCNA expression in epithelial cells was lower in melatonin administrated animals, suggesting that the melatonin could be a direct or indirect regulator of cell proliferation. In addition, the melatonin administrated animals showed lower serum testosterone levels, suggesting that both hormones could participate in the hormonal regulation of the cellular proliferation in seasonal species such as the viscacha.

A107

POTENTIAL BIOLOGICAL CONTROL OF THE MITE *VARROA DESTRUCTOR* BY THE ENTOMOPATHOGENIC FUNGUS

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Apis mellifera plays a very important role in beekeeping and in the pollination of crops that man consumes daily. Several potential factors have been studied as possible causes of declining pollinator health, such as parasites and pathogens, exposure to agricultural pesticides, habitat loss, and/or climate change. Within the group of biotic stressors is *Varroa destructor*, a mite that causes a parasitosis called Varroosis; whose main effects are physiological and physical damage when feeding on the hemolymph of bees. Various synthetic miticides have been developed to target *V. destructor* mites. However, the long-term application of miticides has resulted in the development of resistance in *V. destructor*. Biological control is an alternative strategy, based on the total or partial destruction of the pest through the use of its natural enemies; when it comes to fungi capable of causing disease in insects or mites they are called entomopathogens. In this work, the pathogenicity and susceptibility of an isolate native to the province of San Luis and a commercial strain of *Beauveria bassiana* were evaluated by bioassays against *V. destructor* and *A. mellifera*. The native fungal isolates were obtained from soil samples from different locations in the Province of San Luis, isolated in Chasé medium and characterized by gender from microcultures according to morphological characteristics and the taxonomic key proposed by Barnett and Hunter (1998). For the bioassays performed, mites and bees were collected. They were individually embedded for 10 s in 5 mL of the conidia suspension for each treatment (1×10^8 conidia/mL) and 5 mL of sterile Tween 80 for control (0.03% w/w). Each group of 5 mites and bees was placed in a sterile Petri dish with 9 cm diameter with food and water *ad libitum*. Five repetitions were performed for each treatment. The boxes were placed in an incubator and kept in the dark at 30 °C and 100% RH (humidity obtained by placing damp cotton on the base of the plate). The mortality of mites and bees was recorded at 24, 48, and 72 h. Dead mites or bees (which showed no movement or stimulus-response) were removed and incubated in 1% water agar at 23 °C. The presence of mycelium on the body of the mite or bees was used as an indicator of fungal-induced mortality. The strains studied were infective producing mortality of 20% for the native strain and 40% for the commercial strain. Although the native strain was less infective, it is a good indication considering its adaptation to the environment where it would be applied. As for the bioassays in bees, the growth of mycelium on the surface was not observed, which suggests the absence of pathogenicity of the strains tested in bees.

A108

HEMATOLOGICAL PARAMETERS IN THREE SPECIES OF *LIOLAEMUS* IN DIFFERENT ALTITUDES OF SAN JUAN, ARGENTINA

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Hematology provides a significant number of data on the different elements of blood, including morphology and the absolute and relative number of different cell lines. In reptiles, as in the rest of vertebrates, the cells present in the circulating blood are grouped into: erythrocytes, leukocytes and thrombocytes, in turn, the values of these cells can present structural and functional adjustments according to the altitude of their habitat, such as modifications in the respiratory surface, of hemoglobin, hematocrit, capillary density, myoglobin levels, among others. In reptiles, there may be little or no correlation between the blood values of lizards and their altitudinal distribution, while some authors have reported high hematological values in lizards of high altitudes. The objective of this work was to present preliminary results on some hematological parameters in specimens of the genus *Liolaemus*. The studies were performed on specimens of three species: *L. darwini*, captured in the area of La Majadita (Chaco Arid), *L. vallecurensis* captured in the area of Tauas (Altoandino) and *L. eleodori*, in the San Guillermo (Puna), during the year 2018. The specimens were captured and taken to the laboratory, and blood collection was done one week after their capture. Data on weight (at the time of capture and blood collection) and length H-C were taken. Immediately after extraction, three blood samples were performed to avoid degenerative cell changes. Subsequently, they were colored with May Grünwald Giemsa, for the morphological description of blood cells. The differential leukocyte count was performed, and some cell types were differentiated: heterophilic granulocytes, lymphocytes, and scarce thrombocytes. The proposed heterophile/lymphocyte ratio as a measure of individuals in response to stress was calculated in *L. darwini*. Due to the small size of the animals, adequate blood volumes could not be obtained to perform the blood count. The calculated measures were erythrocytes, lymphocytes, and heterophiles for the three species and the nucleus/cytoplasm ratio. In *L. darwini* heterophilic lymphocytes and granulocytes were more abundant than in *L. eleodori* and *L. vallecurensis*, it was also the only species in which some thrombocytes were observed. The proposed heterophile/lymphocyte ratio was calculated in *L. darwini* as a measure of the individuals in response to stress, which presented a 0.42 ratio, while in *L. vallecurensis* and *L. eleodori*, the amounts of heterophiles and lymphocytes observed were too low to obtain a relationship.

A109

INFLUENCE OF MELATONIN AND THE ANNUAL REPRODUCTIVE CYCLE ON THE ADRENAL CORTEX PROLIFERATIVE ACTIVITY OF A SEASONAL BREEDER (*LAGOSTOMUS MAXIMUS MAXIMUS*)

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The adrenal cortex is a major source of steroid hormones and its function is key for physiological adaptation and survival when facing homeostatic challenges. The viscacha (*Lagostomus maximus maximus*) is a rodent of nocturnal habits whose physiology and behavior vary according to modifications of environmental signals. The objective of this study was to assess the influence of melatonin and androgen on the proliferative activity of the adrenal cortex of the male viscacha through immunohistochemical detection of PCNA (proliferating cell nuclear antigen). A melatonin administration experiment was carried out randomly dividing animals into two groups: an experimental group (N=4) and a control group (N=4). PCNA expression was also studied in male viscachas along the year to assess the effect of the reproductive cycle along with serum testosterone determinations (N=4 per season). In the adrenal cortex, PCNA expression was observed in adrenocortical cells (PCNA-A) and in endothelial cells (PCNA-E). PCNA-E is considered a marker of neovascularization, a process necessary to cope with the gland nutrient and substrate requirements and to facilitate access of hormone products into the bloodstream. Melatonin administered animals showed a significant ($P<0.05$) lower number of PCNA-A (0.51 ± 0.07) when compared to the control group (1.40 ± 0.45). No significant difference could be established in the number of PCNA-E. During the male viscacha reproductive cycle, regression period animals exhibited the lowest serum testosterone levels (124 ± 16 ng/dL) showing significant differences ($P<0.01$) when compared to recovery (spring, 592 ± 59 ng/dL) and reproductive (424 ± 45 ng/dL) periods. In the annual reproductive cycle study, the highest number of PCNA-A was observed during late autumn (1.96 ± 0.27) exhibiting significant differences ($P<0.01$) when compared to all analyzed groups. On the other hand, winter animals exhibited the highest number of PCNA-E (2.96 ± 0.90) and showed significant differences ($P<0.01$) in relation to all other groups. No significant differences could be established in both the number of PCNA-A and PCNA-E between spring (0.39 ± 0.07 , 0.48 ± 0.10) and summer (0.32 ± 0.07 , 0.34 ± 0.15) animals. Our results indicate that proliferation in the adrenal cortex of the male viscacha is influenced by melatonin and androgen variations. Moreover, the influence of environmental stressors over the hypothalamic-pituitary-adrenal axis cannot be disregarded. In this seasonal breeder, the adrenal cortex proliferation regulation might be an important mechanism of adaptation to survival and reproductive demands when facing changing environmental conditions.

A110

VALORIZATION OF FILTERING LANDS IN THE RATION OF TWO CATEGORIES OF BEEF

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Waste from industrial activities generates air, water, and soil pollution. In the corn syrup, refining process, mixed cellulosic – earth filters filtering earth is used, leaving a cellulosic depleted residue, difficult to handle due to its nature and volume. On the other hand, sustainable livestock management implies practices that minimize the negative environmental impact and simultaneously obtain a product of high final biological value. The hypothesis suggests that the use of industrial waste is key to achieve sustainable productions. The objective of the work was to evaluate the feasibility of the inclusion of depleted filter earth in the diet of beef-producing bovines. In the trial, we used 28 Aberdeen Angus animals, divided into two groups based on the animal category (steer, NOV; and young whole male, MEJ). The ration was designed according to the animal nutritional requirements to gain 1 kg of live weight per day, with the inclusion of 20% of the filtering material (Nutritional composition: DM: 70%, CP: 14%, TND: 75, ME: 2.85 Mcal kg MS-1). Daily DM intake was estimated from the average of three consecutive days of supply and rejection measurement and live weight gain was register every 15 days. The evaluation period was 150 days. The results between the groups of animals were statistically analyzed by repeated mixed measures software R model. The ration responded as expected, good acceptancy, reaching the average final live weight of 378 ± 32 kg PV⁻¹ (366 ± 31 kg NOV and 390 ± 32 kg MEJ), average daily weight gain of 1.09 kg PV. Day⁻¹ (1.20 Kg MEJ and 0.99 kg NOV) and average feed conversion efficiency of 7.17 (6.31 MEJ and 8.03 NOV). The established slaughter criteria indicate the shortening of the fattening cycle, inferring lower greenhouse gas production (GHG), due to the lower total DM intake of the MEJ. The replacement of the 2.7 Tn of corn diminished the production of 1389.85 Kg CO₂ Eq. (equivalent carbon dioxide) of GHG, without even considering the positive costs of using second generation material as the filter residue. It is concluded that filter earth residues can be used as a component of bovine finishing rations and increasing efficiency in more than 20% in the MEJ category, which impacts on environmental, social, and economic benefits of the beef production process.

A111

SEASONAL STUDY OF THE EXPRESSION OF THE S-100 PROTEIN IN PITUITARY PARS NERVOSA OF VISCACHA (*LAGOSTOMUS MAXIMUS MAXIMUS*)

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The pars nervosa (PN) of the neurohypophysis is mainly constituted by pituicytes and other neuroglial cells such as microglial cells, astrocytes and oligodendrocytes, endothelial cells of blood vessels and myelinated axons. The PN pituicytes express the S-100 protein that has a regulatory role within the cell and on different target cells. The aim of this work was to study the seasonal expression of S-100 protein in pituitary PN of adult male viscachas (*Lagostomus maximus maximus*). The animals were captured in their habitat during the most representative months of their annual reproductive cycle. This cycle has three periods: reproductive (RepP), gonadal regression (RegP), and gonadal recovery (RecP). Four pituitary glands of each period were removed and processed for optical microscopy. The S-100 protein was used as markers of pituicytes and their expression was detected by immunohistochemistry. The

primary antibody used was the rabbit polyclonal anti-S-100 protein. Immunohistochemical staining was performed using a streptavidin-biotin-peroxidase complex method with the 3,3'-diaminobenzidine tetrahydrochloride (DAB) as the chromogen. A morphometric study was performed, and the percentage of immunopositive area for S-100 (% IA-S-100) was measured. The results obtained from these periods were statistically analyzed. Most of the pituicytes presented a cytoplasmic immunostaining pattern. However, some of them also exhibited nuclear staining. The nuclei were round, oval, and irregular in shape, with varying density of chromatin and an evident nucleolus. Numerous immunostained cytoplasmic processes in contact with blood vessels were observed. The % IA-S-100 varied throughout the annual reproductive cycle. The % IA during the RepP (13.09 ± 1.49) and RegP (9.77 ± 1.56) were significantly lower than in the RecP (25.61 ± 2.36 ; $P < 0.001$). These results demonstrated that the highest expression of S-100 protein was in the RecP. This period agrees with the increase of seasonal rainfall patterns during spring, so the viscachas have greater water availability in the environment. It is likely that the pituicytes participate in the seasonal regulation of the neurohormonal secretion through the expression of the S-100 protein. This protein is associated with functions such as modulation of enzymatic activity, stimulation of adenylate cyclase, maintenance of cell shape, and mobility. However, further research is needed to elucidate the relationship between S-100 expression and the neurohormonal PN secretion of *Lagostomus*.

A112

DETERMINATION OF CHEMICAL AND PHYSICAL QUALITY IN MEAT FROM HEAVY LAMBS

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Ovine meat production in the central region of Argentina is a profitable activity and allows faster return on investment than cattle breeding, however, it is a secondary production with low meat consumption per head. Promoting the production of heavy lamb will allow increasing the consumption by sale in cuts. Breed is a factor that affects the physical and instrumental quality of meat. The objective of this work is to evaluate the influence of two racial biotypes in heavy lambs on the physical and instrumental characteristics of meat. Twenty male lambs were studied, 10 from Corriedale breed (C) and 10 from Hampshire Down (HD). Weaning was carried out at 19–20 kg of live weight; then lambs were fed for 60 days on grazed in oats, alfalfa hay, and supplemented with corn (220 g/animal/day). Sacrifice was made between 31–36 kg of live weight, after 12 h of rest and fast. Hot carcass weight (HCW) was determined and after 24 h at 0–4 °C cold carcass weight (CCW) was taken. Temperature and pH were measured in left *Longissimus dorsi* (LD), (5th–10th rib) at 3 times post sacrifice: 0 h, 45 min, and 24 h. Left LD muscle samples (5th–13th rib) were taken to determine color by colorimeter, cooking losses (CL%), water retention capacity (WRC%), and tenderness with Warner Bratzler shear forces. Statistical analysis was performed using ANOVA. The results show average HCW values were 14.11 ± 1.38 kg in C and 14.00 ± 0.86 kg in HD and CCW 13.82 ± 1.44 kg in C and 13.59 ± 0.75 kg in HD. Chemical and physical determinations of meat quality did not show significant differences between breeds in the majority of the analyzed parameters. Both genotypes presented pH 24 hours values above the optimum (5.6–5.9); therefore, factors pre- and post-slaughter should be evaluated. Tenderness results were acceptable according to standardized values (below 4.5). These meats showed lower values of WRC and CL than those observed in other studies. The color indicator a* showed significant differences between biotypes ($P < 0.05$), C exhibited higher values than HD. Racial type only influenced meat color, where C lambs showed a more reddish color than HD; the rest of the meat quality characteristics evaluated were not affected by breed type and both exhibited acceptable meat quality values.

A113

APOPTOSIS IN PITUITARY PARS DISTALIS OF MALE VISCACHAS: QUANTITATIVE ANALYSIS DURING THE REPRODUCTIVE CYCLE

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Apoptosis, or programmed cell death, is a genetically controlled cell process whereby cells induce their own death in response to certain stimuli. The balance between this process and cell proliferation is key to maintaining tissue homeostasis in the pituitary gland during physiological endocrine events. A series of cysteine-dependent proteases, called caspases, participate in the intracellular mechanisms involved in apoptosis. The aim of this work was to localize and quantify apoptotic cells that express cleaved caspase 3 (CASP3) in different regions of pituitary pars distalis (PD) of adult male viscachas during their annual reproductive cycle (reproductive, gonadal regression and gonadal recovery periods). In each period, four pituitary glands were collected and processed for light microscopy. CASP3 was detected by immunohistochemistry and morphometrically quantified by image analysis. The immunoreactive (-ir) cells were counted and expressed as a percentage of the total number of cells per microscopic field. The values were statistically analyzed and expressed as mean \pm SEM. The immunostaining pattern was mainly cytoplasmic, although nuclear labeling was also observed. Abundant CASP3-ir cells were located around blood vessels. The total percentage of these cells did not differ significantly among the three periods studied ($P > 0.05$). However, there were significant differences in CASP3-ir cells between different PD zones during the reproductive cycle. In the reproductive period, numerous CASP3-ir cells were mainly located at the

rostral end, in the ventral and dorsal regions ($P<0.01$). Few positive cells were found in the ventral region of PD ($P<0.05$) during the gonadal regression period. In recovery gonadal period, the percentage of CASP3-ir cells did not differ significantly ($P>0.05$) in different PD zones. In addition, each of the different zones of PD was compared throughout the reproductive cycle. The percentage of CASP3-ir cells increased significantly at the rostral end, and it decreased significantly at the caudal end and dorsal region in the reproductive period compared with the gonadal recovery period ($P<0.05$). In the ventral region, the percentage of CASP3-ir cells was significantly higher ($P<0.01$) in the gonadal recovery period compared with the gonadal regression period. The results obtained demonstrated that there are regional variations in the expression of CASP3 throughout the reproductive cycle in the PD parenchyma of male viscachas. These differences might be due to a differential distribution of proapoptotic factors that reach the PD through the blood vessels of the pituitary hypothalamic portal system. It is likely that in the reproductive period, these factors mainly affect the rostral end and then be distributed to other PD zones. The changes in apoptosis rates are probably related to the maintenance and cellular renewal processes and to the variation hormonal secretion of different cell populations depending on the physiological demands of this rodent throughout the year.

A114

IMMUNOHISTOCHEMICAL STUDY OF THE PROLIFERATIVE ACTIVITY IN THE MALE AND FEMALE GONADS OF VISCACHA

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The proliferating cell nuclear antigen (PCNA) is an essential regulator of the cell cycle and has been used as a marker of proliferating cells in several studies. The viscacha (*Lagostomus maximus maximus*) is a wild South American rodent with nocturnal habits and seasonal reproduction. The objective of the present work was to study the PCNA expression in ovary and testis by immunohistochemistry, relating the results with the animal sexual maturity. The animals were captured in their habitat near San Luis city between 2013 and 2018. The females and males were classified into mature (>3 kg, 5–7 kg, respectively) and immature (<3 kg, 1–4 kg, respectively), according to their corporal weight and the light microscopy observations of the gonads. The testicular and ovarian samples were surgically removed and processed for optical microscopy. Ovarian and testicular structures were identified by optical microscopy and the percentage of immunopositive cells was determined. In females, immunostaining was observed in several granulosa and in few theca cells. The higher percentages of PCNA-positive cells were observed in granulosa cells of multilaminar primary follicles (40.02 ± 2.89) and unilaminar primary follicles (22.26 ± 1.17) of mature viscachas in relation to those observed in immature animals (18.18 ± 1.84 , 14.81 ± 1.17 , respectively). In male viscachas, the immunostaining was observed only in germinal cells, while the Sertoli cells were negative for PCNA-immunodetermination. The percentage of PCNA-positive cells in the seminiferous tubules was significantly higher in adults (29.75 ± 0.68) than in immature animals (7.36 ± 0.47). The immunostaining was moderate in the germinal and interstitial cells of studied groups. Our results showed that proliferative activity in female and male gonads develops in different cell types. In females, the highest proliferative activity is found in granulosa cells while in males it is observed in germ cells. In addition, adult animals showed greater proliferative activity than that observed in immature animals. These results suggest that the cell proliferation observed in the gonads, both male and female, ensures the maintenance of the gonadal structure and function during the animal reproductive life, thus ensuring the reproduction and species survival.

MICROBIOLOGY AND IMMUNOLOGY

A115

METHODOLOGY FOR REUSING YEAST IN A CRAFT BREWERY

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Beer is one of the most widely consumed alcoholic beverages in the world, which reaches all social status and is an industrial sector that generates millions of dollars a year. The craft breweries are in Argentina an innovative industrial sector that has experienced explosive growth in the last fifteen years, particularly high in the San Luis Province (Argentina). The growing demand for quality craft beers has driven significant technological development, process optimization, and cost reduction by the breweries. The yeast is critical to beer, which involves much more than converting sugars into alcohol, it contributes to flavor and aroma of beer too. The reusing of the yeasts is a wanted process that is being by brewers, as it generates significant savings in the production of new batches of beer. This reusing implies the collection of the yeast after the fermentation process from the bottom of the fermentation tank. The handling and collection of healthy yeast, avoiding biological contamination, is crucial to achieving a good beer quality. With the aim

to generate a technological transfer to the craft breweries, we developed a procedure of handling and collection of yeast for reuse in breweries. The work was carried out in the Kerze brewery, located at San Luis Province. Yeast samples were daily taken from the bottom of the fermentation tank during all the fermentation process (7 days). In the yeast samples viability was measured using standard methodology. The results indicated that the optimal time to the yeast collection for reuse is from 3 to 5 days of fermentation, when the total number of yeast cells is higher (1.43×10^9) and the best viability is recorded (90%). After the collection, the yeasts were stored 24 h in a refrigerator at 4 °C and its viability measured, showing viability minor than 50%. The results indicate that for yeast reusing, it must be harvested on the fifth day of fermentation and reused for a new batch within 24 h and stored at 4 °C. The implementation of this operative procedure for yeast collection generates an optimal process for reusing beer yeast (quantity of cells and viability necessary) and avoiding losses to craft breweries. Future studies are needed in order to increase the storage time of recovered yeast.

A116

PREVALENCE OF RECURRENT VULVOVAGINAL CANDIDIASIS AND *CHLAMYDIA TRACHOMATIS* IN WOMEN OF SAN LUIS CITY, ARGENTINA

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Vulvovaginal candidiasis is a condition that affects a great number of fertile women. It is considered the second cause of genital infection after vaginosis due to GAM (*Gardnerella*, anaerobes, *Mobiluncus*, *Mycoplasma*) complex. A total of 665 samples obtained between 2017 and 2019 were analyzed to determine the yeast species involved and its association with *Chlamydia trachomatis* in sexually active women. The average age was 34.83 ± 16.03 years. Of the total samples analyzed (n=655), 23.66% (n=155) were yeasts of the *Candida* genus. Yeasts were isolated by standard methods for fungal cultures on Sabouraud glucose agar and subsequently identified in CHROM-agar. The identification of the isolates showed: *Candida albicans* 93.55% (n=145), *Candida krusei* 3.87% (n=6) and *Candida tropicalis* 2.58% (n=4). *Chlamydia trachomatis* is a common sexually transmitted pathogen with significant implications on human reproductive health. It was also analyzed if *C. trachomatis* detection was requested by the general physician. Vaginal samples were collected by dacron swabs for *Chlamydia* test and this microorganism was diagnosed by the CHLAMY-CHECK-1 test. Out of 100 women with a positive *C. albicans* culture, 53% (n=53) were positive for the *Chlamydia* test, a much higher value than those observed in previous works. This could be mainly attributed to the fact that these exudates were performed on women with recurrent vulvovaginal candidiasis. In this study, we evaluated vaginal dysfunction through the analysis of basic vaginal states (BVS) using the methodology of the balance of the vaginal content (BAVACO). *C. albicans* and *C. trachomatis* were associated with BVS V and BVS II in 66.03% and 20.75%, respectively. Although *Chlamydia* infection is not associated with drastic changes in the composition and metabolic activity of the vaginal ecosystem, we observed changes in the vaginal microbiota when infections by bacterial and leishmaniform cells were associated. The microbiota plays a crucial role in maintaining the health and function of the female genital tract, preventing the colonization of urogenital pathogens and sexually transmitted infections. It would be convenient to perform an investigation in sexually active women affected by these clinical manifestations to identify associated germs and to evaluate the advantages of an early diagnosis that favors timely treatments to reduce complications and to interrupt the transmission chain of infection.

A117

ASSESSMENT OF HEPATIC ENZYMES AND BILE ACIDS IN GESTATIONAL INTRAHEPATIC CHOLESTASIS

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The appearance of pruritus during the pregnancy is related to the more frequent hepatopathy on pregnant women, called gestational intrahepatic cholestasis (GIC). The pruritus appears on feet and hands mostly at night and it is related to a bile transport disturbance in the maternal hepatocyte, producing a bile acid (BA) accumulation in the blood. While this is a condition that does not affect the pregnant woman and disappears with birth, it is a risk factor for the fetus. Hepatic enzymes, total bilirubin (TB), and direct bilirubin (DB) are indicators for the diagnosis of this pathology; however, the gold standard method is the determination of BA. The aim of this study was to correlate the values of hepatic enzymes, DB and TB with BA values in pregnant patients. The study group consisted of 105 pregnant women with a GIC clinic diagnosis who attended MPTB. We applied enzymatic methods for the determination of BA (Randox) and hepatic enzymes (Wiener) and a colorimetric method for DB and TB (Wiener) in an autoanalyzer Metrolab CM250. Statistical analyses were performed with GraphPad Prism 5.0. The patients' age ranged between 15 and 42 years old and the gestational weeks were between 25 and 40. A positive BA correlation with alkaline phosphatase (APho), DB and TB were observed, with correlation indexes (IC) of 0.42, 0.52, 0.53, respectively ($P=0.001$). Meanwhile, there was no BA correlation to glutamate

oxalacetate transaminase (GOT), glutamate pyruvate transaminase (GPT), gamma-glutamyl transpeptidase (GGT) and lactate dehydrogenase (LDH), with IC of 0.15, 0.11, 0.05, 0.13, respectively (*P*, not significant). No correlation was observed between patient age and gestational weeks. The predictive positive value obtained for BA was 42%. It can be concluded that to demonstrate GIC, the most sensitive test is the BA determination without dismissing APHo, DB, and TB contributions. These data are coincident with reports of bibliography.

A118

ANTIMICROBIAL EFFECT ON PERISHABLE FOODS OF CELL FREE SUPERNATANT FROM *PEDIOCOCCUS SPP SL19* ISOLATED FROM GOAT MILK. SAN LUIS. ARGENTINA.

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In recent decades, the food industry has focused on reducing the use of chemical preservatives. Numerous investigations aim to discover new biomolecules from microorganisms to be used as food preservatives. Lactic acid bacteria (LAB) have been used for centuries in food processing and are very important for their contribution to the value of products and for their ability to produce antimicrobial substances. In perishable fresh foods of animal origin, the presence of *Escherichia coli* (enterobacterium of fecal origin) may indicate improper handling and/or improper storage. LAB strain selected for this study was isolated and named as *sl19*, from samples of goat milk collected from stainless steel drums in a dairy farm (San Luis, Argentina). The LAB strain was biochemically typified as *Pediococcus spp* and designated as *Pediococcus spp sl19*. The purposes of this study were to evaluate the antimicrobial activity of the *sl19* LAB strain and study its effect on the conservation of cow milk. The antimicrobial activity of cell-free supernatant (CFS) and neutralized cell-free supernatant (NCFS) of a culture of the strain under study was evaluated against *E. coli* (liquid medium method). To 1 mL of sensitive strains suspension (6×10^8 UFC/mL) was added 0.5 mL of CFS or NCFS and 0.5 mL of medium Trypticase Soy Broth (TSB). The mix was incubated for 6 h a 37 °C. Absorbance measurements were made at 700 nm. The inhibition percentage (%I) was calculated according to the formula $I = 1 - A_s/A_c$, considering A_s and A_c as sample absorbance and control absorbance, respectively. In positive control assay, the CFS was replaced by MRS broth. For the processing of cow milk as perishable food, aliquots of 8 mL of milk were treated with 1 mL of CFS or 1 mL MRS broth (controls) and 1 mL of indicator strain suspension (10^8 CFU/mL). Aliquots were incubated at 37 °C for 12 h, 1 mL of 10-fold serial dilution was poured on a Petri dish using 1.5% TSA medium. The plates were incubated at 37 °C for 24 h. *E. coli* counts in samples were determined according to National Standard Test Methods for Food Microbiology. CFS and NCFS from *Pediococcus spp sl19* showed inhibitory activity of 70% and 34% respectively, against *E. coli*. Statistical analyses were carried out with InfoStat software using Friedmann nonparametric variance test ($P < 0.05$ was considered to be significant). The inhibitory substances present in the CFS of the culture of LAB strain caused the inhibition of the growth of *E. coli* in milk (values of the counts expressed as CFU/mL) when compared with the growth of the indicator strains in the controls (aliquots of samples treated with MRS broth). There is a significant difference (< 0.0001) between the counts of controls and those treated with CFS (Friedman test). These studies confirm that *Pediococcus spp sl19* produces antimicrobial substances, probably organic acids among others. These antimicrobial substances could be used to prevent the spoilage of milk caused by *E. coli* and to avoid the presence of pathogen strains of *E. coli* on food, which are the causal agents of some food-borne illness.

A119

EXPLORING ANTI-CYANOBACTERIAL ACTIVITY OF *BACILLUS VELEZENSIS* SL-6

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Different *Bacillus* species showed strong antimicrobial activity by producing diffusible metabolites through ribosomal and non-ribosomal biosynthesis and volatile organic compounds (VOCs). The latter have complex compositions and their relative abundances are influenced by many biotic and abiotic factors. Bacilysin, a non-ribosomal dipeptide produced by *B. velezensis* FZB42, has been described as an antagonist of *Microcystis aeruginosa*. The aim of this study was to explore the anti-cyanobacterial activity of these metabolites from *B. velezensis* SL-6 against unicellular and filamentous organisms. Likewise, the presence of non-ribosomal peptide synthetase (NRPS) for bacilysin was evaluated. Antagonistic activity of diffusible metabolites produced by *B. velezensis* SL-6 against *Tolypothrix tenuis* and *M. aeruginosa* was tested by agar plug-diffusion method using an inoculum of each one containing ~1 µg/mL chlorophyll *a* in a double layer agar of Watanabe (W) or BG11 agar medium, respectively. *B. velezensis* SL-6 was spread on a Glucose Peptone Agar (~ 10^8 CFU/mL), then 10 mm plugs were cut and deposited over plates containing each cyanobacterium lawn. Incubation was performed at 25 ± 2 °C with 3000 lux for 10 days. *B. velezensis* FZB42 was used as a reference strain. To evaluate the anti-cyanobacterial effects of VOCs, a double chamber Petri dish was used. *Bacillus* and cyanobacteria were inoculated in agar media previously described. The plates were incubated into plastic bags at the same conditions previously reported. To screen local isolate SL-6 for genes involved in the biosynthesis of bacilysin (*bacB* gene), the PCR was carried out. Thermal cycling

conditions were: initial activation at 94 °C for 5 min; 35 cycles of 94 °C for 1 min, 51 °C for 1 min and 72 °C extension for 1 min; and a final extension at 72 °C for 7 min. A total of 5 µL of each amplification reaction was analyzed by electrophoresis using a 1.5% agarose gel followed by GelRed staining, visualized, and photographed under ultraviolet light. *B. velezensis* SL-6 showed the ability to produce diffusible and volatile anticyanobacterial compounds. Former compounds were active only against *M. aeruginosa*; two defined inhibition zones were observed around the plug (15.5 and 46.7 mm). FZB42 strain presented the same pattern of inhibition (19 and 50.3 mm), without showing any statistically significant difference $P > 0.05$ (N=3). VOCs inhibited the growth of the two tested cyanobacteria. The SL-6 strain showed an amplicon of 747 bp corresponding to the *bacB* gene of NRPS for the synthesis of bacilysin. These are the first results of a screening of anticyanobacterial activity from *B. velezensis* SL-6 produced by a wide diversity of biocompounds.

A120

POLLEN IN INDOOR ENVIRONMENTS? POLLEN REPORT ON AN INDOOR ENVIRONMENT AND ITS VARIATION DUE TO METEOROLOGICAL FACTORS

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Outdoor air content has a direct influence on indoor air microbiota. In the same way, weather factors that affect the seasons of the year also influence air ecology in different environments. These facts also relate directly to pollen allergy episodes of those people who are in contact with allergen potential particles such as pollen grains. The objective of the work was to identify the content of biological particles in the air of a building of the National University of San Luis and to relate it to some meteorological factors. In addition, through surveys, it was tried to relate the amount of pollen grains with allergy episodes of people who worked in the vicinity of the sampler. A continuous aerobiological record was carried out inside the building of the National University of San Luis, between August 2012 and July 2013. A Lanzoni VPPS 2000 volumetric trap was used in a basement hall. Samples were dyed with basic fuchsin and then read through an optical microscope at 1000X. Different pollen grains of type were identified: Cupressaceae, *Morus*, *Populus*, *Eucalyptus*, *Pinus*, *Ligustrum*, *Urtica*, *Ulmus*, *Parietaria*, Amaranthaceae/Chenopodiaceae. Correlation indexes between pollen registers and some weather variables such as temperature, relative humidity, wind speed, and solar radiation were analyzed. All results proved to be positive: temperature ($r=0.59$), relative humidity ($r=0.6$), wind speed ($r=0.8$) and solar radiation ($r=0.79$). In addition, it was possible to establish a significant positive relationship ($r=0.73$) between the occurrence of allergy episodes, in people working in close proximity to the aerobiological sampler, and pollen peaks recorded during the months analyzed. Indoor and outdoor pollen grain quantity and type were related. According to the results obtained, we conclude that people who are hypersensitive to certain pollen grains may be affected by symptoms of pollen allergy even inside a building. Therefore, it would be safer to keep windows closed especially in those periods when pollen concentrations are the highest.

A121

ISOLATION OF THE MICROALGAE *SCENEDESMUS* SP.

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The freshwater green microalga *Scenedesmus* sp., one of the commonest genera worldwide, is found in rivers, ponds, and lakes. It is sensitive to most toxic substances and in general, more sensitive than organisms of higher trophic levels. Additionally, it is easy to handle in laboratory conditions, has short generational time and rapid physiological responses, which allows the effects of pollutants to be evaluated in short terms, and makes it a valuable bioindicator of environmental impact. The objective of this work was to isolate and grow *Scenedesmus* species for future use in environmental monitoring bioassays. The sample was obtained from an artificial pond with a phytoplankton net. In order to separate algal populations, standard plating methods were applied with two different media: BBM (Bold's Basal Medium) and BG11 added with 1.5% agar-agar and incubated at 25 ± 2 °C in a humid chamber with continuous illumination of ~3000 lux for 16 days. Following the isolation, the individual colonies were transferred to liquid media and incubated in the conditions before mentioned. Then, inocula of 3.4×10^4 cells/mL were transferred to batch liquid cultures and shaken by hand twice daily. The growth was evaluated by measuring the optical densities (OD₅₈₀) and chlorophyll *a* and *b* contents. Cell count examination was performed using a hemocytometer (Neubauer). The original sample was examined under a light microscope (40X magnification) and showed common green microalgae and unicellular and filamentous cyanobacteria. *Scenedesmus* sp was isolated and could grow in both culture media showing the highest growth response (OD₅₈₀ 0.97) and total chlorophyll content (10.6 µg/mL) in BG11. The final cell count was invalidated in BG11 by cell clumping, while in BBM reached 4.35×10^6 cell/mL. Morphological characterization using light microscopy revealed the presence of cells of ovoid and fusiform shapes with single to four celled coenobia even, to a lesser extent, eight-celled ones. In this preliminary study, *Scenedesmus* sp showed faster growth response in BG11; however, a more homogeneous growth with four celled coenobia and a very low number of unicells was observed in BBM. More studies related to growth nutritional conditions should be performed in order to minimize morphological variations observed in *Scenedesmus* sp due to its phenotypic plasticity.

A122

SEROLOGICAL SURVEY OF HEPATITIS B IMMUNITY IN HEALTHCARE WORKERS IN MATERNIDAD PROVINCIAL “DRA TERESITA BAIGORRIA”

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Healthcare workers (HCW) are exposed to preventable infectious diseases, especially hepatitis B virus (HBV). Vaccination is the most cost-effective method of preventing and controlling HBV infection. In Argentina, vaccination against HBV is mandatory for HCW since 1992 (Law 24.151), it is incorporated into the immunization schedule for newborns from the year 2000 to contribute to the elimination and control of HBV, and in 2013 it was universalized. The aim of this study was to determine the immunity of HCW against HBV. A descriptive and prospective study was carried out with HCW, who stated they were vaccinated (1997-2019), at MPTB from August to September 2019. Exclusion criteria: HCW with hepatitis B surface antigen (HBsAg) reactive. Blood samples were used to measure antibodies against the HB surface antigen (anti-HBs) and HBsAg by ELISA using commercial kits the Biokit and Wiener lab respectively. Anti-HBs levels of ≥ 10 mIU/mL were considered reactive and protective. The statistical analyses were performed with the software GraphPad Prism 5.0. Of 151 randomly selected HCW 82% (124) were females and 18% (27) males, the average age was 40 (range 25–67). The distribution according to professional category was: 30% (45) nurses, 16% (24) technicians, 13% (19) physicians, 12% (18) administrative staff, 8% (12) biochemists, 7% (11) obstetricians, 3% (5) personal responsible for cleaning, 3% (5) pharmacists, 2% (3) ambulance driver, 2% (3) social worker, 1% (2) physiotherapists, 1% (2) sanitary workers, 1% (2) phonoaudiologists. All HCW were non-reactive for HBsAg. The overall prevalence of anti-HBs in the HCW was 62%. The group the HCW aged 25–34 had a significantly higher prevalence (67%) compared with the groups HWC aged 35–45 (62%, $P < 0.02$), aged 46–55 (44%, $P < 0.001$) and aged 55–67 (58% $P < 0.01$), respectively. There was 7% (11) were unaware of their vaccination status and who were vaccinated in the last 6 years, presented higher protection (66%). In the most exposed professions, biochemists, nurses, physicians, and technicians, the prevalence of protection was 42% (5), 64% (29), 79% (15), and 71% (17), respectively. The most important result of this study was that nearly two-thirds (62%) of the HCW had a protective anti-HBs level, coinciding with published data. In any case, an anti-HBs level < 10 mIU/mL is equivalent to not being vaccinated, as some vaccinated subjects will not respond to vaccination and, in responders, anti-HBs titers decrease over time, becoming negative in many cases. Although HBV infections have declined substantially since the introduction of HBV vaccination, the risk of exposure to HBV persists in the healthcare setting. Therefore, there is a need to improve vaccination programs in HCW, in order to protect them from infected persons and vice versa.

A123

ADVANCES IN THE STUDY OF INTRACELLULAR TRANSPORT OF *BRUCELLA ABORTUS* 2308 (VIRULENT STRAIN) IN MACROPHAGES

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In recent years, this research team has progressed in the study of intracellular transport of the virulent strain (2308) of *Brucella abortus* in macrophage cell lines (J-774 and Raw) through multiple microscopic approaches. We demonstrated the transit of *B. abortus* 2308 through compartments of the endocytic pathway, marking early endosomes and lysosomes with gold (20 and 60 nm, respectively). We have shown that *B. abortus* 2308 occupies two different types of compartments: phagolysosomes and modified phagosomes, significantly reducing their fusion to endosomes. On the other hand, macrophages transfected with GFP-Rabs were used to evaluate the location of Rab 5 and 11 proteins, involved in the vesicular transport of the cell. Confocal microscopy showed that *B. abortus* 2308 (stained with Rhodamine) recruits Rab11 to the phagosome membrane that contains them, in the form of discrete patches. In addition, *B. abortus* 2308 co-locates with vesicles that overexpress Rab 5 in a large proportion. The permanence of Rab 5 and 11 associated with phagosomes containing *B. abortus* 2308 suggests that the bacteria actively retain these Rabs to avoid maturation of the phagosome that contains them. Subsequently, the effect of kinase inhibitors (AKTi) on the multiplication and intracellular survival of *B. abortus* 2308 at different times post macrophage infection was studied by confocal microscopy. These results allowed confirming that *B. abortus* 2308 uses the AKT/AS160 pathway to activate Rabs involved in the transport of nutrients necessary for its replication and the generation of a safe intracellular site. The compound used as an AKT inhibitor could constitute a new pharmacological approach for the treatment of brucellosis.

A124

DETERMINATION OF CYANOBACTERIA PRESENT IN BIOLOGICAL CRUSTS OF SOILS AFFECTED BY FOREST FIRE

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Forest fires are one of the main landscape modelers which cause, among other things, destruction of vegetation and soil losses with the consequent breakdown of ecosystem balances. Biological Crusts (BCs) are an intimate association between fungi, algae, cyanobacteria, bryophytes, and liverworts that are widely distributed in the world's soils. They are the first colonizers in ecosystems that have suffered alterations of this magnitude and play a fundamental role in erosion and runoff control, in addition to their ability

to fix atmospheric nitrogen and carbon. The objective of this work was to determine the diversity of cyanobacteria in biological crusts (BCs) in places affected by fires. We worked with soils from Córdoba mountains, from Quebracho Ladeado and La Calera towns, and from San Luis mountains, Los Lobos town. Three days after the fire event, five samples of BCs in burnt sites and three of their corresponding unburned control samples were taken to analyze the presence and determine the cyanobacteria present in the studied sites. The BCs were hydrated and analyzed in their fresh status by direct observation under an optical microscope. The cyanobacteria were classified as non-fixative (NF) and fixative (F) and the latter, in turn, were classified as fixatives with heterocysts (FCH) and fixers without heterocysts (FSH). Preliminary results show that the *Nostoc*, *Phormidium*, and *Oscillatoria* genera are found in all three sites. Differences in amounts and percentages of cyanobacteria identified were observed between soils affected by fires and those that did not suffer the effects of fire. Quebracho Ladeado had a total of 75 cyanobacteria on burnt soil, 76% were NF, and the remaining 24% were F, of which 5% presented heterocysts (FCH) and 95% did not (FSH); 86 cyanobacteria were counted in the control sample being 80.2% NF and 19.8% FSH. Los Lobos presented 18 cyanobacteria in the soil affected by the fire, 66.7% were NF and 33.3% FSH, while 185 cyanobacteria were counted for BCs in the control sample: 88.1% NF and 11.9% FSH. Finally, La Calera presented values of 188 cyanobacteria in BCs of burnt soils; 85.6% were NF and 14.4% were FSH, while in the control sample, 59 cyanobacteria were found: 84.7% were NF and 15.3% were FSH. The values observed in burnt soils, with respect to their control sample, indicate that the intensity of the fire is directly related to the quantification of the cyanobacteria present in the analyzed BCs. This allows us to conclude in advance that fire has immediate effects on the amount and composition of cyanobacteria present in the Biological Crusts of the studied soils. The magnitude of this effect is directly related to the fire intensity.

A125

A NOVEL METHOD FOR PURIFICATION OF JARILLA PROTEINS BY RAPID PROTEIN LIQUID CHROMATOGRAPHY (FPLC)

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Larrea divaricata Cav. (Zygophyllaceae) is a plant native to South America commonly known as "jarilla". In Argentina, folk medicine uses plants widely, including *L. divaricata*. The proteins obtained from crude jarilla extracts have been used for the immunogenic characterization of this plant and to establish its relationship with different pathogens such as *Pseudomonas aeruginosa*, *Proteus vulgaris*, *Klebsiella pneumoniae*, and *Escherichia coli* O157:H7, where a strong cross-reactivity has been observed with proteins belonging to these bacteria. In addition, the antigenic similarity of different protein fractions of *L. divaricata* with cellular and extracellular proteins of *Candida albicans* has been demonstrated. In this study, purified protein fractions of the jarilla crude extract (JPCE) were investigated using rapid protein liquid chromatography (FPLC). A novel method was used, with an extraction buffer with reducing components to prevent the oxidation of polyphenols. These compounds interfere with the chromatographic separation of proteins by damaging the columns used. The objective was to separate the proteins from the JPCE, to obtain a high degree of purity of each protein component. The samples (leaves of *L. divaricata*) were collected in the city of San Luis, Autopista de los Serranías Puntanas and Provincial Route No. 3. Subsequently, they were frozen at -80 °C, thus allowing mortar spraying. Protein extraction was performed by mixing the powder obtained with extraction saline buffer under reducing conditions (0.5 mM KH₂PO₄, 8 mM Na₂HPO₄, 5 mM EDTA, 50 mM NaCl, 5mM Cysteine, pH 7.4) in 15% (w/v) ratio. The extract was partially purified using membrane concentrators with a 10 kDa cut. The total purified proteins were precipitated with 96% ethanol (4:1) and the precipitate obtained was subjected to rapid liquid chromatography of ion exchange proteins. Protein concentration was determined by the Bradford protein assay. The different fractions were resolved by SDS-PAGE, where a protein profile similar to those obtained with other previously reported methods was observed. Antigenic preparations were obtained according to fractions of band patterns and similar protein concentrations: J1 (33-103 kDa), J2 (22-26kDa), J3 (14-26 kDa) and J4 (14kDa). These findings could be important in the development of new methods to prevent infection of pathogenic microorganisms, based on plant-specific cross-reaction proteins without the need to expose the patient to the pathogen, in a safe and effective way. The high purity protein components obtained by this methodology can be used, in future studies, as immunogens in possible vaccine formulations.

A126

PREVALENCE OF URINARY TRACT INFECTION IN PREGNANT WOMEN OF MATERNIDAD PROVINCIAL "DRA TERESITA BAIGORRIA". DIAGNOSTIC UTILITY OF URINARY STRIP AND GRAM STAIN

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Urinary tract infection (UTI) is the most common bacterial infection during pregnancy. Regarding the literature, the prevalence can be up to 20 %. Untreated UTI can be associated with serious obstetrics complications such as premature membrane rupture or premature birth. The objectives of the work were to determine the prevalence, most frequent germs, and the diagnostic utility of urine strip and gram stain for diagnostic of UTI in pregnancy, taking the culture as a diagnostic gold standard. 435 samples of urine of pregnant women during the period of March 2019 to August 2019 were analyzed. All of them come for patients of the laboratory of

Provincial Maternity. The urine strip was used to determine leucocitary esterase and nitrites. A Gram stain of whole urine was made. The samples were placed on cystine lactose electrolyte deficient (CLED) medium. The positive samples were identified by a phenotypic biochemical test. To determine the diagnostic capacity of the urine test strip and Gram stain, the Receiver Operating Characteristic (ROC) curves were analyzed. The prevalence of UTI was 12.87% and the main bacteria were *Escherichia coli* (85.7%), *Klebsiella pneumoniae* (3.57%) and *Enterococcus faecalis* (3.57%). Sensitivity, specificity, positive and negative predictive value were: leukocyte esterase (62.5%, 88.4%, 45.5%, 93.8%), nitrites (55.4%, 95.9%, 67.4%, 93.3%), gram stain (100%, 92.4%, 67.5%, 100%), leukocyte esterase + nitrites (71.4%, 94%, 74.1%, 97.5%), leukocyte esterase + nitrites + gram stain (96%, 98.6%, 85.7%, 99.7%). The ROC curves showed an area under the curve of 88%. From the analysis of these results, the ROC curves allowed us to verify the overall validity of the test strip and Gram stain for the diagnosis of UTI in pregnant women. The best test performance was the combination of were leukocyte esterase + nitrites + Gram stain with optimal sensitivity, specificity, and positive and negative predictive value.

A127

CHARACTERIZATION OF FLAGELAR PROTEIN FROM CULTURES OF *CLOSTRIDIUM CHAUVOEI*

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The anaerobic Gram (+) mobile bacterium, *Clostridium chauvoei*, is the black leg pathogen, a disease that causes severe toxemia and high mortality in cattle, sheep, and many other domestic and wild animals. It is considered the most important *Clostridium* that produces economic losses in cattle. The immunity of *C. chauvoei* is mainly considered as anti-cell, somatic and antiallergic antigens, being the most studied. Bacterial flagella, anchored at one end of the cell membrane, are special locomotion organelles in most active mobile bacteria. In flagella, identical structural units of single flagellin are joined by non-covalent bonds, which in turn helically assemble the flagellar filaments. Unlike other PAMPs, flagellin has only a peptide nature, which makes it a robust stimulator of innate and specific immunity. The objective of this work is to obtain, purify, and characterize the flagella of *C. chauvoei*. The strains of *C. chauvoei* ATCC 10092 were grown in a clostridial medium using a Batch type system in anaerobiosis generated by the VAS-PAR method. Logarithmic phase cells were washed and resuspended in distilled water. The deflagellation of the cells was carried out by shaking them in a tube containing glass beads for 10 min. Then, the deflected cells were removed by centrifugation at 3500 x g for 20 min. The supernatant obtained was subjected to centrifugation in two stages. First stage, it was centrifuged at 16000 x g to remove cell debris for 20 min. Second stage, the supernatant was centrifuged at 40000 x g to sediment the flagella for 2 h. The flagellar solution was obtained by resuspending the pellet in PBS. Protein content was determined by the Bradford Method. Next, the samples obtained at each stage were subjected to SDS-PAGE using the buffer system described by Laemmli. The protein profiles obtained revealed the presence of a common band of 50 kDa. In the final stage, the presence of two bands corresponding to a molecular mass of 47 and 15 kDa was observed. It is known that the flagella of *C. chauvoei* have a profiled protein composed of bands whose molecular masses are 46, 73, and 100 kDa. On the other hand, the presence of the main band of 46 kDa, which has been described as 56 kDa in other studies is attributed to the electrophoretic method differences. The scourge of *C. chauvoei* has strong immunogenicity and is proposed as a candidate for vaccine development. This work provides a low-cost protocol for obtaining the scourge in order to continue studying its participation in the immune response against *C. chauvoei* infection.

A128

ULTRASTRUCTURE OF THE ADHESION OF *PSEUDOBUTYRIVIBRIO XYLANIVORANS* TO CANCER CELLS

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Pseudobutyrvibrio xylanivorans (Px), a wild-type bacterium isolated from the rumen of Creole goats, has been intensively studied during this decade. Px strains generate an acidic environment through the production of butyrate and lactate. In addition, Px produces bacteriocins that inhibit some potential pathogenic bacteria and produce the biohydrogenation of linoleic acid. Butyrate, an important nutrient for normal colonic cells, reduces proliferation and induces apoptosis of human colorectal adenocarcinoma (HCRA) cells. Bacterial adhesion to the epithelium of the digestive tract of the host is a fundamental requirement for the selection of probiotics since it influences the time of permanence of bacteria in the gut. Recently, we have shown that Px exhibits significantly greater adherence to HCRA cells, compared to demonstrated adherence to He-La cells, a non-colonic epithelial cell line. In order to deepen the knowledge of the mechanism of adhesion of Px, we use scanning electron microscopy (SEM) to examine adhesion to SW480

cells (Dukes type B colorectal adenocarcinoma cells). SEM micrographs showed that 1 h after the interaction. Px was trapped on the surface of the cancer cell, on laminar structures surrounding discrete segments of the bacterium, as described above for the membrane wraps (MWs). For all the above, we consider to deepening the study of the molecular mechanisms that underlie the phenomenon of adhesion of Px to HCRA cells.

A129

ANTIMICROBIAL EFFECT ON A BASE CREAM OF FREE CELL SUPERNATANTS FROM *LACTOBACILLUS PARACASEI* SSP *PARACASEI* 1. *SL57* ISOLATED FROM GOAT MILK. SAN LUIS. ARGENTINA

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The emergence of resistance to available antifungals and the side effects of these medications have restricted their use as long-term prophylactic agents for *Candida* sp. The probiotics are presented as an alternative for the treatment of candidiasis. The protective role of lactic acid bacteria (LAB) lies in their capability to decrease pH and synthesis of bacteriostatic and bactericidal substances. These substances include hydrogen peroxide, lactic acid, carbon dioxide, and bacteriocins which are defined as peptides produced by bacteria that inhibit or kill other related and unrelated microorganisms. There are few studies on the use of lactic bacteria in creams. Strain selected for this study was isolated and named as *sl57*, from samples of goat milk collected from stainless steel drums in a dairy farm (San Luis, Argentina). The LAB strain was biochemically typified as *Lactobacillus paracasei* ssp. *paracasei* 1 designated as *L. paracasei* spp. *paracasei* 1 *sl57*. The purposes of this study were to evaluate the spectrum of antimicrobial activity of a LAB strain and study its antimicrobial effect on base cream. The antimicrobial activity of cell-free supernatant (CFS) and neutralized cell-free supernatant (NCFS) of a culture of the strain under study was evaluated against *Candida albicans* for liquid medium method. To 1 mL of sensitive strains suspension (6×10^8 UFC/mL) was added 0.5 mL of CFS and 0.5 mL of medium Trypticase Soy Broth (TSB). The mix was incubated for 6 h at 37 °C. Absorbance measurements were made at 700 nm. The inhibition percentage (%) was calculated according to the formula $I = 1 - As/Ac$, considering As and Ac as sample absorbance and control absorbance, respectively. In the control assay, the CFS was replaced by MRS broth. Because the base cream has components (cetearyl alcohol, EDTA) that could inhibit the growth of *C. albicans*, dilutions of the cream were treated with *C. albicans* to determine the appropriate dilution in which *C. albicans* grows correctly, to be treated, and it was concluded that it was 10^{-2} . Aliquots of 8 mL of the dilution of base cream were treated with 1 mL of CFS or 1 mL MRS broth (controls) and 1 mL of indicator strains suspension (10^8 CFU/mL). Aliquots were incubated at 35 °C for 12 h, 1 mL of 10-fold serial dilution was poured on a Petri dish using Sabouraud Agar medium. The plates were incubated at 37 °C for 24h. *C. albicans* counts in samples were determined. CFS and NCFS from *L. paracasei* spp. *paracasei* 1 *sl57* showed inhibitory activity of 54% and 24% respectively against *C. albicans*. Although there was no significant difference, it is observed that CFS tend to inhibit the growth of the indicator in the treated base cream (values of the counts expressed as CFU/mL), when compared with the growth of the indicator strains in the controls (aliquots of samples treated with MRS broth). These results raise the need for more extensive studies to investigate experimental conditions (working with concentrated CFS for example) that potentiate CFS inhibitory activity so that it can be used for the topical treatment of infections.

A130

PRELIMINAR STUDY OF THE GLYCOPHENOTYPE OF *YERSINIA ENTEROCOLITICA* SECRETED VIRULENCE FACTORS AND THEIR INTERACTION WITH GALECTIN-1

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Yersinia enterocolitica (Ye) is a Gram-negative bacterium that causes gastrointestinal infection and eventually, extraintestinal manifestations. Ye evades the immune response by injecting *Yersinia* outer proteins (Yops) into the cytosol of host cells using type III secretion system. In addition, the lectin Galectin-1 (Gal-1) is a proto-type member from the family of galectins widely distributed in immune compartments. Knowledge about the interconnection between Yops and endogenous host proteins is limited. The aim of this work was to characterize the interaction between Gal-1 and Yops and obtain preliminary *in silico* models of noncrystallized virulence factors of Ye. Our results showed the presence of Gal-1-permissive glycoepitopes in the Yops. Moreover, we demonstrated that Gal-1 binds to Yops by glycan-protein interactions and that Gal-1 protects some Yops of trypsin digestion. Additionally, the sequence corresponding to the virulence factor, YopE was obtained from the National Center for Biotechnology Information database (NCBI) and comparative modeling was performed by searching for resolved protein structures in the Protein Data Bank (PDB). We used the catalytic domain of YopE from *Y. pestis* (1HY5), YopE of *Y. pseudotuberculosis* (1L2W) and the human Helicase RECQL4 (5LST) as a query. The results provided potential templates for different primary structural regions. The alignments were used to perform the YopE homology modeling using MODELLER 9v12 software. The preliminary models obtained were evaluated using PyMOL and COOT software. In addition, the intrinsic disorder was assayed using DisEMBL and DisProt software. Future studies are needed to understand cellular and molecular mechanisms to clarify the potential clinical relevance of our findings, focusing on the interactions between Gal-1 and Yops during Ye infection.

A131

SEARCH FOR THE PSEUDOMONAS GENUS IN *LARREA DIVARICATA* CAV. SAMPLES FROM DIFFERENT AREAS OF SAN LUIS, ARGENTINA

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Larrea divaricata Cav. ("jarilla") is a plant widely spread in America and is used in folk medicine to treat several pathologies. Studies in our laboratory have shown that proteins from aqueous crude extracts of jarilla exhibit cross-reaction with proteins of *Pseudomonas aeruginosa*, a human opportunistic pathogen. Although in our previous works *P. aeruginosa* was not detected in the jarilla's microbiota, it was considered appropriate to carry out further studies on the possible presence of plant pathogenic *Pseudomonas*, such as cultivation at appropriate temperatures and in suitable media for these environmental species. The aim of the present study was to search the *Pseudomonas* genus in the microbiota of jarilla specimens collected from different geographical areas of San Luis, Argentina, because this bacterial genus is a frequently isolated plant pathogen or belongs to the normal microbiota. For microbiological analysis, homogenates of the different samples were diluted in peptone water, incubated at 25 °C for 24 h and then plated in King B agar for *Pseudomonas* spp. King B agar plates were incubated at 25 °C for 24 h. Colonies that grew on King B agar were exposed to ultraviolet light to observe the presence of the fluorescent pigment, characteristic of some species of this genus. Also were subjected to the TSI agar test, in order to demonstrate if they were colonies of non-fermentative Gram-negative bacilli (NFB) or not. In addition, Gram staining was performed to confirm the development of Gram-negative bacilli, characteristic morphology of the genus. Triplicate trials were performed. The microbial load analysis was negative for the *Pseudomonas* genus. King B plates with growth showed no fluorescence. The TSI test was negative for NFB. Gram staining did not show the presence of Gram-negative bacilli. In conclusion, the results of this study provide data referred to the presence of *Pseudomonas* genus in Jarilla's microbiota. In the samples analyzed and through the tests performed, this genus was not isolated from the plant microbiota.

A132

CHEMICAL COMPOSITION AND ANTIBACTERIAL ACTIVITY AGAINST *STAPHYLOCOCCUS AUREUS* ATCC 25923 OF THE ESSENTIAL OILS FROM *BACCHARIS SPARTIOIDES* ("PICHANA") EXTRACTED BY STEAM DISTILLATION

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Baccharis spartioides is a plant aromatic member of the Asteraceae family. Its branches are strongly scented because of the presence of a wide variety of volatile compounds that have demonstrated antimicrobial activity. The present work aims to determine essential oil composition of *B. spartioides* (BsEO) and assess the antimicrobial activity against *Staphylococcus aureus*. BsEO was obtained by steam distillation, and its composition was determined by GC-MS. The antimicrobial activity was evaluated by a disc diffusion test in which the diameter of the clear zone around the disc was measured and expressed in millimeters. It was assessed the antimicrobial activity by increasing volumes of BsEO impregnated on disc using 6 µL of pure limonene (97%) and Amikasin (30 µg) as discs control. It was considered diameters >20 mm by significant inhibitory effect. The major constituents of BsEO were limonene (44%), sphaulenol (12%) and caryophyllene oxide (8%). Since limonene is BsEO major constituent, we considered this component as responsible for antimicrobial activity. Thus, the calibration curve of µL of limonene vs diameter was made and it was determined that 6 µL of BsEO was the minimal volume for significant inhibitory effects. Major antimicrobial activity was expressed in discs impregnated with 5, 6, 7, and 8 µL of BsEO, which diameters were inferior to 20 mm. Results indicate BsEO present low antimicrobial activity (<20 mm) but inhibition diameters founded (10-15 mm) could correspond to bacteriostatic activity.

A133

ASSESSMENT OF VIRULENCE GENOTYPIC MARKERS IN *YERSINIA ENTEROCOLITICA* BIOTYPE 1A STRAINS OF DIFFERENT ORIGINS BY PCR

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Yersinia enterocolitica B1A comprises a heterogeneous group of strains that encompasses a wide variety of serotypes. They have been considered non-pathogenic microorganisms due to the lack of plasmid and chromosomal virulence determinants that characterize pathogenic strains; however, strains of this biotype are commonly reported not only from healthy individuals but also from patients with gastrointestinal disorders. The comprehension of pathogenic mechanisms of B1A strains should focus on certain chromosomal virulence determinants associated to adhesion and invasion in intestinal cells (*myfA* and *ail* genes), production of heat-stable enterotoxin (*ystB*), some proteases (*hreP*) and iron-chelating receptor (*fepA*), all of them related to growth and survival in the host during infection. The synthesis of insecticidal toxins (*tccC*) is also considered a virulence determinant in *Y. enterocolitica*

B1A. In this work, virulence-associated genes such as *ystB* (146 bp), *myfA* (272 bp), *hreP* (757 bp), *fepA* (438 bp) and *tccC* (1035 bp) were studied in 23 local *Y. enterocolitica* B1A strains of different origins (animal, food, environmental and human clinical samples) by PCR. Strains belonging to serotypes O:5 and O:7,8-8-8,19 (six isolates each), O:41,42-41,43 (four isolates), O:5-4,32-4,33 (three isolates), O:6,30-6,31 (two isolates), O:12,25-12,26 and NA (non-agglutinable/non-determined serotype) (one isolate each) were analyzed. DNA extraction was performed by the “boiling” technique and the amplification products were revealed by agarose gel electrophoresis. The frequency of detection of these genes in decreasing order was: *fepA* and *ystB* (22/23), *hreP* (21/23), *tccC* (3/23) and *myfA* (1/23). Regarding the relationship between genes and serotypes, *fepA*, *ystB* and *hreP* genes were demonstrated in strains of all serotypes, meanwhile *tccC* was observed in O:41,42-41,43 and O:7,8-8-8,19 strains, and *myfA* was only detected in O:7,8-8-8,19 strains. The serotype O:7,8-8-8,19 was associated to the presence of all genes. Regarding the relationship between genes and strain sources, *ystB*, *hreP* and *fepA* were demonstrated in chicken samples (3 isolates), porcine products (five isolates), ground meat (six isolates), human clinical samples (three isolates), and wild boar, hake fillet and wastewater (one isolate each). The *myfA* gene was observed in porcine skin (one isolate) and *tccC* was present in porcine skin (two isolates) and wild boar (one isolate). Interestingly, human samples belonged to serotypes O:5 (two isolates) and O:7,8-8-8,19 (one isolate) showed to be carriers of most of the studied genes, except *myfA* and *tccC*. Our results suggest the existence of alternative virulence mechanisms in *Y. enterocolitica* B1A and that the pathogenic potential of this biotype might be strain-dependent.

A134

SANITIZING TREATMENTS ON FRESH VEGETABLES ARTIFICIALLY CONTAMINATED WITH *YERSINIA ENTEROCOLITICA* AND SHIGA TOXIN-PRODUCING *ESCHERICHIA COLI*

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Fresh vegetables constitute essential ingredients of ready-to-eat salads in a healthy diet. From orchard to table, they may become contaminated by numerous microorganisms including *Yersinia enterocolitica* (Ye) and Shiga toxin-producing *Escherichia coli* (STEC), which are known foodborne enteropathogens responsible for moderate and severe human diseases, respectively. There is great interest in developing safe and efficient sanitizing treatments avoiding modifying the organoleptic characteristics of vegetables. In this study, 2% citric acid (CA), 200 ppm sodium hypochlorite (SH), commercial vinegar with 5% acidity (V) and 0.5 g/L acidified sodium chlorite (ASC) were assayed as sanitizers on pear tomatoes (*Lycopersicon esculentum* Mill), fresh lettuce leaves (*Lactuca sativa*) and strawberries (*Fragaria ananassa*) artificially contaminated with Ye or STEC. Fourteen units of each vegetable were used in each experiment. They were submerged in 1×10^8 CFU/mL suspensions of each pathogen by 2 min at 22 °C. Vegetables were removed and placed in sterile containers for 12 h at 4 °C to facilitate bacterial adhesion. Solutions of each sanitizer were prepared on the same day of each experiment. Samples were individually placed inside sterile plastic bags and agitated for 1 min in 50 mL of one of four sanitizing solutions. After washing, each sample was subjected to a 100 mL sterile distilled water rinse for 1 min to remove chemical residues and placed on sterile paper towels to dry. Vegetables were individually placed in new bags, added with 20 mL of sterile saline solution, and agitated for 1 min to detach bacteria. To perform bacterial counts after sanitizing, these suspensions were serially diluted, and volumes of each dilution were spread on Mac Conkey agar for Ye or Sorbitol Mac Conkey agar for STEC. In each group of vegetables, untreated/contaminated products were processed to determine the initial pathogen load on the surface. Bacterial counts were reported as log₁₀ CFU/vegetable. Bacterial reduction in each vegetable group by each sanitizing treatment was calculated as the difference between the count of untreated/contaminated group and the count of treated/contaminated group. After sanitizing, vegetables were stored at 4 °C for 7 days to assess changes in appearance. Lettuce leaves showed the highest initial loads of each pathogen as compared to other vegetables. Major bacterial reductions were produced by V (4.3 log in Ye and 4.5 log in STEC, both on lettuce) and ASC (3.76 log in Ye and 4.5 log in STEC, both on lettuce), followed by SH (2.28 log in Ye on tomatoes and 2.74 log in STEC on strawberries) and CA (2.83 log in Ye on tomatoes and 2.32 log in STEC on strawberries). After 7-day storage, tomatoes treated with any sanitizer and V- and ASC-treated remaining vegetables exhibited acceptable organoleptic characteristics; meanwhile, lettuce and strawberries treated with other sanitizers showed slight changes of color and texture and fungal growth. Vinegar and ASC are proposed as the best sanitizers to control Ye and STEC on fresh vegetables.

A135

SURVEILLANCE STUDY OF THE CARRIER STATE OF MULTIDRUG RESISTANT BACTERIA IN THE LABORATORY OF THE PROVINCIAL MATERNITY “DRA. TERESITA BAIGORRIA”

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The presence of colonized patients is one of the main routes for the spread of multidrug-resistant bacteria and its control is a clinical and public health priority. Surveillance studies are essential for the early detection of colonization by these bacteria. The objective of this study was to establish the carrier state of multidrug-resistant (MDR) bacteria of epidemiological importance and the antimicrobial resistance profile in patients of the Provincial Maternity. Between January 2018 and July 2019, 56 nasal and rectal swabs corresponding to patients of the intensive care unit (ICU) of the Provincial Maternity and patients with risk factors derived from other institutions, were processed. Different microbiological methods based on culture were adapted for the detection of the carrier state of multidrug-resistant bacteria. Nasal swabs were cultured on salad mannitol agar with antimicrobial disk cefoxitin (30

µg) for the detection of methicillin-resistant *Staphylococcus aureus* (MRSA). Rectal swabs were cultured on Eosin Methylene Blue agar (EMB) with antimicrobial disks strategically placed for detection of extended-spectrum β-lactamase (ESBL) producing *Enterobacteriaceae*, plasmid-mediated AmpC β-lactamase and carbapenemase-producing *Enterobacteriaceae* (CPE). For detection of glycopeptide resistant *Enterococcus sp.* (VRE), rectal swabs were cultured on bile esculin azide agar added with 6 µg/mL of vancomycin. From the analyzed data, microorganisms with some drug resistance mechanism were isolated in 23 samples (41%) as described: CPE OXA-48-like (1.78%), CPE KPC (1.78%), MDR *Acinetobacter sp.* (1.78%), ESBL producing *Enterobacteriaceae* (23,20%) and MRSA (3.57%). MDR or VRE. *Pseudomonas sp.* strains were not detected. In the population studied, these results show a low percentage of multidrug-resistant bacteria. This fact justifies the importance of local epidemiological surveillance studies that allow early detection of colonized patients and the prevention of the appearance of outbreaks, as well as the design of active control strategies.

A136

ISOLATION OF BACTERIOPHAGES AGAINST STRAINS OF SHIGA TOXIN-PRODUCING *ESCHERICHIA COLI* (STEC) IN MENDOZA

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Strain of *Escherichia coli* Shiga Toxin (STEC) are human pathogens whose most severe clinical manifestations are Hemolytic Uremic Syndrome (HUS) and/or Thrombotic Thrombocytopenic Purpura (PTT). In our region, it is important to control the spread of this pathogen. HUS is the most common cause of Acute Kidney/Renal Failure in Children (ARF) and it is the second cause of Chronic Kidney/Renal Failure (CRF). It is responsible for 20% of kidney transplants in children. In our country, there are about 400 cases annually. 2% will die and around 20% will be left with an IRC, in addition to other serious complications (M. Salud Nación 2018). The STECs that causes HUS are found mainly in the feces of cattle (the main reservoir), although it has also been found in other animals as sheep, goats, and rodents. In this way, milk, water, fruit, vegetables can be contaminated. It can also be transmitted from person to person (through contamination of the hands). Like any bacterial infection, its growing resistance to antibiotics is worrying. An alternative approach is the use of bacteriophages as a biocontrol tool for these pathogenic bacteria, present in meat. The aims are: Isolation of specific phages from STEC and evaluate the lytic activity of phages against STEC strains. Ground meat, sausages and wastewater were used to isolate phages. The technique used was to make an enrichment in the indicated culture medium and on the 2nd day, with the addition of CaCl₂, subsequent centrifugation and filtration. To isolate the phages, the Spot Test was performed. A total of 6 specific STEC phages were obtained, isolated from ground beef, sausages and wastewater. Phages were selected to form a therapeutic cocktail and lytic phages were chosen. STEC strains were provided by outbreaks of HUS from a pediatric hospital. Its effectiveness was evaluated with the reduction of viable cells (VC) that were spread on the surface of the meat, and after spreading the phages to be able to compare the VC reduction (STEC). Viable cell count was performed with and without phage application.

A137

OBTENTION OF INVERTASE FROM *SACCHAROMYCES CEREVISIAE* AND *ASPERGILLUS NIGER*

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Invertase is an enzyme that catalyzes the hydrolysis of substrates as sucrose to produce a 1:1 mixture of glucose and fructose sweeter than sucrose called invert sugar. It is mainly used in the food and pastry industry. *Saccharomyces* and *Aspergillus*, have developed to be good producers of this enzyme. The aim of this project is to optimize the culture parameters for the production of Invertase by *Saccharomyces* and *Aspergillus* and carry out enzymatic activity analyzes. The strains of *Aspergillus niger* (NRRL1419) and *Saccharomyces cerevisiae* (strain 2) were grown in Sabouraud liquid medium. The strains were suspended in acetate acetic buffer 0.1 M/NaCl 0.5 M pH 5. The effect of sonication on enzyme production was evaluated. Some samples were sonicated to break down cells and obtain greater enzyme production. Both strains were exposed to an induction process in sucrose solution 10 g/dm³. Finally, the glucose oxidase method was used to measure the activity of the enzyme. Protein concentration was determined by spectrophotometry (505 nm). The strains of *Saccharomyces cerevisiae* and *Aspergillus niger* that were subjected to an induction process with sucrose solution showed higher activity values than those strains that were suspended in extraction buffer, obtaining values of 283 U/min for *S. cerevisiae* strain 2 and 4802 U/min for *A. niger* NRRL 1419 using sucrose solution. All enzyme activity values for samples without sonication were higher than those samples with sonication. Values calculated using extraction buffer were 44 U/min for samples with sonication and 113 U/min without sonication for strain 2 of *S. cerevisiae*. For *A. niger* NRRL 1419 the values were 46 U/min with sonication and 246 U/min without sonication. It is an easily reproducible system, and high enzyme activity values were obtained. There were no inhibition problems during the process.

A138

DEFENSIVE RESPONSE OF SENESCENT CELLS TO STAPHYLOCOCCUS AUREUS INFECTION

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Senescence is defined as a state where somatic cells lose replicative capacity. This mechanism is triggered by different stimuli such as oxidative stress, telomere shortening, DNA damage, and chromatin abnormality. The characteristics of senescent cells include irreversible growth arrest, enlarged morphology, expression of cyclin-dependent kinase inhibitor (CDKI), the formation of senescence-associated heterochromatin foci, and senescence-associated secretory phenotype (SASP). These changes attempt to prevent damaged cells from proliferating and generating expansive damage. However, this mechanism can be counterproductive when activated in cells of the immune system as shown by a decrease in the effectiveness of the immune response against pathogens in older people. However, this mechanism can be counterproductive when activated in cells of the immune system as shown by a decrease in the effectiveness of the immune response against pathogens in older people. What could explain the increase in morbidity and mortality of infectious processes in relation to aging. Our work focuses to understand evaluating the response of senescent cells against *Staphylococcus aureus* infection. This pathogen is able to replicate intracellularly and to produce a wide spectrum of pathologies in humans ranging from skin infections to life-threatening invasive conditions. Previous results from our laboratory have demonstrated that autophagy plays an important role in the replicative cycle of *S. aureus*. In addition, it has been demonstrated that senescence blocks autophagy flux. Due to that, we propose to study the role of autophagy in the response of senescent cells against *S. aureus* infection. Taking advantage of the cellular model where senescence is induced by oxidative stress or transient p21 overexpression (CDKI) we were able to evaluate the autophagy response against *S. aureus* infection. Thus, we have observed that the activation of senescence modifies the intracellular distribution of the endocytosed microorganisms as well as the number of bacteria recovered by cell at 4 h post-infection. Furthermore, the use of autophagic inducers, as starvation or rapamycin, did not modify the effect observed under senescence activation on *S. aureus* replication.

A139

EVALUATION OF BIOMASS PRODUCTION IN WINTER PASTURES IN PLOTS SUBJECT TO CHEMICAL FERTILIZATION AND WITH CIANOBACTERIA IN THE PROVINCE OF SAN LUIS

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In the province of San Luis, there are different systems of livestock production, breeding (natural and megathermic pastureland), complete cycle and milking yards with forage resources such as winter and summer pastures, perennial meadows (“alfalfa”) and preserved forages. In the cald í forest phytogeographic area, one of the most marked production deficiencies is winter, especially if working with productive applications of high nutritional requirements such as milking yards or fattening of steers. The inclusion of a proportion of pastures, such as *Secale cereale* (rye) in the forage chains contributes to maintaining the productive level at that time of the year. To obtain high productions (MS/ha), it is necessary to consider the need for fertilization. In this sense, the hypothesis that biofertilization with cyanobacteria generates a forage production higher than the normal rye growth curve was raised. The objective of this work was to measure in a comparative way the production of forage biomass simulating grazing subjected to different types of fertilization. A randomized plot design with 4 replications and four treatments was arranged on the FICA (UNSL) site: Witness (T1), Cyanobacteria (T2), Cyanobacteria + SPS (T3) and Urea + SPS (T4), distributed completely at random. All the plots were farmed on March 10, 2019, applying irrigation with cyanobacteria in the corresponding plots on March 14 and planted (in its whole) on April 10 of the same year with a density of 30 kg/ha, together with the application of SPS in T3 • and T4 (100 kg x ha). T4 was fertilized with 50 kg x ha of Urea when the crop was in macollage (May, 16). The plot size was 1 m², and the cutting unit of ¼ m². Depending on the phenological state of the crop, 4 cuts (at the height of the fist) were made during the crop cycle; each sample was weighed to obtain the Green Weight (GW) value; then, they were taken to a stove with forced air at 60 °C for 48 hours. After this time, the Dry Weight (DW) value was taken, and the percentage of dry matter (% DM) was determined in order to estimate the yield per hectare (Kg/Ha). The cumulative production results (kg/ha/treatment) were: T1 = 3610, T2 = 3357.5, T3 = 3710 and T4 = 4300. A statistical analysis of the data was performed ($\alpha = 0.05$). It determined that there are no significant differences between the treatments. Thus, it is concluded that the farming of Rye in cultivated soils with low moisture content in the province of San Luis does not show a yield increase due to the application of urea or cyanobacteria to the soil. In this context, the use of biological fertilizers (cyanobacteria) and the use of chemical fertilizers (urea) generate the same effect on crop yield.

A140

UPDATE OF WOUND BOTULISM IN ARGENTINA (1992-2019)

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Botulism is a highly lethal neuroparalytic disease, caused by botulinum neurotoxins (BoNT) produced by *Clostridium botulinum* (*Cb*) types A to G and less frequently by other Clostridia, whose natural reservoir is mainly the soil (So). Serotypes A, B, E, and eventually F affect the human. Currently, two forms of the disease are recognized: (1) Intoxication, caused by preformed toxin: a) food botulism (classic and more recognized), b) accidental (in the laboratory), c) intentional (bioterrorism) and d) iatrogenic (derived from the pharmacological use of the toxin). (2) Toxiinfection, caused by colonization and subsequent production of toxin *in situ*, comprising: a) wound botulism (WB) and b) intestinal toxemia. The latter includes infant -the most frequent worldwide (in children under 12 months of age)- and adult (with previous intestinal pathologies) botulism. Although WB is unusual, in recent years the notification of cases has increased. BoNT produced in wounds contaminated with *Cb* spores travels systemically producing descending flaccid paralysis. It is associated with trauma, surgery, subcutaneous or intravenous drug use and sinusitis due to intranasal cocaine abuse. The clinical presentation is similar to food botulism, without digestive symptoms, with an incubation time of 5 to 15 days and with the history of a previous wound. The purpose of this work is to describe the characteristics of the cases reported in the country, their diagnosis, treatment, and evolution. The diagnosis was made by detection of BoNT in serum samples and wound debridement material, by the mouse bioassay method (gold standard) and investigation of *Cb* in wound material by inoculation in enrichment and suitable for toxin production media (Tarozzi Broth). They were incubated 4 days at 34 °C, in the centrifugation of the broth supernatant, BoNT presence was investigated with subsequent serotyping by specific neutralization tests. Between 1992 and 2019, 8 cases were reported and confirmed: 5 corresponded to the male sex, the age range was between 12 and 50 years, with a mortality of 37.5%. The types of injuries were: 1 surgical, 2 stabbing, 3 exposed fractures, and 2 of healed lesions. Serotype A was identified in 7 (seven) and B in the remaining 2 (two). The provinces with notified cases were: La Pampa, Entre Ríos, Santiago del Estero, Mendoza, Santa Fe, and the province of Buenos Aires. Since the pathology is not always related to the entrance door, it is necessary to strengthen surveillance for the detection and establishment of early treatment in cases of WB. Early clinical suspicion would allow to promote a favorable evolution and reduce mortality.

A141

ATMOSPHERIC BIOPARTICLES IN THE DRIEST PERIOD OF THE YEAR IN GENERAL ALVEAR, MENDOZA

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The aerobiological quality of a region results of relevance in health and agricultural matters. The knowledge of the aerial pollen concentration in pollinosis periods or the aerial fungic spore concentration in agricultural production seasons provides paramount information. This is especially important in General Alvear, a city commonly affected by recurrent health problems in people exposed to allergens and diseases in plants of agricultural production. This preliminary study of aerial bioparticles was implemented in the driest period of the year (July and August 2019). The aerial bioparticles were sampled with a Lanzoni volumetric pollen trap (VPPS 2000) and the samples were read with an optical microscope at 400X. The peaks of pollen abundance registered were: Cupressaceae (31.86 grain/m³ air), Oleaceae (16.2 grain/m³ air), Rosaceae (5.13 grain/m³ air), and Ulmaceae (3.6 grain/m³ air). In lower proportion, Chenopodiaceae-Amaranthaceae, Asteraceae, Fabaceae, and other pollen types were also found. With respect to spores, the major types were: *Aspergillus-Penicillium* (20.52 spores/m³ air), *Periconia* sp. (17.01 spores/m³ air), *Alternaria* sp. (12.6 spores/m³ air), *Torula* sp. (10.51 spores/m³ air), *Paraphaeosphaeria* (7.51 spores/m³ air), and *Didymosphaeria* sp. (7.2 spores/m³ air). The type and proportion of pollens and spores registered were similar to those reported in the same period from other provinces of Argentina. However, the Rosaceae type had a greater representation than others reflecting the abundance of fruit trees and ornamental plants of this family in the city. Among pollen types, highly allergenic species were registered. These biological particles when combined with environmental dryness, which favors their persistence in the air, aggravate the pollinosis symptoms. The present study has as a priority to elaborate an aerobiological calendar of fungi and spores for future applications in health and agricultural production.

BIOCHEMISTRY, PHYSIOLOGY, PATHOLOGY AND PLANT PRODUCTION

A142

WEEDS PRESENT IN DEFORESTED AREAS OF LIVESTOCK USE IN THE DISTRICT OF “CALDÉN” IN SAN LUIS PROVINCE (ARGENTINA)

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The study was carried out in the Southeast Central region of the province of San Luis, Caldín district (*Prosopis caldenia*). This area had an important productive transformation which increased deforested areas as the implantation of annual agricultural crops and forage production needed. Thus, study of weeds gained progressively great importance in the region due to the change in land use. Previous studies in this region have concluded that agriculture produced an increase in introduced herbaceous species but without significant differences neither in richness nor diversity. Other authors concluded that an increase in introduced species occurred at the expense of native species, which implies a change in the floristic composition of such agroecosystems. At present, there are no previous studies specifically on weed species, native or exotic, that occur in dismantled sites, whether under agricultural or forage use. The aim of this work was to evaluate the presence of weed species in deforested areas and cultivated with maize for livestock use. Sampling was carried out from the spring of 2018 to the spring of 2019 in recent dismantled areas under livestock use. Weed species were sampled at different stages, collected, preserved, and deposited in the “Departamento de Ciencias Agropecuarias” herbarium (VMA) and identified by the traditional botanical methods. Richness and frequency in deforested areas cultivated with maize for livestock use were calculated. The families best represented in the 2018 spring survey were the Asteraceae (55%), Poaceae (36%), and the Brassicaceae (5%). The most frequent species was *Cynodon hirsutus* (Poaceae) as well as species of *Stipeae* tribe (Poaceae) (23%). In general, the summer spring emergency species were found in the seedling stage, with few representatives each of the remaining 4 families. While, in autumn 2019, the best-represented families were Asteraceae (38%), Poaceae (15%), and Brassicaceae (8%), with less frequency than another five botanical families. The autumn–winter emergency species were sampled mainly to the seedling stage. The most frequent were *Descurainia erodiifolia* (Brassicaceae), *Gamochaeta filaginea* and *Gnaphalium gaudichaudianum* (Asteraceae). The climatic conditions within this period with late frosts and little rainfall influenced the livestock management, therefore low supply of fodder was determining the occurrence of species in the studied plots. Within the framework of the Stimulus to Scientific Encouragement Vocations Scholarship EVC-CIN 2018 & CyT UNSL P-140916

A143

MORPHOGENESIS AND STRUCTURE OF *THINOPYRUM PONTICUM* “AGROPIRO ALARGADO”

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Thinopyrum ponticum (agropiro alargado) is a temperate forage specie suitable for transforming unproductive areas and rehabilitating its capacity for livestock use. It has a high level of adaptation and can be developed in areas with limiting environmental conditions –dry, saline or with low temperatures– where animal production is affected by the decrease in the quantity and quality of the forage. The objective was to describe the morphogenetic and structural variables at tiller level, of *T. ponticum* in the autumn and winter season, during the growth period. The pasture is established on a depressed and slightly saline area into an experimental field of the agronomical department of UNSL, located in Villa Mercedes (S.L.). In mid-February 2019 a cleaning cut was made, and at the beginning of regrowth 15 random tillers were subsequently identified. Once a week the appearance and leaf senescence, green leaf length (LFV), tiller and plant phenological stages were recorded. With the collected data, morphogenetic and structural variables were determined. Phyllochron (Fc) expressed in thermal time (°C), mean leaf lifespan (VMF: °C), leaf appearance rate (TAH: leaves. °C.day⁻¹) and foliar elongation rate (TEF: cm.day⁻¹) conform the morphogenetic variables. The total number of leaves at tiller (N°H), Fc, VMF (Fc * N°H), TAH (1/Fc) and TEF (daily increase in LFV, from the appearance to the maximum foliar expansion) was determined. The foliar accumulation and the TEF were related to the average temperature and determined a base growth temperature of 4 °C. Fc and VMF were calculated between the 3rd and 7th sheet. Of the total of identified tillers, only 8 were able to be evaluated, which present: N°H: 8, VMF 519 ± 108 °C.day⁻¹; Fc: 260 ± 54 °C.day⁻¹, TAH: 0.004 ± 0.001 leaves. °C.day⁻¹. The variable TEF varied between 0.37-0.56 cm.day⁻¹ in the first three leaves and among 0.14-0.43 cm .day⁻¹ in the last three. All the tillers reached VMF in a vegetative state. The maximum LFV was 16 to 20 cm in the first three sheets and 8 to 9.5 cm in the last three. Agropiro alargado has significant foliar accumulation but with a low number of green leaves on tiller (1.7 ± 0.3), limited foliar height, and slow leaf renewal during the autumn-winter season. These aspects should be considered before the possibility of its use as fodder in the cold season.

A144

EFFECT OF CO₂ AVAILABILITY ON PHOTOSYNTHESIS S SPEED OF *FICUS CARICA* LEAVES AND DISTRIBUTION OF CHLOROPHYLL IN BIPHASIC SYSTEMS

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Photosynthesis is a process whereby organisms with chlorophyll, such as green plants, algae, and some bacteria, capture energy in the form of light and transform it into chemical energy. In this work, we studied the effect of CO₂ availability on photosynthetic speed, in *Ficus carica* leaves. The chlorophyll content of hydroalcoholic extracts of the leaves was determined spectrophotometrically and the distribution in two-phase systems composed of ammonium sulfate and ethanol was analyzed. The floating disc test was used to explore the process of photosynthesis in the leaves. Foliar disks generally float due to the many

intercellular spaces used for gas exchange. When the air spaces are infiltrated with solution, the overall density of the blade disc increases, which causes the discs to sink. The infiltration solution includes a small amount of sodium bicarbonate. The bicarbonate ion serves as the carbon source for photosynthesis. By providing the necessary components for photosynthesis (light, CO₂, and H₂O), oxygen will be produced in the leaf. As photosynthesis is carried out, oxygen is released inside the leaf that changes buoyancy and causes discs to rise. As cellular respiration takes place and also consumes oxygen, the rate at which the discs rise is an indirect measure of the net rate of photosynthesis. Breathing, which uses the oxygen produced by photosynthesis, is also observed in the foliar disk test. Some oxygen will be used in the leaf breathing process. Leaf discs float because the net result is that photosynthesis produces more oxygen than is used in breathing. The rate of photosynthesis recorded for the 1% sodium bicarbonate infiltration solution was 0.12 min⁻¹, being slightly lower for 0.5% sodium bicarbonate solutions. The total chlorophyll content in the extracts was 24 mg/L, corresponding, 9.8 mg/L to chlorophyll a and 14.2 mg/L to chlorophyll b. The distribution constant in the biphasic systems formed by 24% ammonium sulfate and 24% hydroalcoholic extract was 2.8 with a yield of 73%. The importance of these biphasic systems lies in the possibility of using the plant tissue without the prior preparation of the extract, recovering in this case, the greater amount of chlorophyll in the upper part, the plant residues being retained at the interface.

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ABSENCE OF *DIATRAEA SACCHARALIS* IN CORN, IN LATE SOWING, IN VILLA MERCEDES (SAN LUIS)

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Diatraea saccharalis (Fabr. 1798) is the species with the greatest economic impact in the Pampas region of Argentina, where it causes an average annual loss of 170 million dollars. During its larval stage, it causes galleries on the stem of the corn plant severely damaging it. Depending on the region, the "stem borer" has between three and four annual generations. Populations of this plague increase from sowing to corn harvest. The first generation from wintering larvae emerges in October and November infesting native and cultivated grasses. The base temperature (T_b) for the larval phase is 7.3 °C and 9.4 °C for the total cycle. In the late 90s, biotechnology created Bt corn, this genetic event allows the crop to resist the attack of *D. saccharalis*. The massive adoption of this technology produced the population level of adults to decrease significantly in the subsequent 10 years. The objective of this work was to determine the incidence and severity of *D. sacharallis* in transgenic and traditional hybrid corn in late sowing. The field test was in the experimental field of the Department of Agricultural Sciences (FICA-UNSL), located on Route Prov. No. 55 Extrem North, during the 2014, 2015, and 2016 productive campaigns. In a randomized design of four treatments (tree transgenic and one traditional hybrid) and three repetitions, the plants of the three central furrows were sampled from the emergence to physiological maturity. Two parameters were evaluated: (a) number of ovipostures in each plant, and (b) incidence: number of plants with exit hole, and severity: number of internodes affected and number of galleries/internode. For the period analyzed, there was no presence of the species throughout the crop cycle. These results could be due, on the one hand, to the conditions of severe winters in the study area, where the average temperature for the months of June, July and August (2014: -0.4 °C, 2015: 2.06 °C and 2016: 0.17 °C) is below the T_b; and on the other hand, to the significant increase in the area with GM crops. It is necessary to continue with the evaluations, being that the low prevalence of the species induces to rethink the management strategies of the corn crop.

A146

FRECUENCY AND COVERAGE OF WEEDS IN CORN CROP IN LATE SOWING IN THE CENTRAL EAST AREA OF SAN LUIS PROVINCE

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The population dynamics of weeds within the corn crop are modified with the sowing time. The late sowing in the central east area of San Luis province allows water recharge in the soil profile during spring. So the crop reduces, on the one hand, water stress, and on the other hand, thermal stress due to high temperatures in December and January; this allows you to arrive in adequate conditions to bloom. The sustainability of agriculture in the central semi-arid region of Argentina is questioned in many situations; on the one hand, crop yield instability; and on the other hand, the presence of weeds, which becomes a conditioning factor that causes production losses of up to 80%. In this region, weed control through phytosanitary products is restricted to cost if we compare it with the greatest agricultural potential region. Furthermore, in most cases, it is routinely used and without comprehensive advice that takes into account their ecology. Accordingly, the weed control requires prior knowledge of the particularities in the species and their interactions with crop management. The objective of this work was to obtain preliminary data on the frequency and coverage of weeds in hybrid corn in late sowing in the central east area of San Luis province. The field test was in the experimental field of the Department of Agricultural Sciences (FICA-UNSL), located on Route Prov. No. 55 Extrem North. The Dekalb 7010 RR2 hybrid was used in the field test and the sowing dates were during the first half of December of the years 2016, 2017, and 2018. The "Interception Lines Method" (Mostacedo y Frediricksen, 2000) was used for sampling. Samples were taken along a 10-meter transect in the three central grooves and 2.5 m were left unsampled to reduce experimental error. First weeds were identified in each transect, then quantify the coverage in centimeters of each identified species. The evaluation was made during two phenological stages of the hybrid: VT (pollination start) and R5 (grain filling). The total coverage between the rows was 120 m. The number of species (2016:

9, 2017: 15 and 2018: 10) was not constant between the different years and it was also variable within each year. Only *Portulaca oleracea* (274 cm), *Digitaria sanguinalis* (117 cm), *Cynodon dactylon* (51 cm), and *Chenopodium album* (47 cm), remained constant. In most of the samples, *Portulaca oleracea* had greater coverage. It is necessary to continue with the evaluations to establish distribution patterns that allow us to know more about the population dynamics of weeds, and thus provide technical tools for the management of corn in the central east region of San Luis province.

A147

VIABILITY EVALUATION AND GERMINATION TREATMENTS OF THREE SPECIES OF WEEDS NATIVE TO “DUNES WITH PASTURES AND CHAÑAR” REGION OF SAN LUIS, ARGENTINA

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The phytogeographic region “Dunes with pastures and Chañar” covers an area of 2,000,000 ha in the central south of the province of San Luis, Argentina. In this region are important patches invaded by perennial *Poaceae* species not preferred by cattle. Seasonal growth of which accumulates combustible material susceptible to disturbances caused by accidental or natural fires: *Elionurus muticus* of summer growth, elongates in spring and fruits in early summer. *Nassella tenuissima* a caespitose herb grows in winter, sprouts in autumn, fruits in November and December-January. *Jarava ichu* also grows in winter and regrows in autumn, flowering and fruiting in August-September to late spring and early summer. These herbs appear in poor or fair condition sandy grasslands, dominated by species of little or no forage value. Effective control of undesirable species requires a deep knowledge of them, for this reason, the aim of this study was to evaluate viability and germination treatments of *Nassella tenuissima*, *Jarava ichu* and *Elionurus muticus* species. For this purpose, seeds of the three species were collected in the experimental field of the “Facultad de Ingeniería y Ciencias Agropecuarias (UNSL)” between the months of October to December 2018 and were stored in paper bags. Studies were conducted in the laboratories of Plant Biotechnology (CIATEJ, Guadalajara, Mexico). Seeds were subjected to a viability test using the tetrazolium technique. Germination of the seeds treated with Gibberellic Acid (GA3) was evaluated in the three species. Moreover, *Nassella tenuissima* and *Jarava ichu* seeds were subjected to Hydrogen Peroxide; on the other hand, these species were exposed to 13 °C for 24, 30, and 72 h with GA3. Water was the reference sample for all treatments. Seed vigor (tetrazolium test) showed a high value for all species: *Elionurus muticus* 90%, *Nassella tenuissima* 91%, *Jarava ichu* 94%. All species showed the highest germination percentage with GA3 treatment. Hydrogen Peroxide did not improve the germination in both *Nassella tenuissima* and *Jarava ichu* seeds. *Jarava ichu* and *Nassella tenuissima* showed a germination percentage of 90% and 75%, respectively, after treatment with GA3 and 72 h at 13 °C. The seeds showed a low germination percentage in conditions of humidity and temperature according to each species. This result would indicate that sexual reproduction in these species is limited. Additionally, seed germination of winter species was favored by cooling treatments probably because of some dormancy mechanism related to this factor.

A148

EVALUATION OF PHYTOSANITARIES BASED ON PHOSPHITES FOR THE CONTROL OF ROYA (*PUCCINIA* SPP.) IN AVENA AND WHEAT IN VILLA MERCEDES - SAN LUIS - ARGENTINA

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Oats (*Avena sativa*) and wheat (*Triticum aestivum*) are prominent cereals for seed production in the province of San Luis, Argentina, where the progress of agriculture has been important in recent years. These crops are affected epidemically by Black stem rust (*Puccinia graminis*) and yellow rust (*Puccinia striiformis*), which produce significant yield losses as well as forage quality. So far, the most used control methodology is the application in total coverage of fungicidal products, mainly those belonging to the triazoles group and the estrobilurinas group. In recent years a new type of phytosanitary products are being launched, phosphites, derivatives of phosphorous acid have new mechanisms of action and movement in the plant, these can help or replace the use of conventional fungicides. These products exacerbate crop defenses by improving their nutrition and stimulate the production of phytoalexins, important organic substances with which plants resist the attack of pathogens, mainly fungi. The study was carried out in order to determine the effect of Cu²⁺ phosphite and K⁺ phosphite on the attack of pathogens (Orange Rust and Yellow Rust) and on crop yields during the 2018/2019 campaign. The trial was sown in mid-June on 12 m² plots with 3 treatments and a witness with 3 repetitions for each. The follow-up of the crops was weekly by visual observation and data collection in registration forms. The scale used to measure severity was that of "Peterson, 1948". The application of the phytosanitary ware was carried out in the stage of tillering stage and in doses of label. The grain harvest was carried out on December 10th, obtaining as a final result for both crops: Oats: excellent response to phosphite of K⁺ (Rend 2983.1 kg/ha), very good to phosphite of Cu²⁺ (Rend 3178.1 kg/ha), regulate conventional fungicide (Rend 2926.2 kg/ha) in relation to the witness that was severely affected (Rend 2259.5 kg/ha). Wheat: excellent response to phosphite of K⁺ (Rend 2188.6 kg/ha), very good to phosphite of Cu²⁺ (Rend 1912 kg/ha), regulate the

conventional fungicide (Rend 2222.8 kg/ha) and control severely affected (Rend 1807.5 kg/ha). In all cases, the yields accompanied the sanitary conditions in which each treatment was developed.

A149

EVALUATION OF PHYTOSANITARIES BASED ON PHOSPHITES FOR THE CONTROL OF ROYA (*PUCCINIA* SP.) IN CENTENO AND TRITICALE IN VILLA MERCEDES - SAN LUIS - ARGENTINA

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In the province of San Luis, Argentina, the crops of rye (*Secale cereale*) and triticale (x *Triticosecale* Wittmack) are excellent alternatives for the production of fodder and grain for animal feed and the sale of seed respectively. Its good adaptation to semi-dry conditions and low temperatures means that year after year they are chosen by a large number of producers with different purposes, among which are also the realization of cover crops in lots with erosion risks. These crops are affected epidemically by stem rust (*Puccinia graminis*) and yellow rust (*Puccinia striiformis*), which produce significant losses in forage quality, the incidence of leaf area (IAF), and final grain yield. So far, the most used control methodology is the application in total coverage of fungicidal products, mainly those belonging to the triazoles group and the estrobilurinas group. Currently, new products for disease control are incurring in the market, systemic translocation and phosphoric acid derivatives are called phosphites, which present new modes of action, which can help or replace the use of conventional fungicides. These products exacerbate crop defenses by improving their nutrition because they are formulated with different trace elements and stimulate the production of phytoalexins, important organic substances with which plants resist the attack of pathogens, mainly fungi. The study was carried out in order to observe the effect of Cu²⁺ phosphite and K⁺ phosphite against the attack of Stem rust and Yellow rust, in addition to its effect on the forage of crops during the 2018/2019 campaign. The trial was planted on June 15th in 12m² plots with 3 treatments and a witness, with 3 repetitions for each. The follow-up was weekly by visual observation and data collection, the scale used to measure severity was that of "Peterson, 1948". The application of phytosanitary products was carried out during the macollage stage and in doses according to the label. The results showed for both crops and as regards fodder it refers: very good response to K⁺ phosphite and Cu²⁺ phosphite, regular for conventional fungicide, while the control was severely affected by both Royas.

A150

MULTIVARIATE ANALYSIS OF THE RESPONSE OF COTTON LAVENDER (*SANTOLINA CHAMAECYPARISSUS*) TO LEAD STRESS

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Cotton lavender is a shrubby aromatic plant with several uses in medical and pesticide issues. The objective of the present work was to determine the physiological response of cotton lavender plants (*Santolina chamaecyparissus*) against lead stress through chemometric tools assessing tolerance for Pb stress. Seedlings were arranged in pots of 1 kg with sand:vermiculite (3:1) as substrate and grown in a greenhouse. Macro and micronutrients were applied with 1 g of Nitrofull fertilizer every 30 days until the end of the experiment. 32 days after the experiment started (AES), plants were watered with PbCl₂ solution until they reached concentrations in substrate 0, 250, 500, 1000, and 2000 ppm of Pb. 25 weeks AES, plant growth parameter was measured with image analysis software. At week 27 AES, samples were collected to determine lipid peroxidation (LP), catalase (CAT), ascorbate peroxidase (APX), and guaiacol peroxidase (GOPX) activity. The data were analyzed by uni and multivariate statistics. Score plot defined two groups in which concentrations up to 1000 ppm of Pb performed one group, and the other grouped 2000 ppm Pb treatment. The loading plot indicated that such difference is due to the influence of LP, CAT, APX, and GOPX, which they increase at 2000 ppm Pb in accordance with univariate statistics. Results indicate that treatments up to 1000 ppm Pb have a slightly-moderate physiological response with respect to the 0 ppm Pb, while at 2000 ppm biomass production is inhibited, and lipid peroxidation and antioxidant enzyme activity increase significantly. In conclusion, the results indicate that cotton lavender has high tolerance in conditions up to 1000 ppm of Pb, presenting effects and responses to oxidative stress at 2000 ppm Pb. Thus, this specie could be potentially useful as a phytoremediator in contaminated areas with Pb.

A151

VALUATION OF THE GRAIN YIELD OF UNPUBLISHED AND COMMERCIAL GERMPLOSM OF *TRITICUM AESTIVUM* IN THE EEA- SAN LUIS – INTA

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The germplasm used corresponds to the cultivars of the Wheat-INTA Territorial Trials Network. The objective of the work was to evaluate the grain yield of unpublished and commercial cultivars in the 2017 and 2018 cycle. Wheat cultivation requires during its cycle (June–December), between 350 and 550 mm. Both water availability and fertility are related to yields. Management was with chemical fallow (glyphosate and metsulfuran), during March–April to accumulate autumn rains. The critical period of wheat cultivation is the period of stem extension with a water demand (3–4 mm/day) and the needs are maximum in the filling of grains (5–6 mm/day). In this period the grain yield is defined. It was fertilized at the time of sowing with diamonic phosphate: (18% of N; 46% of P₂O₅), during tillering with urea 100 kg/ha equivalent to 46 kgN/ha and was fertilized in flag leaf with 46 kgN/ha. In a random block design was analyzed with INFOSTAT 2018 with the DGR Test. The 2017 cycle was sown in June and harvested in the second week of December. Thirty long-cycle cultivars were evaluated, resulting in an overall average of 2147 kg/ha of grain with a minimum of 800 and a maximum of 4080 kg/ha. These cultivars stood out with significant difference: Klein serpiente, Baguette 801, Aca 360, Sy 110, Biointa 2006, Ms inta 415. While the short cycles, with 25 participating cultivars, had an average yield of 2012 kg/ha with a maximum of 3840 and a minimum of 240 kg/ha. The cultivars with the highest yield were: Sy 300, Sy330, Sy200, Biointa1006, Aca910, Ms INTA615, Ms INTA815, Ms INTA816. The cumulative rainfall was 414 mm from March to December. In 2018, short cycles due to drought dried up, while 30 long-cycle cultivars averaged 1498 kg/ha of grain with a minimum yield range of 200.00 and a maximum 3100 kg/ha. These cultivars were highlighted sy 211, sy 120, aca 914, msINTA 116, buck meteor, ms inta 817, ms inta 415. The cumulative rainfall was 250 mm from March to December. It is acceptable to conclude that during the 2017 cycle, the availability of water was adequate with a good average of yields in short cycles and in long cycles. While in cycle 2018 the low rainfall did not allow to produce short cycles and in long cycles the yields were low, however, there are cultivars that with low rainfall produce average yields of 2500 kg/ha.

A152

EVALUATION OF OPTIMUM HARVEST TIME AND YIELD OF *ERAGROSTIS CURVULA*, CV. ERMELO (LOVEGRASS) SEED

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Lovegrass (*Eragrostis curvula*) is a perennial forage native to Africa, the cultivar Ermelo was introduced to Argentina in the 1960s, with better productivity and quality than the cultivar Tanganika, widely distributed in the country for high tolerance to drought and winter cold (–17 °C). It reproduces by apomixis, of the type mitotic diplosporia pseudogamy. It is a form of asexual reproduction and produces seed without pollen involvement and is genetically identical to the mother plant. Plants 50–90 cm tall, with large pyramidal panicles with olive-green spikes to dark greens. The objective of the present work was to evaluate the evolution of seed yield and optimum harvest time. The pasture was managed during July 2018, the foliar remnant was cleaned with weed remover and in mid-August, it was fertilized with 100 kg/ha of urea equivalent to 46 kg/ha, in a lot of the EEA SL-INTA. Two samples were taken per week with 10 repetitions per date, with an experimental unit of one-quarter of a square meter. The panicles were collected with the floral stem approximately 40 cm long. Sampling lasted from 01/11 to 30/12/2018. The collected panicles were stored in a cool, dry place. They were then hand-threshed during February and March 2019. The number of panicles per square meter was heterogeneous with an average of 160, with a minimum of 50 and a maximum of 300. The yield of cariopsis or seed per panicle was very stable and with significant differences per week of cumulative sampling. The first two weeks were 0 mg/panicle, and in the magnifying glass, only very small rudiments of cariopsis were observed and in few cases. The seventh week was the highest with 118 mg/panicle, while the eighth week started the natural seed spread by significantly lowering the weight, reaching 0 mg/panicle in the last sampling. From the sixth week, a change of color from green to yellow is observed at the base of the stem from the bottom to the top. The maximum yield was 200 mg/panicle with significant differences in the seventh week of sampling. It dropped significantly in the eighth week by dissemination. The first and second weeks were almost 0 mg/panicle. In other drought years, it has been observed that the highest yield was in the fourth week. It is concluded that the optimum harvest time and the highest yield was in the seventh week of sampling.

A153

PREVALENCE OF POTENTIALLY PATHOGENS MICOTOXIN PRODUCERS IN CORN SEEDS, IN THE CENTRAL EAST REGION OF THE PROVINCE OF SAN LUIS

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Corn is the most important cereal in the country and its grain and fodder play an important role in the production systems of San Luis. Seed quality is also crucial as a raw material in many food industries. Where the quality is an important component of the consumer's health, some phytopathogens can produce mycotoxins and these can be acquired by ingestion of products of plant or animal origin and exposure to environments or dust particles that contain toxins. These fungi are always present, but they generate toxins when favorable environmental conditions persist, such as high temperature and high relative humidity. It is important to assess the health

of the seed in order to determine the need to apply prevention and control measures, therefore the epidemiological implications of the presence of certain pathogens in grain production must be known. With this objective, grains were collected from 25 monitoring sites located in the central-eastern region of the province of San Luis, with different agroecological conditions. Of each of them, 150 grains were analyzed in DCA of three repercussions. Health tests were performed according to ISTA and phytopathological standards of current use. Colonies were quantified at 3, 5, and 7 days after planting, an ANOVA and Tukey test (1%) was performed. The fungal load per treatment ranged between 7 and 47% (no significant differences were observed), but for the species present *Aspergillus flavus* (AF), *A. niger* (AN), *Fusarium verticilloide* (FV), *Rhizopus microsporus* (RM), *Penicillium variabilis* (PV) and *P. paxili* (PP), potentially mycotoxin producers (with significant differences between FV (C), AF-RN (B) and the remaining (A). Given the importance of mycotoxicosis, alternative post-harvest strategies should be considered for the management of these pathogens.

A154

EFFECT OF SANITARY MANAGEMENT TREATMENTS ON THE FUNGAL AMOUNT OF CORN GRAINS IN VILLA MERCEDES, – SAN LUIS – ARGENTINA

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The increase of the world population caused a strong expansion of the borders and intensification of the use of arable land. This led to an increase in health problems and the need to generate sustainable management alternatives. Fungi can be associated with cereal seeds and in some cases, produce mycotoxins that can generate mycotoxicosis in animals and humans. Therefore, the need to know the fungal load in corn grains and determine the incidence of mycotoxin-producing pathogens under different sanitary treatments. 150 grains were analyzed per plots with treatments: T0: control, T1: fungicide (F) + insecticide (I), T2: herbicide (H) + I, T3: H + F and T4: H + F + I and three repetitions. Seed health tests were performed in 2% APG medium. The seeds were disinfected with 70% alcohol, 2% NaClO and sterile distilled water. The identification was by observation of reproductive structures. Colonies were quantified at 3, 5, and 7 days after planting; ANOVA and Tukey test (1%) were performed. The fungal load per treatment ranged between 35 and 42 % (no significant differences were observed). Herein shows that early fungicide applications for other problems do not cover advanced phenological stages. It was found: *Aspergillus flavus*, *A. niger*; *Fusarium verticilloide*, *Rhizopus microsporus*, *Penicillium variabilis*, and *P. paxili*, potentially mycotoxin producers (with significant differences between *Fusarium verticilloides* (C), *Aspergillus flavus* (B) and the rest (A). Given the importance of mycotoxicosis should be considered alternative strategies post-harvest.

A155

EVALUATION OF THE DORMITION BREAK IN GATTON PANIC (*PANICUM MAXIMUM*) WITH DIFFERENT METHODS OF CHEMICAL SCARIFICATION

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Gatton Panic (*Panicum maximum*) is a perennial tropical megathermal forage, is native from Africa and Asia with a wide adaptation and distribution in the Argentinean phytogeographic region of the arid Chaco (Salta, Jujuy, Tucumán, Santiago del Estero, Chaco, Formosa, and North of Córdoba). During the production of seeds, which coincides with times of scarce rainwater or drought, occurs a greater accumulation of abscisic acid (AA) in antecios (lemma and pãlea), that phytohormone induces the dormition of cariopse. The aim of the work was to break the dormition with chemical scarification and improve emergency uniformity. The determination of viability by tetrazolium gave a potential total of 30 % of viable cariopses. Two trials were carried out, in the first one sulphuric acid (SA) was used in two immersion times of the seed: 10 and 20 minutes, 4 concentrations and four repetitions. The treatments were: T0 (control-water); T1: 10%; T2: 20%; T3: 30%. At the end of the immersion times, seeds were washed with water to stop the effect. They were placed to germinate in camera with 8 h of light at 30 °C and 16 h of darkness at 20 °C, with readings every 4 days for 21 days. The second test was performed with sodium hydroxide (SH), in 4 immersion times: 10, 20, 30, 40 min and 6 concentrations: T0: water; T1: 10%; T2: 20%; T3: 30%; T4: 40%; T5: 50%. In all cases, the temperature did not exceed 10 °C during the immersion. The best treatments for rupture of the dormition with sulphuric acid were T0, with 10 or 20 min of immersion. In other treatments, at different times and concentrations decreased germination affecting its viability. The best results with SH were in T0 at all times of immersion, with values around 28% of germination. Germination decreased when concentration and time increased. It is concluded that washing with water for 10 to 40 min at low temperatures and subsequent drying breaks dormition and improves the germination percentage and emergency uniformity.

A156

GERMINATION OF *ERAGROSTIS CURVULA* (LOVEGRASS) CV. ERMELO ON THREE HARVEST DATES WITH DIFFERENT WATER POTENTIALS WITH POLYETHYLENE GLYCOL -PEG6000

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Eragrostis curvula (lovegrass) is a tropical perennial forage native to East Africa. The introduction of lovegrass in the semi-arid region of Argentina was in the 1950s, which revolutionized livestock farming in this area since this forage has a high tolerance to drought and winter cold. It is a polyploid species, mainly reproduced by forced apomixis of the pseudogamma mitotic diplosporia type. Apomixis is the development of the embryonic sac of the megasporocyte that by mitosis forms the tetranucleated embryo sac, with an egg cell, two synergies and a polar nucleus. Reproduction is asexual and seeds are genetically identical to the mother plant. This species adapted quickly and has a wide distribution in Argentina. The aim was to determine the effect of different water potentials with PEG6000, on three harvest dates in a lot of EEASL-INTA. The treatments were T0: water, T1: -0.5 MPa, T2: -1.0 MPa, T3: -1.5 MPa, at 25 °C with 8 h of light and 15 °C with 16 h of darkness. The experimental units were Petri dishes with 20 seeds with 4 repetitions, with PEG-6000 solution as substrate, in order to simulate variations in soil moisture availability. Germination percentage (GP) was controlled every 4 days until 21 days. Statistical analysis was performed with the Duncan Multiples Range Test. INFOSTAT 2018. At 21 days of germination, in the first harvest date (11/11/2018) at time T0 there was 30 % GP, in T1 there was 10 % GP and in the rest 0 % GP. On the second date (30/11/18), it was found in T0: 73.3 %, in T1: 46.7 % and in T2: 3.3 %. In the third harvest date (28/12/18) there was in T0: 50 %, T1: 25 %, T2 and T3: 6.7 % and 3%, respectively. This last fact is very important since in the same seed production lot, depending on the harvest date, we can find germination percentages with high water potentials, which is directly related to low soil moisture for arid areas. This suggests that this technique can be very useful to detect in different lots, seeds with different germination percentages with -1 to -1.5 Mpa. It is concluded that, depending on the harvest date, in the same lot the germination percentages will be different in response to different water potentials with PEG600.

A157

CONTAMINATION WITH CADMIUM AND ZINC, STUDY OF CYTOTOXICITY AND GENOTOXICITY IN *ALLIUM CEPA*

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There are problems associated with the contamination of soils by heavy metals, generating competition between contaminating metals and essential metals. Zinc (Zn) is an essential element, however, in excess; it causes alterations in important processes for the development of the organism. Cadmium (Cd) is a heavy metal with no biological functions, which at low concentrations generates damage at the cellular and molecular level. They have the same load as the essential metals; the plant absorbs them using the same transporters (IRT1-ZIP) for Cd and Zn, getting to interfere with the entry, transport and use of the microelement. The toxicity symptoms of these ions are common for several plant species such as *Glycine max*, *Allium cepa*, among others. The phenological aspect observed is: reduction of biomass, decrease in elongation and darkening of the main roots, chlorosis and leaf necrosis, also causing changes in oxidative stress parameters. The objective of this work was to evaluate the cytotoxic and genotoxic effect caused by the interaction of Cd and Zn in the apical meristem of *A. cepa*. For this test, the cytotoxic and genotoxic effects of both ions were examined through the mitotic index (MI), chromosomal aberrations (CA), and nuclear aberrations (NA). Seedlings after 2 days of development and adaptation under hydroponic conditions, with Hoagland nutrient solution, were subjected for 3 days, to different concentrations of ZnCl₂ (0, 0.6, and 4.8 mM) and a constant concentration of CdCl₂ (40 µM). The results showed a significant decrease in MI in all treatments with respect to the control ($P < 0.001$). In the genotoxicity analysis, NA were observed, to a greater extent in the last two treatments with Zn [4.8] s/ Cd and Zn [4.8] c/ Cd ($P < 0.001$). With respect to CA, C-mitosis and increased chromosomal condensation showed a significant increase in treatments s/ Zn c/ Cd, Zn [0.6] s/ Cd and c/ Cd and Zn [4.8] s/ Zn and c/ Cd with respect to the control ($P < 0.001$). Based on these results, we can conclude that cytotoxicity and damage are observed at the level of the genetic material both in preparations without Zn, without Cd and with Zn+Cd. There was also a progressive increase in the extrusion of the nucleolar material, generating cellular apoptosis, which could be attributed to the plant's response to the high toxicities of Zn with Cd.

A158

VARIETY CABERNET FRANC (*VITIS VINIFERA*) IN VITRO PLANTS PRODUCTION

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The vine (*Vitis vinifera*) is considered one of the most important fruit crops in the world for its cultivated hectares and its economic value. Plant biotechnology can provide valuable tools that allow the best use of these plants. For this reason, it is necessary to obtain information about the viability of this species and its different varieties by using the *in vitro* culture. The objective of this study was to obtain mother plants of the Cabernet franc variety through *in vitro* propagation and thus determine its viability and performance for being propagated with this technique. We worked with plants of the Cabernet franc variety, obtained from vineyards of Sol Puntano, San Luis. Uninodal cuttings were cleaved from these plants, properly disinfected and seeded in 50% Murashige Skoog (MS) nutritive media with the addition of 0.01% indole acetic acid. After eight weeks of growth, the data on explant establishment, rooting, the number of leaves, and callus formation were recorded weekly and statistically analyzed. Then, they were transplanted to a 1:1 mixture of sterile perlite: fertile soil acclimatized for 4 weeks under adequate humidity and temperature conditions and analyzed their *ex vitro* behavior. For the descriptive analysis of the data of the number of leaves in the different weeks, the average, median, and mode central tendency were calculated, along with measures of dispersion, standard deviation and variance. With the establishment and rooting data, frequency tables were constructed. The normality tests (Kolmogorov–Smirnov test; $P < 0.05$) and homoscedasticity (Levene's test; $P < 0.05$) were also performed. It was concluded that none of the assumptions were fulfilled, therefore, to analyze the correlation the Mann–Whitney and Kruskal–Wallis *U*-tests were used. The results showed that it is a viable variety for *in vitro* culture, with average values of 87.9% in induction and 58.62% in rooting and 5 average leaves per cut. There was also a correlation between rooting – quantity of leaves, being the third week where it was most marked. The percentage of unwanted callus was 0%. The percentage of acclimatized plants was 44.82%. Cabernet franc *in vitro* mother plants showed us that they were a genetic material of high-level performance and health, with good behavior in *ex vitro* acclimatization.

A159

ROOTING AND SPROUTING OF PINOT NOIR (*VITIS VINIFERA*) BRANCHES IN DIFFERENT SUBSTRATES

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The Pinot noir grape is a variety with origin in the French Burgundy with excellent qualities, with medium vigor, with early maturation, not very fertile and lying down, it is very sensitive to fungal diseases, sensitive to mildew and oidium, very sensitive to botrytis, cluster moth, scarring, and mites. Pinot noir is implanted in vineyards with incipient grape production and winemaking in the province of San Luis, which has generated demand for information on this variety. In San Luis, the wine industry is developing with special interest, which establishes a need to generate information of a scientific nature on the differences between varieties in relation to vegetative and enological production, health, agronomic behavior, the influence of terroir, etc. The plant material was obtained from vineyards of the Sol Puntano, in San Luis city. The objective of this study was to select the best substrate to propagate Pinot noir vine branches, where it could develop a good amount of roots and aerial shoots. For this, 100 vine branches with 7 buds were kept cold until their treatment. Under laboratory conditions, they were disinfected with 15% sodium hypochlorite solution and immersed in commercial rooting solution for 15 min. In plastic containers, containing three types of substrate: 1- sand, 2- perlite, and 3- 1: 1 mixture of sand:perlite, the vine branches were placed with three buried buds. They were irrigated with running water and placed in an incubation chamber, at a temperature of 25 ± 2 °C and cycles of 16 h/8 h of light/dark. Root and shoot number data were recorded weekly for three months. The number of roots for vine branches was 15 in sand, 23 in perlite and 21 in sand:perlite. The average number of leaves was 4.5 for all substrates. For the analysis of vine branches rooting, first the descriptive tests of the different levels of rooting were performed for each of the substrates. Subsequently, the normality tests (Kolmogorov–Smirnov test; $P < 0.05$) and homoscedasticity (Levene test; $P < 0.05$) were performed, resulting in none of the assumptions being fulfilled, so a Kruskal–Wallis was performed for each of the two variables (sprouted and rooted) at the second week, resulting in that the differences were not significant ($P > 0.1$) in relation to the substrates used. The relevance of the passage of time ($P < 0.05$) was confirmed by a Cochran Q test that resulted in $P < 0.05$; and a Wilcoxon test $P < 0.05$ in relation to the number of roots and shoots. In conclusion, it should be noted that although the better performance of the vine branches, was observed in the 2-perlite substrate, the differences were not statistically significant. The first stage of mass production of vine branches takes in the nursery has been completed. It has been proven that the substrate used is indistinct for the mass production of rooted and sprouted vine branches.

A160

ANALYSIS OF PATHOGENICITY, FUNGICIDE RESISTANCE AND PATULIN PRODUCTION IN STRAINS OF BLUE ROT PRODUCER *PENICILLIUM* SP.

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The blue rot produced by *Penicillium expansum* is a post-harvest disease that causes significant economic losses in fruits and vegetables. The application of fungicides is the main way to combat this disease. However, there are strains resistant to them, and added to this, public demand for reducing the use of pesticides has grown. On the other hand, this fungus is a producer of patulin. This compound is a mycotoxin that causes acute and chronic diseases. The goal of this work was to analyze *Penicillium sp.* strains in terms of their ability to produce blue rot in apples, resistance to fungicides, and patulin production. In addition to select the best candidates for future studies using different control methods. We worked with 10 strains of *Penicillium sp.* isolated of pear and apple rot from Alto Valle de Río Negro, one isolated from San Luis, and two from references. The pathogenicity tests were performed on commercial *Red delicious* apples washed and then wounded to inoculate them with 20 µL of a suspension of 10⁶ conidia/mL. Apples were stored 7 days at 25 °C and the diameter of the rots was measured (mm). The fungicide resistance at the doses recommended for standard postharvest treatments was evaluated “in vitro”. A suspension of spores of the fungus was dispersed in plates with PDA. Then, holes were made, and there were inoculated with 5 fungicides Captan, Tecto (Thiabendazole), Carbendazim, Scholar (Fludioxonil), Penbotec (Pirimethanil) or water. Development or not of fungus was observed at 3 and 7 days of incubation at 25 °C. Patulin determinations were performed by culturing the strains in PDA medium 7 days at 25 °C. Then, the toxin was extracted and determined in HPLC-UV according to the modified AOAC technique. Regarding pathogenicity, INTA-1, INTA-2, INTA-6, INTA-10 strains developed rot diameters between 39–36 mm. On the other hand, the rest of the strains presented diameters smaller than 33mm. All the strains tested were resistant to Captan fungicide. Also, INTA-6 and INTA-10 strains showed total resistance to Tecto and Carbendazim and partial resistance to Penbotec and Scholar. On the other hand, the strain isolated in San Luis presented total resistance to Tecto, Carbendazim, Penbotec, and Captan. INTA-10 strain presented the highest production of patulin of 119.78 µg/g PDA. However, the rest of the strains presented production below 45 µg/g PDA. From the results obtained, it is observed that all strains showed resistance at least one fungicide, several strains to 2 or 3 of them. The longer the fungicide was present, the greater resistance was observed. Strains also presented different capacities to generate blue rot in apples and to produce patulin. In addition, it was concluded that the INTA-10 strain is the best candidate to continue studying different control methods. It is highly pathogenic, resistant to fungicides, and has the highest production of patulin.

A161

PRELIMINARY STUDIES OF PHENOLOGY OF DURAZNERO (*Prunus persica* L.)

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Fruit development should be based on knowledge of certain agroclimatic factors. Performance and quality are genetically determined, but strongly influenced by climatic factors, such as frost. Knowing the phenology of the different varieties for fresh consumption and industry, in regions not typically producing fruit, will help to make an adequate choice of varieties, optimizing productive resources. In the province of San Luis, there are four zones with different edafoclimatic characteristics, which determine the productive potential of a particular variety, being favorable for one of the four zones, and in others, it will be harmed. The work was done in 2016–2017 and 2017–2018 seasons in six peach varieties: Spring crest, Flavor crest, Maria bianca, Carson, Andros, and Bowen, in Sol Puntano San Luis experimental property. To carry out this study, we worked with a plot of experimental - productive varieties of one hectare; were the phenological states were evaluated according to Baggioini. The evaluations were carried out every 3 three days for 5 months from the beginning of vegetative activity until harvest (July–November). Frosts were recorded for each date, correlating these with the phenological events to determine their incidence in the different varieties. The meteorological data were taken from the Weather Station Network (REM) recording the temperatures and rainfall. Phenological states were recorded by variety and climatic conditions by date. The peach phenology begins in mid-July, the occurrence of frost is not critical until mid-August, because it is observed in yield states: winter and swollen. The early varieties: Spring crest, Flavor crest, and Maria bianca are in full bloom status (F) in the first half of September, more precisely September 12 as the average date; while late varieties (Carson, Andros, and Bowen) are in full bloom status (F) in the second half of September, more precisely on September 21 as the average date. During the period 25/8–25/9 the occurrence of critical damage temperatures, was recorded, requires the active defense to ensure a satisfactory set. The deviation in the early varieties was ±7 days with respect to the average and the late varieties ±10 days in relation to the average. The late-flowering varieties: Carson, Andros, and Bowen have development possibilities, are not affected by critical temperatures of flowering damage. Early flowering cultivars are more susceptible to the occurrence of frost, they concentrate flowering in the indicated critical period, while late cultivars do so outside the critical period, guaranteeing a lower probability of loss of fruits. Productivity in these conditions requires to implement a system of defense against frost and protection with anti-hail mesh. These results can help with the development of alternative or complementary strategies to improve the ecological, economic, and social sustainability of the fruit systems of the central west of the province of San Luis.

A162

EFFECT OF DIFFERENT OSMOTIC POTENTIALS WITH POLYETHYLENGLYCOL ON THE GERMINATION OF *TETRACHNE DREGEI* NEES

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Tetrachne dregei Nees (tetrachne “green grass”), is an herbaceous forage species belonging to the family of the Poaceae. It is native to South Africa and Pakistan. It is a species with good forage quality for livestock considered provisional for the semi-arid temperate region in EEA-INTA San Luis. There are 40-year plots that survived winter with a frost of -17°C . The difficulty to achieve its implantations is a limitation for the implantation of the extensive cultivation of this species. The purpose of this paper is to investigate the effect of different osmotic potentials regulated by different concentrations of polyethyleneglycol 6000 on the germination of seeds. The seed was collected from 10-year-old plots, previously fertilized with 100 kg/ha with 46% urea. Four treatments were performed 1–0.0 MPa–control (distilled water), 2–0.5 MPa, 3–1 MPa, 4–1.5 MPa with three repetitions each, 20 seeds were used for a box, so 60 seeds were counted for treatment. Germination energy was calculated at three days and germination power at seven days. Germination conditions were as follows: 30°C (8 h light) and 20°C (16 h dark). The following results were obtained: control (with water) was obtained from EG 16% and the PG of 73%, in treatment 1 (0.5 MPa) the EG gave 0% the PG 3% the rest of the treatments was negative. It was concluded that *Tetrachne dregei* Nees germinated without difficulty in control conditions at 0.0 Mpa of osmotic potential and present strong sensitivity to osmotic stress as opposed to tolerance to saline stress present by the species, according to our previous studies.

A163

SPATIAL PATTERN OF *Puccinia sorghi* “COMMON CORN RUST” IN VILLA MERCEDES, SAN LUIS

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The “common corn rust” *Puccinia sorghi* is one of the main diseases crop and every year it presents epidemic characteristics that are featured in different areas of Argentina. Optimal management of the disease requires the generations of information about its biology but no studies of its spatial distribution are available in the country. In order to define dispersion patterns, the disease severity of 50 geo-referenced plants was assessed weekly, in a completely randomized design with three repetitions in vegetative and reproductive stages from January to March, in 2014, 2015 and 2016 production campaigns in Villa Mercedes, San Luis. The severity was assessed by measuring the pustules number per third of the leaf (tripartite method), in functional plant leaves. Disease maps were generated for exploratory analysis and patterns were defined by means of the Morisita index of aggregation. Morisita is used to evaluate the spatial distribution pattern of disease, it has two variables, the total number of plots (n) and the number of individuals per plot (x), $I_g = n [\sum(x^2) - \sum x] / (\sum x)^2 - \sum x$. The infection foci determined by disease maps were constant in the evaluation months of each year. Morisita indexes reflected two dispersion patterns according to the degree of severity. When the severity is less than 5% it behaves uniformly ($I_g < 1$), but when the severity increases above 5% the distribution pattern randomizes ($I_g \approx 1$). These results reflect the disease pathosystem. When the plants stop generating new leaves, the number of pustules is concentrated due to the production of uredospores and the increase of senescent tissue favors the production of teleutospores and hinders the horizontal contagion of the epidemic. The effect of “concentration” change the diseased distribution pattern but such effect was not enough to make aggregations.

A164

SCATTERING MODEL OF *Puccinia sorghi* “COMMON CORN RUST” IN VILLA MERCEDES, SAN LUIS

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The “common corn rust” *Puccinia sorghi* is one of the main diseases of the crop and every year it presents epidemic characteristics in different areas of Argentina. For the optimal management of the disease, it is necessary to generate information about its biology and in the country, there are no studies of its spatial distribution. In order to identify the foci of infection and record the distance of inoculum displacements, the severity of georeferenced plants was evaluated weekly in plots of 5 x 2.5 m, distributed in a completely randomized design with three repetitions in vegetative and reproductive stages during the months of January to March, in 2014, 2015 and 2016 production campaigns in Villa Mercedes, San Luis. Severity was assessed per third of the leaf (tripartite method), in all the functional leaves of each plant. Epidemic maps were generated to detect the foci of infection and linear matrices and graphs were generated with the distance and severity variables per plant taking the center of the infection focus as “0”. With the R statistic, the field data were adjusted with the models Linear ($y = ax + b$), Exponential Negative ($y = ae^{-bx}$) and Gregory ($y = ax - b$). The best adjustments were with the Gregory Model. The evaluation of all the generated matrices determined that the last 2 evaluations have a strong negative correlation (-0.80). The gradient slope was smooth, -0.62 for 2016, -0.37 for 2015 and -0.99 for 2014. The significance increases slightly as the evaluations progress, which would indicate that at the beginning when the disease is installed,

even contagions are not evident but as the system matures, the plants adjacent to the focus of infection will express the lesions resulting from the contagion of secondary inoculums and that as the distance increases the percentage of severity declines.

A165

PHYSIOLOGICAL RESPONSES OF “JARILLA HEMBRA” (*LARREA DIVARICATA*) TO PLUMBUM CONTAMINATED AREA: MULTIVARIATE ANALYSIS OF STRESS VARIABLES

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Larrea divaricata is a native plant adapted to a wide range of arid environments. Our group is assessing this species as a phytoremediator for lead (Pb) contaminated areas, so, the aim is to determine its physiological response under stress caused by Pb bullets of a shooting range field. Plants growing at greenhouse were planted in soils with 0 and 1000 ppm Pb (0 Pb and 1000 Pb) in the same field, and they were compared with plants that continued at the greenhouse (GH). After 6 months, samples of aerial parts were collected for lipid peroxidation (LP) analysis and determination of catalase (CAT), ascorbate peroxidase (APX), and guaiacol peroxidase (GOPX) activity. Results were analyzed by uni (ANOVA and Tukey test) and multivariate statistics (PCA). Univariate statistics determined no significant differences in LP between treatments although plants 1000 Pb showed a significant decrease of CAT, APX, and GOPX activity respect to plants 0 Pb. The behavior of GH plants was similar to 0 Pb of field plants, although univariate statistical did not show significant differences between GH and 1000 Pb plants either. Nevertheless, PCA showed in scores plot of PCA three different groups: 0 Pb, 1000 Pb and GH. Loadings plot showed that 0 Pb was differentiated from the other groups by the influence of APX and GOPX variables in PC1, while 1000 Pb and GH were differentiated by the influence of LP and CAT variables in PC2. Results of PCA indicate that the activity of CAT, APX, and GOPX is reduced in 1000 Pb treatment on field respect to 0 Pb and GH. In addition, lipid peroxidation is minor in GH plants than both field treatments (0 Pb and 1000 Pb) while CAT activity is higher in GH than in field treatments. Effects of oxidative stress are evident at field treatments in comparison with GH plants as well as the decrease of antioxidant response in plants under Pb contamination respect to both controls (0 Pb and GH). Multivariate analysis was capable of differentiating three growth conditions and determine their physiological status. In conclusion, although the first pilot essay shows the effects of Pb over *L. divaricata* physiology, more studies are needed to determine whether *L. divaricata* is potentially useful as a phytoremediator plant.

A166

EVALUATION OF THE FORAGE MASS AND QUALITY IN A BOVINE BREEDING SYSTEM IN *DIGITARIA ERIANTHA* AND *ERAGROSTIS CURVULA* IN THE SOUTH OF SAN LUIS

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El Centenario Establishment with 54,000 hectares, located in the south-central part of San Luis, called sandy land area with grasslands and shrubby chañar (*Geoffroea decorticans*), is exclusively dedicated to an extensive dry breeding rodeo (400 mm), in which 80 to 90% of the psammophytic grassland cover is dominated by a non-fodder species (*Elionurus muticus*), with an average receptivity of 15 to 20 ha/EV. The objective of this work was to determine the production and seasonal quality of *Digitaria eriantha* and *Eragrostis curvula* and the need to supplement it. A module of 2118 ha was evaluated where there was degraded grassland and replaced with 100% of digitaria and lovegrass. In this unit, there were 390 suckler cows providing a receptivity of 5.4 ha/EV. Regarding the initial situation with degraded pasture, an increase in receptivity was obtained between 176% and 268%. Four samples were taken, one for each season of the year (Summer, Autumn, Winter and Spring). Sampling was carried out from the watery point to the farthest point of the lot, diagonally, every 200 m using the modified Botanal system. The cuts were made with a rectangle of 0.50 x 1 m with a cutting surface of 0.5 m². The samples were dried in an oven at 65 °C and dry matter was obtained from each cut, the total forage mass was determined and their respective analyses were carried out in the laboratory (crude protein and digestibility). During spring–summer and part of autumn, grazing was on lovegrass and during that period the digitaria rests without grazing. During the spring–summer, the cumulative production of digitaria was 4782 kg/ha with digestibility of 42.3% and 8.3% of CP, during the winter grazing of digitaria was obtained an average of 3.5% CP, indicating the need for supplementation to improve quality during winter grazing. The lovegrass during grazing maintains levels of availability close to 50 kg/ha with a quality of 7% of CP, indicating that quality supplementation is not necessary. It is concluded that the availability for the breeding rodeo is in balance while the quality during the winter is lower than the requirements of the suckler cows indicating the need to supplement.

A167

EVALUATION OF THE BOTANICAL COMPOSITION OF FECES BY THE MICROHISTOLOGICAL TECHNIQUE IN A BOVINE BREEDING SYSTEM IN THE ESTABLISHMENT "EL CLARÍN", MENDOZA

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"EL CLARÍN" is located in General Alvear (Mendoza) and has 5500 ha and 16 lots. The historical isohyet is 450 mm. 60% of its surface is psammophytic grassland and 40% is "jarilla arbustal". The most important native species are *Trichloris crinita*, *Nasella tenuis*, *Aristida mendocina*, *Panicum urvilleanum*, *Poa lanuginosa*, *Pappophorum caespitosum*, and *Elyonurus muticus*. In some lots, there is a low proportion of desirable species and low productivity, for that reason *Eragrostis curvula* (lovegrass) has been implanted. The objective of this work was to evaluate the botanical composition of the diet, through the microhistology feces technique of a system of cattle breeding in the grazing lots during the four seasons of the year. Monthly samplings were made on fresh feces, not less than 40 stools (dung) by sampling, in the water fountains during the morning. Samples were dried in a forced-air stove at 65 °C for 48 h, using a Wiley mill with 1 mm mesh. Were decolorized with 5% sodium hydroxide (NaOH) and 5 min of boiling and washing and were dehydrated with 25% ethyl alcohol. We used safranin stain. The preparations were mounted on slides with "Jelly Glycerin" and analyzed and identified in a light microscope. In the natural grassland lots enriched with *Eragrostis curvula*, 60% was found in the spring-summer and autumn, and low proportions in winter. In winter, where forage availability decreases, 60% of *Aristida mendocina* was observed. They were 27% and 19% in spring and summer respectively. Also in winter 13.3% of *Poa lanuginosa* and lower percentages of *Botriochloa springfieldii* and other broadleaved were found. In lots with mount used only autumn-winter time and closed the rest of the year, 48% of *Aristida mendocina* and 20.5% of *Trichloris crinita* in winter and 15% of *Trichloris crinita* in autumn were observed. We concluded that the breeding cows selects and regulates its diet based on the presence and time of the year, showed an important selectivity.

A168

EVALUATION OF THE FORAGE MASS AND QUALITY IN A BOVINE BREEDING SYSTEM AT THE ESTABLISHMENT "EL CLARÍN" MENDOZA

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The establishment "EL CLARÍN" is located on Route 188, km 716 (Gral. Alvear) Paraje La Mora, the phytogeographic region of "Monte". Livestock is developed in a complex environment: with highly variable rainfall within the historic isohyet of 450 mm. It has 5500 ha with 16 lots. 60% of its surface is psammophytic grassland and 40% is "jarilla arbustal". The most important native species are: *Trichloris crinita*, *Nasella tenuis*, *Aristida mendocina*, *Panicum urvilleanum*, *Poa lanuginosa*, *Pappophorum caespitosum*, and *Elyonurus muticus*. In some lots, there is a low proportion of desirable species and low productivity, for that reason *Eragrostis curvula* (lovegrass) has been implanted. The objective of this work was to evaluate the quantity and quality of the forage mass present in the lots in the four seasons of the year of a bovine breeding system. Vegetation surveys were carried out, using the tenth of a square meter of Daubenmire and a quarter or a half square meter (Botanal). The species, frequency, coverage and cut were determined to obtain dry matter (kg/ha) quality (%CP) and digestibility (%D). Lots were sampled diagonally every 200 m perpendicularly from the watery point. To ensure the observation of all available species at the main times of the year, the surveys were carried out in the four seasons of the year. The samples were dried at 65 °C during 48 h, then, were ground and sifted (1 mm). Were determined dry matter, crude protein and digestibility percentage. In poaceae, in autumn, were found ranks of 3.6 to 9.6 %CD, 30.7 to 47.7 %D and availability (Dkg/ha) of 70 for lots with native species at 1500 kg/ha with lovegrass and %D=4. In winter, the ranges were 3 at 6.5 %CD, 30.5 to 51.5 %D and Dkg/ha of 30 for lots with native species at 500 kg/ha with lovegrass. In spring, the ranges were 5.4 to 12.9 %CD, 38.5 to 57.9 %D and Dkg/ha of 150 for lots with native species at 2000kg/ha with lovegrass. In summer, the ranges were from 8.3 to 12.3 %CD, 38.8 to 53.8%D and Dkg/ha of 200 for lots with native species, at 2500 kg/ha with lovegrass. In conclusion, lovegrass increases significantly the forage availability, and the native individually have good quality, but not the quantity of forage mass for the requirement of the suckler cows.

A169

ISOLATION AND SELECTION OF TOLERANT RIZOBIOS TO SALINITY AND CADMIUM

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Microbial communities are a fundamental part of soil biota. Against phytotoxicity effects, soil microorganisms associated with the rhizosphere play a very important role in protecting plants from damage caused by heavy metals and other types of contaminants. The objective of the present study was to identify the rhizobia tolerant from saline soils and select those that have a greater tolerance to salinity and cadmium. For the isolation of rhizobia strains, an induction test of the nodulation in vitro was carried out in the laboratory of the Institute of Agricultural Sciences, CSIC (Madrid). The study was conducted in test tubes with alfalfa seedlings as trap plants, and as growth support, a modified Jensen culture medium with deficiency of Nitrogen. With the aim of promoting the nodulation, the seedlings were inoculated with a solution containing 1 g of soil previously sampled in EEA INTA San Luis and 9 mL of distilled water. At 15 days, the pink-colored nodules were removed and sterilized for the isolation of rhizobia strains. The sterilized nodules were cut transversely and the bacteria in the central area of the nodule were pecked on tryptone yeast extraction (TY) agar plates that were incubated at 28 °C for 3–5 days. For the extraction of bacterial DNA, the UltraClean KIT was used and, the 16S rRNA gene was amplified by conventional PCR. Ten isolates of rhizobia were obtained which, once the purified DNA was obtained, the samples were sent to SecuGen S.L to be sequenced. The 16S rRNA sequence of gene similarity was performed using the BLASTN tool from the NCBI website. Likewise, the sensitivity of rhizobia isolates to NaCl and Cd was determined by evaluating growth inhibition on TY agar plates. To study NaCl tolerance, the TY agar medium was supplemented with an increase in NaCl concentrations (0–800mM). On the other hand, to evaluate the tolerance of the metal, CdCl₂ was added to the TY medium with an increase in concentrations of (0–200 µM). The plates were left incubating for 48 h to subsequently perform the assessment. All isolates obtained, 10 in total, belong to the bacterial species *Sinorhizobium meliloti* with a 99% identity percentage. They have high a tolerance to NaCl, with a MIC value of 400 mM NaCl, which indicates that exceeding these concentrations said isolates cannot grow; only one strain had an inhibitory value of 600 mM NaCl, being the most salinity tolerant. On the other hand, most of the isolates studied have a tolerance to Cd, only three of them have the highest MIC since they grow at 200 µM of CdCl₂.

A170

BOTANICAL CHARACTERIZATION AND PHOTOACTIVITY DIFFERENCES BETWEEN *BIDENS SUBALTERNANS* VAR. *SUBALTERNANS* AND *BIDENS SUBALTERNANS* VAR. *SIMULANS* AGAINST SPECIES OF GENUS *CANDIDA*

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The finding of new antimicrobial agents has become the main objective of health sciences. Because of this, search for a new therapeutic alternative for the inactivation of pathogenic microorganisms resistant to traditional chemical agents is imperative. Plants are an important source of chemical diversity providing unlimited opportunities for isolating new compounds or generating photoactive extracts with high antimicrobial potential. Although medicine has contributed to the survival of immunosuppressed patients, a high percentage died due to microbial infections. Drugs available for the treatment of superficial or systemic mycosis often show high toxicity, produce recurrence or develop resistance by pathogens. New techniques developed from photodynamic therapy could be considered according to their ability to decrease the microbial load using plant extracts already used in popular medicine. Therefore, the main objective of this work was to characterize the species *Bidens subalternans* var. *simulans* and *B. subalternans* var. *subalternans* and establish the high potential for photodynamic inactivation against yeasts. For this purpose, aerial and subterranean parts were collected and characterized. The characterization of the species allowed us to identify parameters macro- and micrographics, both quali- and quantitative, applying macro-/micromorphological and taxonomic study methods to provide characters for effective quality control of raw drugs and herbal medicines containing them. The dry and ground vegetal material was extracted successively with solvents of different polarities; then the effectiveness of the extracts to inactivate yeasts of the genus *Candida* was observed using a microdilution assay recommended by Clinical and Laboratory Standards Institute under UVA light irradiation. It was possible to determine the high and different photoinactivation potential of extracts of *B. subalternans* var. *simulans* and *B. subalternans* var. *subalternans*. The test was performed in triplicate and the minimum concentration without microbial growth was determined visually as spectrophotometrically. Minimum inhibitory concentration (CIM₁₀₀) with 100% inhibition was determined in the range of 400–1000 µg/mL for *B. Subalternans* var. *subalternans* and between 15–30 µg/mL for *B. Subalternans* var. *simulans* against different yeasts of the genus *Candida*. In addition, from chromatographic and spectrophotometric determinations, we have presumed that photoactive metabolites as thiophenes and polyacetylenes are present in the extracts, whose phototoxicity also appears to be related to the synergy between these components.

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PRELIMINARY STUDY SHOWS GERMINATION OF SEEDS *CEREUS AETHIOPS*

IMPROVED WITH ACID SCARIFICATION

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Seed dormancy is defined as a state in which an alive seed is unable to germinate. This is an ecological adaptation mechanism that lets plants survive in an unfavorable environmental condition; the seed germination ability can be regulated by environmental signals. *Cereus aethiops* seeds, like many other cactus species, show low germination capacity due mainly to their hard-lignified integuments, obstructing water absorption and radicle protrusion. The purpose of this study was to accelerate the initiation of the germination process by the action of chemical agents as the sulfuric acid (H₂SO₄) at 98% and chloric acid for three lengths of time: 1, 2.5, and 5 min were tested as germination enhancers. Germination assays were performed in Petri dishes and under controlled conditions at constant temperature (20 ± 23 °C) and a photoperiodic regime of 12 h light per day, and continuous darkness. All assays followed a completely randomized design with three replications of 30 seeds each treatment. We found that chemical scarification using sulphuric acid produced germination percentages generally higher than those for seeds incubated in chloric acid. This happened under photoperiodic conditions gave the highest germination percentage in the shortest time. None of the assays carried out to establish if different agents could remove the inhibitory effect of continuous darkness on seed germination. This showed that they are positive photoblastic seeds.

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MINIMUM INHIBITORY CONCENTRATION (MIC) AND FUNGICIDE /FUNGISTATIC EFFECT OF EUGENOL, ISOEUGENOL, CARVACROL AND THYMOL ON *LASIODIPLODIA TEHOBROMAE* ISOLATED FROM VINES WITH SYMPTOMS OF GRAPEVINE WOOD DECAY IN SAN JUAN PROVINCE

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The vine disease known as “Grapevine wood decay”, which is caused by ligninolytic fungi, is internationally considered to have a great impact, as it causes the death of plants. The production of wine is the most important economic activity in the province of San Juan, so any pathology that alters the yield and quality of the vines has a strong economic impact. These pathologies are produced by a pool of ligninolytic fungi, which cause Eutypiosis, Esca, Petri Disease, BAD (Black Dead Arm), Malvón Leaf. Recently, *Lasiodiplodia tehobromae* has been reported as a causative agent of irreversible destruction for the disease. We isolated the ligninolytic fungi from vine plants in San Juan. After morphological and molecular analyses, the presence of *Lasiodiplodia tehobromae* was confirmed. Minimum inhibitory concentrations (MIC) of Carvacrol, Thymol, Eugenol, and Isoeugenol were determined through a bioassay using 5, 4, 3, 2.5, 2, 1.5, 1, 0.75, and 0.5 mM for each compound. Three replications of each treatment were performed. In order to evaluate if the compounds performed as a fungicide or fungistatic agent, a subsequent bioassay was performed when the absence of mycelium was detected in the MIC bioassay. It was obtained which MIC for Thymol, Carvacrol, Eugenol, and Isoeugenol were 0.75, 0.75, 2.5, and 3 mM, respectively. Also, Thymol, Carvacrol, Eugenol, and Isoeugenol were found to be fungicidal agents at 3, 3, 1, and higher than 5 mM, respectively, according to visual inspection in the fungicide/fungistatic bioassay, demonstrating to be potential natural agents to continue their studies in the treatment of this disease.

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YIELD AND INULINE CONTENT IN TUBERS OF TWO *HELIANTHUS TUBEROSUS* L. CULTIVARS

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The Jerusalem artichoke tuber, *Helianthus tuberosus* L., has the potential to expand as a commercial product in different areas, from its use as forage through its horticultural use, to its industrialization to obtain inulin. In the latter case, its use for the production of gluten-free or starchy flours is also found. This aspect occurs because the plant stores its reserves in the form of inulin (fructose polymer) instead of starch (glucose polymer). Inulin is considered a prebiotic since it is not degraded by the digestive enzymes of the organism but reaches the intestine where it is used by the microorganisms generating health benefits. The plant regrowth during spring and between 14 and 16 weeks the stages of tuber formation begin. In the autumn, senescence begins and the translocation of nutrients to the reserve organs occurs. The objective of this work was to establish the yield for two cultivars (white and red) in the semi-arid region of San Luis during a crop cycle and compare the inulin content of the tubers for these varieties. A comparative performance test was carried out between two Jerusalem artichoke cultivars (red and white) at the INTA San Luis Experimental Station. The planting was carried out in September 2017 on plots of 5.25 m² with three rows each and with a planting frame of 0.50 x 0.50 m. A completely randomized design with 5 repetitions was used. At the end of the cycle (217 days after de regrowth), after the

last frost when the plant completed the translocation to the reserve organs, the tubers of the intermediate rows of the plots were harvested. The yield of fresh tubers for each variety was calculated from the harvested material. The tubers were processed (washed, cut, and dried) and then ground and screened to determine inulin content. Inulin extraction: it was performed in 10 g samples with water (1:16 w/v) at 75 °C for 90 min in a thermostatic water bath with constant agitation. Then, the samples were filtered and the solid was subjected to two other extractions under the same conditions. The inulin content was determined by the Somogyi-Nelson method. Both cultivars were able to produce tubers. The results showed a greater production of tubers for the white cultivar (68.0 vs 42.1 Tn/ha, white and red, respectively) with significant statistical differences ($P < 0.05$). On the other hand, difficulties were observed for the “red” variety at the beginning of the trial for the crop establishment, this could be verified by emergency and plant numbers measurements in further studies. Regarding inulin contents, the white treatment showed higher levels than the red (13.05% vs. 11.35%, respectively); the tests were performed in triplicate and statistically analyzed with the Student’s *t*-test program and obtaining a level of significance less than 5%. From the results, it is concluded that the white cultivar showed a better productive adaptation in the semi-arid region with adequate yields and with higher inulin contents.

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APPEARANCE OF LEAVES AND NUMBER OF STEM IN TWO *HELIANTHUS TUBEROSUS* L. CULTIVARS

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Jerusalem artichoke (*Helianthus tuberosus* L.) is a species that has multiple options for use, but also one of the greatest advantages of the crop is its adaptability to different environmental situations and productive regions. It can be grown in soils with a wide pH range (4.4 to 8.6) and under different rainfall regimes (between 310 to 2800 mm). The main factors that can restrict its performance in our region are the edafoclimatic conditions (mainly water and nitrogen) and competition with weeds. Its growth begins in the spring with the regrowth of the tubers and each plant develops a different number of leaves and stems. The rapid intergrow coverage and good foliar development allow a competitive advantage over the presence of weeds. The objective was to measure the appearance of leaves and the number of stems per plant for two cultivars of Jerusalem artichoke: red and white. The planting was carried out in September 2017 on plots of 5.25 m². Each plot consisted of 3 rows and a planting frame of 0.50 x 0.50 m. The design was completely randomized with 6 repetitions. The number of leaves and stems per plant was measured every 15 days, for which a subsampling was done by randomly selecting 3 specimens of each plot to establish an average value of the number of leaves and stems per plant. Regression models were made that related the number of leaves and stems with time. In addition, an ANOVA was performed to compare the highest values achieved (12/26/19). For each variety, a quadratic regression model was obtained that related the variation of the number of leaves with the course of time. Both regressions showed adequate adjustment values ($R^2 = 0.89$ and $R^2 = 0.67$; white and red, respectively). There is an evolution in the number of leaves that reach their highest values between mid and late December. On the other hand, it is detected that the white variety shows an earlier regrowth and a greater rate of foliar appearance at the beginning of the crop, which implies that it requires less time to develop the first leaves. As for the regressions for the number of stems, there was no relationship since the number of stems was defined at the beginning of the crop and did not change over time. When comparing the highest values achieved (12/26/19) between both variables, it was determined that the white cultivar reached higher levels with significant statistical differences ($P < 0.05$), for both the number of leaves and the number of stems (12.69 vs 21.61 leaves/plant; 1.33 vs 3 stems/plant). From the results obtained, it is concluded that the white variety had a greater number of leaves and stems in the semi-arid region of the province of San Luis and also had an earlier regrowth with a faster rate of foliar appearance that could allow a better capacity of competence to settle against the invasion of weeds, by covering the interrow more quickly.

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ALPERUJO INOCULATED WITH *PLEUROTUS OSTREATUS* ANTI-OXIDANT ACTIVITY

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In San Juan province, between 60000 and 100000 tons of olive are milled for olive extraction using two-phase extraction system. As a byproduct it is obtained 45000 tons of Alperujo (AL) which final disposition is difficult due to its contamination effect, derive from its high polyphenol content (PC) and lipids. The use of biotechnological processes, alone or combined with physico-chemicals ones are a possibility for AL treatment. It is remarkable the *Pleurotus ostreatus* ability for degrading PC in solid waste or by-products from agro-industrial processes. The use of AL as non-conventional animal feed is limited because its low content of crude protein and also for PC usually considered as anti-nutritional. Some phenolic compounds present in AL have antioxidant activity (AA). The antioxidants present in food protect structures against free radicals formation. This process is related, not only to physiological aging, but also to a series of cardiovascular, degenerative, such as Alzheimer’s, Parkinson diseases, as well as different types of cancer, so the presence of antioxidant compounds in food are considered beneficial for health. In previous works, substrate formulation containing AL among other agro-industrial waste from San Juan was studied to obtain optimal conditions for *Pleurotus ostreatus* production. The aim of this work was to determine AA in fermented substrate after fruiting bodies harvest. Three kg of substrate (AL 40% p/p, fodder oat 40% p/p, grape stalk 10% p/p and Alamo chip 10% p/p, pH 6, initial water content 30%) were held in polypropylene bags (B), sterilized for 15 min at 121 °C and 1 atm. Then was inoculated with 3% fodder oat grains (cultivated during 15 days at 27 °C in dark conditions). Inoculated substrate was incubated 15 days at 27 °C in dark conditions, and then a thermic shock

was applied at 4 °C for 48 h, in order to induce the fruiting stage. Finally, conditions were maintained at 17 °C and 80% relative humidity with 12 h of 200 Lux photoperiods for 25 days. As a control, a parallel test was performed, (C) prepared with fodder oat 100% (p/p). The essay was done by triplicate. First harvest for Substrate (HB1) and Control (HC1) was done after 30 days. Second harvest, (HB2) and (HC2), 10 days later. AA was measured using a test based on the stability free radical DPPH and it was determined in harvest HB1, HB2, HC1, HC2, in control and in substrates covered with mycelium at 15 days (C1 y B1), at 30 (C2 y B2) and 40 days (C3 y B3). The results show that in control, AA remained stable over time, around 29.17%. Instead, a significant increase was noticed from B1 (30.33%) to B2 (61.16%), remaining almost constant in B3 (60%). An increase was also noted when compare HC1 (51.3%) with HB1 (67.7%) and HC2 (40.1%) with HB2 (87.7%). These results would indicate that the presence of AL increased AA in the fermented mixtures, improving their nutritional properties. In this way, the AL could be considered as a suitable raw material for mushroom production, and the fermented substrates after harvest could be an alternative for animal feed.

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MINERAL CONTENT IN AROMATIC SPECIES FOR CULINARY AND MEDICINAL USE

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Aromatic plants provide nutrients that can have health benefits; in fact, almost all aromatics are also known as medicinal. Throughout the history of mankind, it has been observed that many people have acquired empirical information on the medicinal properties of a large number of plants in their environment. A wide variety of herbs and spices, individual and combined, have been extensively investigated to assess their effects and synergistic actions on health and diseases. The value of aromatic plants and spices in nutrition is due to the fact that when they are consumed, they incorporate minerals, in addition to flavoring using aromatics it allows us to enhance flavors and reduce the consumption of sodium and salt. The objective of this work was to determine the content of minerals, calcium (Ca), sodium (Na), potassium (K) and phosphorus (P) in rosemary (*Rosmarinus officinalis*), sage (*Salvia officinalis*) and santolina (*Santolina chamaecyparissus*) used for culinary and medicinal purposes. Plant material was first washed with Milli-Q water and dried at 60 °C for 72 h and subsequently mashed to turn it into powder in a mortar with liquid nitrogen. 250 mg of homogenized plant material were acid digested in an ultrasonic bath (30 min at 110 °C) with 5 mL HCl 8%. The samples were analyzed by MIP OES (Agilent MP 4100). Certified reference material RM-Agro E1001a (*Brachiaria brizantha* cv. Marandu) from the EMBRAPA Pecuaría Sudeste (São Carlos, SP, Brazil) was included in each batch for quality control. The mineral concentrations were: 5.47 g/kg, 7.01 g/kg, 12.62 g/kg and 1363 mg/kg, for Ca, Na, K and P, respectively, in santolina; 7.34 g/kg, 5.94 g/kg, 11.46 g/kg, and 982 mg/kg for Ca, Na, K and P, respectively, in rosemary; and 6.79 g/kg, 10.19 g/kg, 24.67 g/kg, and 1762 mg/kg for Ca, Na, K, and P, respectively, in sage. The results obtained were evaluated and compared with the limit or recommended values, established by national organizations (ANMAT 1999, 2004-CAA), being in acceptable values that contribute to human health not only through its proven therapeutic activity but also to the daily intake of essential minerals.

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COMPARATIVE TESTING OF TWO CORN HYBRIDS (DENT AND SEMIDENT) GRAIN PERFORMANCE USED IN BOVINE FOOD

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Corn grain represents for our and nearly all of the world countries, the most used component as an energy supplement in cattle feeding. In intensive pastoral systems of meat production, feedlot plays an important role in animal's termination phase and corn grain is normally used in rations formulation. The crop yield has a direct influence on the rodeo feeding cost. Corn grain is composed of an outer envelope (pericarp) and the seed (endosperm and embryo). The endosperm is divided in the corneal endosperm and the flourey endosperm. Depending on their proportion, hybrids are called "Flint" or "Dent", with a large number of intermediate hybrids (semi-dent). In Flint type corneal endosperm predominates, while in Dent type mealy endosperm is proportionately greater. Dent type corn is softer and has a lower physical resistance to chewing and starch greater accessibility for ruminal bacteria, improving degradability with respect to Flint type. The objective was to evaluate the yield of two corn hybrids used in the zone, with different grain characteristics, in Villa Mercedes area, San Luis (33°47'25"S, 65°34'12"O). Production of the two different hybrids characterized by their respective seedbeds was evaluated as semi-dent (Syngenta_840) and dent (KWS_4200). A completely randomized design was made with five repetitions. It was seeded on 2018 December 1, with a density of 46000 seeds/ha, with similar agricultural work performed in the zone. It was fertilized on seeded with 46 kg/ha of diammonium phosphate and at the end of December in the state of 4–6 leaves (V4–V6) with 86 kg/ha of urea at broadcast fertilization. Each repetition covered 10 planting lines separated at 52.5 cm and 900 m in length (4725 m²), randomly distributed. It was harvested upon reaching an adequate moisture content for its good conservation on 2019 August 10, and the results were corrected at 14.5% of humidity. A *t*-test ($\alpha = 0.05$) was performed to compare means and significant differences were found between treatments: KWS_4200 had a yield of 7132 ± 340 kg/ha and Syngenta_840 6100 ± 216 kg/ha. Under conditions of this experience, KWS_4200 hybrid has a double advantage: its performance and nutritional characteristics of its endosperm.

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**DRY MATTER, BRIX AND PH OF SORGHUM CHOPPED FOR SILAGE INFECTED WITH
ERGOT**

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Maize and sorghum silage is a diet key component of most feedlot systems in San Luis province, Argentina. Silage is a process whose main objective is damp forage conservation with the minimum loss of dry matter (DM), maintaining the original nutritional value. One important reason in cattle feed is that both the corn and sorghum silage cultivation, have a high potential for quality forage production. Silage reserves despite increasing costs per kg of DM generate security and substantially increase production livestock yields, against the alternative traditional winter crops use. Good silage has pH ranges varied from 3.3 to 4.2 and its temperature is similar to the ambient temperature. Additionally, to obtain good silage, chopped material to ensile key would be in crop and percentage of MS optimal phenological state. The silage process involves lactic fermentation, therefore pH, temperature and organoleptic characteristics are indicative of this process. The sorghum ergot is a fungal disease generally caused by *Claviceps africana* and *Sphacelia sorghi* that infect the crop during anthesis. The environmental conditions that favor their infection are days with high relative humidity and low temperatures, situations that increase in late sowing. Chopped whole plant characteristics were evaluated, of a sorghum crop (BMR 135, Gentos) affected by ergot with damage from 81 to 100%, in Villa Mercedes, San Luis. The plants were cut at 30 cm of height, 7 times between March 11 and July 3, in 4 plots randomly distributed within the crop, and its growth was classified into three stages (EC1, EC2 y EC3). The material was chopped with a MAINERO 4771 were also dry matter content was determined at a constant temperature of 60 °C (MS), Brix (°B) by a manual refractometer (0-32 °B) and pH using a digital pH-meter. The growth was at the beginning of anthesis (EC2) until completion of growth (EC3). ANOVA ($P < 0.05$) was performed for different EC and chopped times. Results show that pH means values significantly gradually increased between EC2 and EC3 (5.0 and 5.6, respectively), over time. MS augmented significantly on successive chopped dates (17, 19, 21, 24, 27, 30, and 41 %), while Brix raised until April 8 (10, 11, 11, and 13) and significantly progressively decreased before the end of growth (9, 8, 7). According to sorghum chopped material evaluation prior to silage, it was observed that coinciding with crop phenological progress, the variables pH and MS have a typical behavior despite Ergot infection, however, °B did not. We can conclude that Sorghum infected with ergot provides a lower quality in terms of energy and grain yield.

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**MINIMUM WATER REQUIREMENTS FOR THE GERMINATION OF SEEDS FROM
CERCIDIUM PRAECOX, “BREA”**

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The physiological processes and the structural characteristics that underlie the germination of a seed, as well as the regulation of the early growth of the seedling, are very important aspects for the establishment of a plant species in its environment. Germination is a key stage in the vegetable cycle, the final number of plants and their yield or survival depends on it. Arid region species, where *Cercidium praecox* “brea” is included, should generally germinate under conditions of low water availability. The objective of this work was to evaluate the minima amount of water required by *C. praecox* seeds to germinate and analyze the effect of low water availability on early seedling growth. Plastic containers equipped with 145 g of vermiculite were conditioned, washed with running water until Field Capacity (FC) was reached and weighed. After 72 h the pots were reweighed and enough water was added to reach 20% (4 mL of water), 40% (38 mL of water), 60% (72 mL of water), 80% (106 mL of water) and 100% (140 mL water) of the FC. At that time, the seeds, previously scarified with hot water, were sown in an amount of 2 (two) seeds per container. Every 72 h the pots were watered maintaining the indicated percentages of FC. After planting the pots were kept at 27 ± 2 °C and natural light. The number of germinated seeds was recorded at 3 (three) and 7 (seven) days after sowing. The result was expressed as Germination Percentage. At 7 (seven) days, root length (R) and aerial part (PA) of the seedlings were measured and the R/PA ratio was calculated. Germination results indicated that the amount of germinated seeds decreases as the percentage of water in the soil decreases, while the R/PA ratio of the seedlings increases. On the other hand, it was observed that the seeds of brea need a minimum amount of 40 mL of water to germinate, although the prolonged lack of water causes in the seedlings a reduction of root growth with respect to the aerial part.

ECOLOGY, ETHOLOGY AND BIODIVERSITY

A180

LARVAL-MASS EFFECT OF NATIVE SPECIES OF FLIES OF FORENSIC INTEREST

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In forensic entomology, the temperature experienced by feeding larval instars is useful to estimate the age of larvae easily from its size (length and weight); however, “larval-mass effect” that occurs due to pronounced gregarious behavior that larvae present when feeding leads to an increase in local temperature. The objective of this work was to investigate the larval-mass effect on larval cultures of *Lucilia purpurascens* and *Lucilia ochriconis* subjected to fluctuating temperatures in the field. The experiments were performed during the spring, summer, and autumn seasons. Eggs of flies were obtained from wild mothers, and for each species were used three replicates of 250 larvae, placed on a piece of fresh beef liver of 300 g. Subsequently, they were conditioned in boxes with fine net and in an enclosure wired up and exposed to the external environment. Temperature and humidity data (external and internal) were measured. With software SPSS with a level of significance $P < 0.05$ a MANOVA was performed. There were only differences in the autumn season ($F=6.732$, $P=0.008$) in the morning and afternoon measurements, both for temperature ($F=6.696$, $P=0.018$), with up to 2.5 °C more for the inner temperature, as for the humidity ($F=6.280$, $P=0.021$), with up to 9.6% less for the inner temperature. It is possible that this thermal increase combined with lower humidity, which gives a higher thermal sensation, helps to maintain feeding activity, and above all increases larval survival. A highly supported hypothesis could be that thermal difference arises from the quantity or volume of larvae used, with approximately 250 larvae report temperature increases below 4 °C, as in our investigation.

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FIRST INVENTORY OF EPIGEAN SPIDERS OF “SIERRA DEL GIGANTE”, SAN LUIS, ARGENTINA

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Spiders constitute one of the most diverse and widely distributed animal groups, but one of the least known, with an identification percentage of less than 10% of the estimated species. They have been proposed as ideal organisms for biological monitoring and as indicators of environmental quality due to their sensitivity to small changes in environmental conditions and habitat heterogeneity. In addition, the arachnofauna of San Luis province is among the least relieved in our country, which makes it important to inventory its biodiversity. The general objective of this work is to know the diversity of epigeal spiders in Sierra del Gigante through the making of an inventory of soil spiders, determining richness and abundance of the species present. This area is of particular interest because it is located in an ecotone between the ecoregions “Chaco Seco” and “Monte”, showing a particular combination of typical vegetation from both regions. The study was carried out in an area near the Naranjo Esquino area, General Belgrano department, San Luis province. The study area lies approximately at the intersection between 32.9167° South latitude and 66.9° West longitude. Six sites were selected, and the samples were taken at the end of spring 2018 and the end of autumn 2019, periods that correspond to dry and rainy seasons. At each sampling site, properly georeferenced, 12 pitfall traps were placed along three 100 m linear transects (4 traps per transect). Traps contained saline solution (salt [kg]-water [lt] in a 1:8 ratio, plus detergent drops). Material from traps was collected after 7 days of activity, stored at 70% ethanol and transferred to the laboratory for cleaning. Subsequently, the spider material was separated from other arthropods from the samples and properly conditioned. Spider material collected was recorded in electronic spreadsheets and sorted by families and species/morphospecies according to the available taxonomic keys. A digital image database was generated. A total of 245 spiders were captured, The presence of 23 families and 44 morphospecies were recorded, the most abundant being Zodariidae, Lycosidae, Salticidae, Gnaphosidae, Xenocnidae, Anyphaenidae, Amaurobiidae and Orsolobidae. The species *Leprolochus birabeni* stood out remarkably for its abundance during the rainy period, while families Anyphaenidae and Amaurobiidae during the dry season. Data found in this study represents in most cases the first records regarding presence and distribution of the taxa for this area.

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INFERENCES OF GRAZING ACTIVITY OF BREEDING COWS BY SPEED OF MOVEMENT

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Various factors are involved in the daily activities of livestock, but the forage availability and spatial heterogeneity are probably among the most influential. Broadly speaking, periods of rest and activity are identified. In the rest period, rumination and resting itself are included, while the grazing activity involves the time spent searching and fodder consumption (displacement and grazing). The knowledge of bovine behavior, including its activity pattern, is fundamental to apply concepts of animal welfare, even in

extensive systems. Data obtained from 9 GPS placed carried by grazing cows chose randomized at the herd were analyzed to inference activity by the speed of movement: five in natural grassland during spring and four in deferred *Digitaria eriantha* pasture at the end of winter. In stationary GPS, the error between registration can be utilized as a threshold for discriminate activity and no activity in animal behavior. The recorded speeds from animal monitoring were grouped into time ranges, and the stationary average speed was determined to be subtracted at its. For both grazing situations, the “activity period” of the breeding cows is concentrated in daylight hours, with a decrease in the period in the central hours. The animals grazing on grassland during spring-summer, show an activity period practically differentiated between morning and afternoon, with lower GPS speed in the hours of higher temperature and incidence of solar radiation (11:00 from 17:00 hs). On mega thermal pasture evaluated at the end of winter, the activity pattern is quite similar, but the activity begins more gradually in the early morning. The activity period in the afternoon behaves the same as in the morning but shows a marked decrease around 1 pm for a shorter period of time with respect to pasture behavior: approximately 1 h on the pasture and 3 h on the grassland. It is possible that in the pasture, as have higher productivity, cattle will graze some remnants in proximity to a water point, while in grassland paddock it will necessarily be moved to farther sites for grazing. It is necessary to continue with these evaluations, but it is possible to estimate the period of activity of grazing cattle from the analysis of the speed of its displacement, obtained by the GPS monitoring.

A183

SIG APPLIED IN THE EVALUATION OF SPATIAL AND TEMPORAL GRADIENT OF BOVINE GRAZING AT THE Paddock LEVEL

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The use of space is a widely studied aspect, both from the environmental ecology and by productivist approaches. The information that can be obtained from georeferenced locations of domestic animals such as cattle provides excellent accuracy and very important estimates can be made. The presence of animals is closely related to the use of the environment and when a particular spatial element that acts as an "attractor" of animals is considered, such as the location of the unique drinking water point in a pasture, it can generate a gradient of space utilization with respect to it. To perform an exploratory analysis of the cattle grazing gradient of the paddock, GPS equipment was placed on the neck of five cows on each paddock, in a low-weight harness that does not interfere with grazing livestock behavior. A 730 has grassland and 400 has deferred *Digitaria eriantha* paddocks were used. From an image obtained with Google Earth, the geo-reference of the paddocks in the center of the province of San Luis with corresponding water point location was carried, and through the free software Quantum GIS (QGIS), shapefiles of the spatial elements were generated. Equidistant (200 m) sectors were delimited concentrically from the location of the water and the georeferenced locations at each sector were counted, to evaluate a spatial utilization gradient from the animal presence. The frequency of records from the GPS device also allows a temporary analysis and by filtering the desired hours the information obtained can be increased. The area of each delimited sector was estimated and, using the relationship with the geo-referenced locations, the concentration or density of them was calculated to obtain an indicator of the grazing livestock in the grassland. In both forage resources, the concentration showed a similar response, and the function responded to a negative power or logarithmic function type, with an adjustment (R^2) greater than 65%. The greater adjustment was obtained with the model $y = 79.23x^{-0.688}$ for grassland ($R^2=0.85$) and $y = 40.018x^{-0.414}$ for pasture ($R^2=0.6616$). On grassland paddock decreased dramatically to 200–400 m, while on deferred pasture is not so steep the decline, until 400–600 m from water. The analysis of georeferenced information of cattle in grazing with GIS tools establishes the basis for investigations of gradients of use of temporary space at the paddock scale. Relationships with variables determined in the field will be useful for the integrative analysis of the interaction between cattle and space, in extensive livestock systems. GIS tools allow a spatial and temporal evaluation of grassland use at paddock level by cattle, providing safe, reliable, and detailed information of the grazing animal behavior.

A184

CO₂: EMISION FOR ONE YEAR IN A SAN LUIS WETLANDS

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Wetlands are flooded environments that can play another vital aspect as carbon sequestering due to low rates of organic matter mineralization. Our aim was to estimate CO₂ emissions over a year in Wetland “Bajo La Salada” physiognomy types. The sample area is located 5 km from Villa Mercedes town (San Luis) at 33°37' south latitude and 65°25' west longitudes, which is 505 m height above sea level. In each physiognomy type determined in previous work, we evaluated the C of CO₂ released by the soil in a closed trap of alkaline solution. Ten-day measurements were made in each season from winter 18 to autumn 19. A control sample, not closed, was placed in the halophyte open prairies (in the middle of the wetland) that correlate positively with the sums of all physiognomy types ($r=0.99$). The results of multifactorial ANOVA show significant differences ($P<0.05$) in emissions and cumulative emissions for each observed factor (station, days, and physiognomy types) and also in all their interactions. Each of the regression curves of the cumulative emission data of these physiognomic types in each station shows R^2 greater than 0.92 (for control

and 62,546 day). It is concluded that: (a) measuring CO₂ production was efficient by using the methodology used; (b) the results of the cumulative CO₂ and CO₂ values show significant differences ($P < 0.05$) in seasonal emissions, for every day and physiognomy types with a strong interaction between these factors; (c) there is a good correlation between the physiognomy types cumulative CO₂ and the observation days, adjusting to linear models; and (d) the open witness accumulated CO₂ showed a high correlation in each season with the sums of the cumulative CO₂ emission of all physiognomy types.

A185

VARIABLE STOCKING IN THE “CALDENAL” TO PRESERVE THE FORAGE RESOURCE

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San Luis "Caldenal" mixed pasture (summer and winter), currently has a low bovine receptivity due to woody species invasion that affects the pasture which competes for resources and restricts access to grazing. Traditionally, the "Caldenal" forage supply is consumed in the winter months coinciding with the winter grass critical moment where their nutritional reserves are affected and the stand of plants is reduced and allowed to rest in the months in which *Eragrostis curvula* (Schrader) is used from October to April of the following year. This situation favors the colonization of summer growth herbaceous in the areas released by the overgrazed winter pasture. As a means to sustaining the “Caldenal” resource within a local livestock approach and likewise to preserve floristic diversity, in the El Mollar Establishment a technical analysis of adequate livestock management was carried out for this purpose, which consisted of a case study, making a bibliographic survey on the productive characteristics to the Caldenal (forage production, time of use, natural grassland species) and its evolution. From this analysis it appears that there is a larger annual forage supply, leaving a remnant that can be used to support the system. A model of pastoral use of summer grass growth was proposed from November to the end of January with its own cattle. Another option is to lease the "Caldenal" to be used with high animal load, in order to free the area to favor the normal growth of winter grasses.

A186

BOTANICAL TOPONYMS IN THE SIL ÍPICA DEPARTMENT, SANTIAGO DEL ESTERO

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With the aim of recovering popular knowledge about the names of places in the departments crossed by the royal way alluding to plant species, their properties and uses, work was carried out in the Sil ípica department. To gather information on the toponymy of the area, the data were collected through bibliographic and map consultation, surveys of key informants in the area, and ethnobotanical walks. The species mentioned before were collected for determination, herbalized, and photo recorded. With the data obtained in each locality and from the bibliography, files were prepared for each species, with the purpose of returning to the communities. There are actually five places on the royal way in the department with plant names: La Higuera, Chañar Pujio, Simbol, Coro Pampa, and Gramilla, predominating the designations that come from the herbaceous stratum. Most of the respondents know the species that give names to the toponyms, but they do not know the reason why they were given, except the residents of the Simbol; the villagers report that these plants still inhabit these places and know where they are. With regard to the uses or properties assigned to them, they are generally known, although they only continue to use them in two locations; when comparing the uses or properties mentioned with those obtained in literature, it is appreciated that there is a loss of consciousness. Although people know in general the plants that give the name to their places, most of them do not know the reason why they were awarded and the use of these species is currently reduced.

A187

EDAPHIC, BENTHIC AND PHYTOPLANKTONIC DIATOMS OF SALINAS DEL BEBEDERO

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Diatoms form a group of siliceous microscopic algae that develop in a wide variety of micro-habitats. They are among the major primary producers and serve as valuable environmental indicators, commonly used to monitoring water quality. Their spatial and temporal distribution allows categorizing the type of ecosystem they inhabit. Our goal was to analyze edaphic, benthic and planktonic diatoms taxocenosis of Salinas del Bebedero, during the dry season of 2018 and 2019. The planktonic samples were obtained with a phytoplankton net, benthos samples by scraping 5 cm² area stones, and soil samples with a shovel and a hole. Physicochemical parameters were recorded using portable sensors. Identification was performed using an optical microscope with a magnification of 400X and 1000X. From the qualitative observations, results obtained were that the pennate forms predominated over the centered ones, being also found common and characteristic species of each habitat. Diatom assemblages planktonic were *Gyrosigma* and *Surirella*, the benthic were *Halamphora* and *Fragilaria*, and in soil *Epithemia* and *Campylodiscus*. Benthic diatoms were more diverse because they secrete a mucilaginous substance that, in addition to providing substrate binding, allows them to slide over the surface. This slip is distinctive of pennate form diatoms with raphe. Diatom assemblage changes study provide an

excellent basis for inferring environmental changes as well as let to know the degree of interrelation and distribution established in extreme environments.

A188

CHARACTERIZATION AND PROPAGATION OF *POROPHYLLUM OBSCURUM* (SPRENG.) DC. IN LOMA BLANCA DEL MORRO (PROVINCIA DE SAN LUIS)

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Porophyllum obscurum belongs to the Asteraceae family and is a native herb or sub-shrub, perennial and gynodioecious species. In Argentina, it is distributed from the north to the center of the country and it can be found in Uruguay, Paraguay, and Brazil too. Its name derives from the Greek “porus”, which means pore, and “phyllon”, which means leaf, in reference to the leaves glands present in much of the genus species. It is commonly known as “deer grass” due to the strong smell, similar to deer meat, which is produced by the aromatic oils emitted when plants are stepped on the field or their leaves are scrubbed. It has diaphoretic and antispasmodic properties. Loma Blanca del Morro is a grassland and mountain range area, with stony soil and rainfall between 500 and 600 mm per year. In general, deer grass grows on dry, sandy, saline, or stony soils. Deer grass large populations were not observed at the sampling site, probably due to medicinal herbs connoisseurs’ extraction. In order to preserve this species, the specimen’s phenology and morphology were recorded in the field and in the laboratory methods of sexual and asexual propagation were tested. Two populations were identified so that two sampling sites were determined, (1) “the hill” at a higher height and with a greater proportion of rocks and (2) “the bottom” at a lower height and with a greater proportion of soil. In “the bottom” 6 plants per m² were registered for the first transect and 20 plants per m² for the second, in “the hill” 6 plants per m² were registered in both transects. The specimens in the site reach a height ranging from 5 to 12 cm. The leaves are linear, up to 4 cm in length. It begins to bloom at the end of October, the full bloom is observed at the end of November and it continues to bloom and bear fruit until the end of April. In the laboratory tests to evaluate the sexual propagation, the fruits were selected for sowing, obtaining a germination value of 70%. Those harvested at “the bottom” site showed greater germination power both in the Petri dishes sowing and in the site substrate. For the purpose of vegetative multiplication evaluation, herbaceous and semileinous cuttings were made, and both types were treated with different rooting hormones obtaining a low percentage of survival.

A189

MORPHOLOGICAL CHARACTERIZATION OF RECENT POLLEN GRAINS FOR THE COMPARATIVE STUDY WITH FOSSILS AND ITS IMPLICATIONS IN *EPHEDRA* L. (GNETALES) DIVERSITY

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Many authors agreed that the study of recent pollen grains contributes to the taxonomy of some vegetal groups. On the other side, comparative studies with fossil pollen grains had allowed reconstructing paleoflora and paleoclimates. The primary objective of this work was to analyze pollen grains of recent South American species of *Ephedra* and to compare them with fossil pollen grains recovered from La Cantera Formation in San Luis province (Early Cretaceous) that were assigned previously to this group. The pollen grains were removed from specimens deposited in an herbarium and stained with basic fuchsin. A minimum of 20 pollen grains was studied per sample, using light binocular microscopy (1000X) and were compared with the fossil palynomorphs. Fossil palynological samples used were located in the paleopalynological collection from IANIGLA CCT from Mendoza. The analysis showed that both morphological and morphometric characters of the grains were uniform in each of the recent species and did not correspond to individual differences. The main shared characters between species were: ellipsoidal to fusiform shape and polylicate sculpture. These results support the previous idea that the great diversity in morphology and size of pollen grains assigned to *Ephedra* and recorded in La Cantera Formation indicating the presence of several species or even several genera in this association. This example shows a case where the study of recent species gives important support to infer the diversity of species that lived in the past.

A190

COMMUNITY ANALYSIS OF CHENOPODIACEAE FAMILY IN SAN LUIS, ARGENTINA

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The members of Chenopodiaceae are important components of the flora and vegetation of the arid regions of the world. The family includes nearly 100 genera and 1400 species. A great number of the halophytes and xerohalophytes of the world belongs to this family. Chenopodiaceae family in San Luis province includes a great variety of species, some of them, natives, poorly studied, and

others being considered as agricultural plagues, and cause of pollinosis in humans and animals. The majority of the non-native species are invaders, being characteristics of disturbed sites. In these places, members of this family form vegetal communities that associate them. The purpose of this work was to study the communities dominated by this family in San Luis city and nearby. The methodology included prospecting study areas in order to identify the study zones: center, suburbs, river, and salinity sites next to Salinas del Bebedero. A total of 32 phytosociological inventories were made, according to the methodology of Zurich-Montpellier school. It was registered a total of 71 species, 19 of them being Chenopodiaceae. The most characteristic communities in San Luis city were the quinoa species, being dominated by *Chenopodium album* and *Chenopodium hircinum*, and communities of white quinoa dominated by *Chenopodium borbassi*. In the suburban areas, especially in marginal lands as border areas close to roads, were prevalent the “cardales” community dominated by *Salsola kali*. The river zone was dominated by *Dysphania ambrosoides*. In salinized areas were registered communities of Chenopodiaceae dominated by: *Sarcocornia neei*, *Heterostachys ritteriana*, *Allenrolfea vaginata*, and *Atriplex lampa*, following a salinity/humidity gradient. It was concluded that in the study area the Chenopodiaceae community is highly frequent in disturbed areas and/or with salinity. Their distribution and phenology must be under consideration in relation to control their expansion, and allergy problems caused by pollen. On the other side, potential uses as tolerant species in degraded lands should be under consideration as a source of biomass for areas under disturbances.

A191

A GREEN AREA (SAN MARTÍN PARK) PROPOSED AS A BIODIVERSITY HOTSPOT OF TARDIGRADES IN THE CITY OF SALTA

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In urban landscapes, fragmentation is generally considered as a major threat to biodiversity, dramatically affects landscape structure, and decreases landscape connectivity. Thus, fragmentation limits the dispersal of many species and strongly shaped metacommunities. Urban green spaces are an important contributor to maintain biodiversity and enabling the sustainable development of cities, as they can increase the connectivity of the landscape. This work was carried out under the hypothesis that the green area "San Martín Park" behaves like a biodiversity hotspot of tardigrades in the City of Salta. A total of 12 samples were taken, each consisting of nine subsamples of lichen pads. Samples were taken from San Martín Park (P) and from two surrounding urban areas: nearby (N) and far away (F). Specimens were treated according to the usual study methodology. Data analysis was performed using PAST, PC-Ord, iNEXT, SPADE, and R programs. A total of 776 specimens belonging to 14 species/morpho-species were identified. The inventory completeness obtained was very good, over 95%. The F habitat had a greater abundance (N=405) but the P habitat was 1.09 and 2.24 times more diverse than the N and F communities respectively. The structures of the assemblages differ in the three communities, P and N communities were more evenness, presenting a greater diversity of guilds. On the other hand, the F community showed a strong dominance of only one species: *Milnesium sp3* (tardigrade group). The partition of beta diversity (BSOR) showed a gradient of species turnover (βSIM) from P to F. The greatest turnover (77%) was observed between the P and F communities, while the difference in the composition of the assemblages in N and F was due to both components: turnover of species (55%) and nesting (βSNE) (45%). In this study, we concluded that San Martín Park has a greater diversity of tardigrades, while the communities that are far away from this green area show a tendency to form a general nested pattern and a loss of species, leading to biotic homogenization in urban areas.

A192

PRELIMINARY STUDIES OF ENVIRONMENTAL IMPACT QUOTIENT FIELD (EIQ-FUR) QUANTITATIVE EVALUATION IN “EL MORRO” BASIN FARMS

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“El Morro” basin is located in the eastern region of San Luis province where agricultural, livestock and mixed activities are developed. The use of pesticides is a usual practice among farmers due to the prevalent pest and disease problems. One of the indices to assess pesticides’ environmental impact is the Environmental Impact Coefficient (EIQ). Issues such as toxicity (dermal, bird, chronic, bee, fish, and beneficial arthropod), soil half-life, systematicity, leaching potential, plant surface half-life, surface loss potential, farmworker, consumer, and ecological effects have been considered in EIQ estimation for a particular pesticide. The result is a single number describing the EIQ of a pesticide active ingredient. The EIQ field (EIQ-FUR) was developed to account for different formulations of the same active ingredient and different use patterns. The objectives were to estimate and compare the environmental impact quotient field (EIQ-FUR) of the crop, livestock, and mixed production systems. A baseline survey was conducted in September 2019. For data collection on pesticide usage, four farms (5.883 has) were selected. Farmers were previously consulted in order to provide information for the survey questionnaire design. Then, the EIQ value was established for the active ingredient of all pesticides that are used in the basin and so the EIQ field use ratings could be calculated by multiplying the EIQ value by the specific chemical, formulation percent active ingredient and formulated product rate used (EIQ field= EIQ value specific chemical x formulation % active ingredient x formulated product rate per hectare). To compare pesticides and production systems, the dose, product active ingredient formulation, and each pesticide frequency of application are needed to be considered. Our results show that the agricultural systems presented EIQ-FUR values higher than the livestock and mixed systems. Agricultural

activities used a greater number of chemical compounds (11 molecules) that increased the EIQ-FUR. On the other hand, we have observed that 36% of the active ingredients used reach high values due to the ecological effects they produce. Similar doses of the active substances were used in all systems. These results will be useful in the development of alternative or complementary strategies to improve the ecological, economic, and social sustainability of farmers. More surveys will be required in order to complete the study of the “El Morro” basin.

A193

BIODEGRADATION OF CHLORPYRIFOS FOR NATIVE MICROALGAE OF MENDOZA LAKES

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Pesticides go downstream to the aquatic ecosystem causing an imbalance and alteration in the way of living of the communities settled there. Chlorpyrifos (CPF) is one of the most commonly used organophosphorus insecticides in Argentina, although in 2000 the United States Environmental Protection Agency (USEPA) rigorously regulates its use. Microalgae play a vital role in ecosystem stability, which is why they are the first to respond to a change in their environment. Under certain conditions, some microalgae can develop in environments where autotrophic metabolism is not viable, so heterotrophic cultures can be used to study some metabolic aspects of them. Previous works show that these organisms react quickly to environmental changes caused by organophosphorus contamination. This is why the aim of this work, is to evaluate the biodegradation of the CPF by a native strain Chlorophyta under two different incubation conditions, light and total darkness. Therefore, a unialgal culture was made in Bold Basal (BBM) medium from an isolated strain of an urban lake in Mendoza province and was harvested in its exponential growth phase. Stock solutions of CPF were prepared from commercial formulations, and 10 ppm were added to the algae culture. These cultures, with and without CPF, were incubated at 30 °C for 6 days in light and darkness conditions, under continuous agitation. The biodegradation of CPF was monitored employing HPLC-DAD. The results demonstrated that the biodegradation in darkness is 70% more effective than in light conditions. It was also found that the occurrence of sub-products of biodegradation is below the detection and quantification limits.

A194

COWS CORPORAL CONDITION AS INDICATOR OF THE BREEDING SYSTEM PRODUCTIVITY IN SAN LUIS

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In a breeding herd, the production objective is to have one calf per cow each year, which is mainly influenced by the cow's nutritional status and the presence of the nursing calf, among other factors. An indicator of the breeding cow's nutritional status is the corporal condition (CC). In systems that forage base is degraded semi-arid natural grassland, the low availability and forage quality causes restrictions on dry matter consumption and depresses the herd CC, that impacts on the system productivity. The pregnancy rate (PR) is a good indicator of this effect. It was proposed to determine the CC, PR, and calves weight (a) during weaning in different cow herds (mature and first calving), and (b) from calving to the end of the service, indicating an order of herd productivity for a breeding system with sandy grasslands. The experience was realized in an extensively farmed livestock located in the sandy area with pastures and isolated chañar woodiest of San Luis, 40 km southwest of Villa Mercedes. The herd 1 were cow's leader of calving (September), with conventional weaning (150 days, end of February), herd 2: cow's body and tail of calving period (October–November) with conventional weaning (150 days, end of March), herd 3: cows at the second mating (December–January), leader of calving (September) and early weaning (60 days, December). The CC (scale from 1 to 5) was determined between calving and the end of the mating period, which began in December, and finished at end of February for herds 1–2, or in January for herd 3. Herd 1 have CC: 2.8 (start of calving period), 2.9 (start of mating), 2.6 (end of mating), with greater calf weight (156 kg) and lower PR (73%). Herd 2 showed CC: 2.9 at calving start, 3 and 2.8 at start and end of mating, respectively, with 81% PR. Herd 3: cow of higher nutritional demand, with similar CC from herds 1–2 at the beginning of calving and mating period, but greater at the end of the service (3.1), with less calf weight (100 kg) and greater PR (88%). The non-parametric Kruskal–Wallis test was performed and differences between CC herd medians were determined, in correlative order ($P < 0.05$). The analyzed system shows a herd increasing productivity order due to changes in CC between calving stage and the end of mating, weaning type and PR, (1) cows of early calving (end of winter) with conventional weaning, (2) cows with late calving (in spring) with conventional weaning, and (3) cows of early calving and weaning, at the expense of weaned calf weight. Tools such as early age weaning, allow improving CC in nutritional restriction conditions and enhancing system productivity.

A195

BIOFILMS FROM EL VOLCÁN STREAM, SAN LUIS, ARGENTINA: ITS EFFECTS IN INSECT DEGRADATION

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In order to reconstruct the processes of degradation of certain organisms in the water and compare them with past processes, this study starts with the determination of the composition of the biofilms formed around dead bodies of aquatic insects (Heteroptera Suborder: *Belostoma bifoveolatum*) collected from El Volcán stream, close to San Luis City. An experiment was assembled where the bodies of insects, killed by freezing, were placed in Petri dishes with a known volume of water from the stream and underwent five treatments, in order to determine the microorganisms associated with the degradation of the insects. The treatments included algacide, antibiotic, and fungicide; the control and four combinations of the three substances, which allowed inhibiting or favoring the growth of certain microorganisms. During the 35 days of the experiment, water samples corresponding were taken to each treatment, as well as qualitative parameters data and were cultivated with specific culture medium (EMB, MC Conkey, MSA, EC medium). The observation of microorganisms from the biofilms was made by Optical Microscope. On the other hand, samples were taken and dehydrated to be observed at the SEM. The results indicate that the organisms that were found forming the biofilms belong to the following groups: “algae” Bacillariophyceae and Zygnematales; Fungi belonging to the Zycomycota and Ascomycota groups, and Eubacteria of the Staphylococcaceae group and Enterobacteriaceae. The treatment in which the growth of Fungi predominated, was the one in which there was less degradation of the organisms used as an experimental model and greater development of the biofilm around the specimen. These observations will be useful in future studies to evaluate the degradation rate of certain organisms in current aquatic environments.

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