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1. METFORMIN TOXICITY AND EFFECT ON TUMOR GROWTH IN CBI MICE CHALLENGED WITH M-406 MAMMARY ADENOCARCINOMA

Capello Gardenal MC, Cáceres JM, Pagura L, Mainetti LE, Rico MJ, Rozados VR, Scharovsky OG¹.

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Metformin is a biguanide used as an oral diabetes medicine. Recently, its inhibitory effect on tumor growth has been informed. Our aim was to explore the likeliness of metformin use in the metronomic treatment of M-406, evaluating its toxicity and effect on the tumor. Inbred CBI mice were challenged s.c. with M-406 (Day 0) and when the tumors reached $\cong 100\text{mm}^3$ volume were distributed in three groups: G (Group) I: Control, no further treatment; GII: received metformin in the drinking water (0,2 mg/ml) and GIII: ídem GII with metformin (0,4mg/ml). Tumor volume, body weight and metformin mean daily intake/animal were determined 2 times/week until mice euthanasia. Glycemia was determined on days 7, 14, 21. Tumor growth curves were adjusted to the exponential model $Vt=S.e^{(k \cdot t)}$, [Vt =tumor volume (mm^3) in time= t (days); S =time of latency constant; k =exponential growth rate] and tumor duplication time was calculated ($TD_{\text{Dup}}=0,69/k$). No differences in TD_{Dup} were observed between groups (GI: 3,15 days, GII: 3,63, GIII:3,52; median). No changes were observed within and between groups in glycemia, body weight and survival. Metformin chronic administration, in the studied doses, would not produce side effects and its antitumoral effect could be evaluated in combination with other drugs administered with a metronomic schedule.

2. BLOOD BIOMARKERS FOR THE EVALUATION OF RESPONSE TO METRONOMIC CHEMOTHERAPY (MCT) WITH CYCLOPHOSPHAMIDE (CY) AND CELECOXIB (CEL) IN PATIENTS WITH ADVANCED BREAST CANCER (ABCP)

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Tumor and metastasis growth depend on angiogenesis. Hence, circulating endothelial cells (CECs), circulating endothelial progenitor cells (CEPs), vascular endothelial growth factor (VEGF) and thrombospondin-1 (TSP-1) could be useful non-invasive biomarkers of response to antiangiogenic treatment. MCT (administration of low drugs doses, at regular intervals, without long rest periods) with Cy+Cel would inhibit tumor angiogenesis. Our aim was to evaluate the levels of CECs, CEPs, VEGF (serum /plasma) and TSP-1 as possible biomarkers in ABCP treated with CY, 50 mg p.o./day + CEL, 200 mg p.o. bid. Blood samples were taken every 15-30 days. The time of patients ($N=13$) permanence varied from 4 to 64 weeks (median=13). The % of CECs was not associated with treatment response, while the % of CEPs increased significantly ($p=0,008$) in non-responder patients. VEGF serum concentration decreased as a function of time ($p=0,004$) and so did the plasma concentration, but not significantly; TSP-1 did not show modifications. Patient with higher permanence in the trial had the lowest VEGF/TSP-1 ratio. The significant decrease of serum VEGF supports the antiangiogenic nature of the treatment; the increase of CEPs % would indicate lack of response; the lower VEGF/TSP-1 rate could anticipate a better response to treatment.

3. TNF α AND IL-1 β DETERMINATION IN ADRENAL GLANDS IN IL-6 DEFICIENT MICE DURING ACUTE INFECTION WITH *Trypanosoma cruzi*

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Interleukin (IL-6) is a multifunctional cytokine involved in inflammatory response and hypothalamic-pituitary-adrenal axis regulation. It is a common finding in inflammatory processes, dissociations between corticotropin, antidiuretic hormone (ADH) and corticosteroids (CT) in blood which postulates the existence of a local circuit in adrenal glands (GA). In IL-6 deficient mice (KO6) in C57BL/6 background (C57) a paradoxical increase in body weight with decreased food intake is observed during acute infection with *Trypanosoma cruzi* (Tc) which aims to water retention in this model. CT, ADH, TNF- α and IL-1 β serum levels were similarly increased in both infected groups. IL-6 was only increased in C57-Tc. At GA, content (pg / ml) and concentration (pg / mg tissue) of the first two ILs were decreased in the C57-Tc group compared to non infected C57 (C) (IL-1 β : Content: C: 944 \pm 99 vs Tc: 850 \pm 43 *; concentration: C: 241 \pm Tc vs 34: 154 \pm 14 *; TNF- α : C: content: 341 \pm 31 vs Tc: 268 \pm 23 *; concentration C: 87 \pm 12 vs Tc: 49 \pm 5 *). Although CT and HAD serum levels were similarly increased in both infected groups, the absence of IL-1 β and TNF- α decrease in the GA in KO6-Tc mice infected may be associated with the fluid retention observed.

4. EFFECT OF A SEQUENTIAL THERAPY WITH MONOFLUOROPHOSPHATE (MFP) AND ZOLEDRONATE (Z). ON THE CORTICAL BONE

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Bone remodeling is a mechanism to maintain bone structure. MFP increases bone formation and Z inhibits bone resorption. Sprague Dawley rats were divided in 5 groups ($n=7$): 1. Sham; 2. OVX (ovariectomized); 3. OVX1: normal Ca diet treated with MFP from day 31 to 90 (400 $\mu\text{mol/Kg.day}$) and Z from day 91 to 150 (1.5 mg/Kg month); 4. OVX2: low Ca diet (0.2%) from day 0 to 30, high Ca diet (2%) from day 31 to 150 without drug treatment; 5. OVX3: low Ca diet and high Ca diet and treatment with MFP and Z as OVX2. At 150 days BMD in OVX1 and OVX3 groups were 13.79 and 9.76% respectively, compared with OVX. The trabecular bone increased in OVX3 and decreased in OVX1 (22.5 \pm 3.3 and 13.4 \pm 1.0%). These contradictory results may be due to changes at the cortical level. The aim of this work was to study the effect of the mentioned treatments at the level of cortical bone. At the end of the experiment 1 mm transverse cuts of left tibia were made at 50% of the diaphysis to analyse cortical parameter. At 30 days no significant differences were found. At day 90 OVX3 showed an increase compared with OVX in: periosteal perimeter (mm) OVX= 8.87 \pm 0.13 vs OVX3= 9.88 \pm 0.2; cortical width (mm) OVX= 0.61 \pm 0.01 vs OVX3= 0.71 \pm 0.01; and cortical area (mm²) OVX= 3.92 \pm 0.1 vs OVX3= 5.06 \pm 0.18 (medias \pm SEM). These differences were not maintained up to 150 days. We conclude that sequential treatment with MFP and Z in rats with increased bone remodeling does not produce changes at the cortical level and this cannot explain the increased BMD observed in OVX1.

5. THE INFLAMMATORY INFILTRATE AS A MARKER OF RESPONSE TO THERAPY IN PRIMARY TUMORS OF BREAST CANCER PATIENTS

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Different types of tumors, including the mammary ones, usually show both intra (ILI) and peritumoral lymphocytic infiltrate (PLI). There is evidence that in tumors from the oral cavity, lung, gastric and malignant melanoma, the intensity of the PLI is related to a good prognosis of the illness. Our objective was: a) to analyze the lymphocytic infiltrate (LI) and the expression of CD4 and CD8 molecules, markers of T lymphocytes subpopulations, in primary tumors of breast cancer patients and, b) to relate them to the clinical evolution (free of illness FI, or relapsed R) after 5 years of the 1st treatment. The archive samples analyzed were, mainly, stage I and II ductal mammary tumors. It was determined by immunohistochemistry, and confirmed by confocal microscopy, the expression of CD4 and CD8 molecules, in ILI and PLI. The LI was evaluated by Hematoxylin-Eosin stain. The quantification was performed in 20 high power fields, utilizing a 0 (negative) to 6 (high) score. A higher ILI was observed in FI patients (n=16) (median [range]:4 [0-6]) when compared to R patients (n=4) (2[0-2]), (P=0,018). The score of CD4⁺ and CD8⁺ cells in the LI showed wide variations. The presence in the 1st tumor of an intense LI could be a marker of good response to treatment, at least during the first 5 years after surgery. Increasing the number of tumor samples analyzed would allow suggesting its use as a marker in the clinical practice.

6. STUDY OF TUMOR GROWTH IN CONDITIONED MEDIA OF MONONUCLEAR CELLS FROM DIFFERENT LINES OF MOUSE

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M-406 mammary adenocarcinoma appeared spontaneously in CBI mice line. CBI line is used as control of an experiment of artificial selection for body conformation from which CBI and CBI/L was derived. When CBI is challenged with M-406, the tumor grows exponentially showing 100% progression. In CBI mice the tumor grows in 100% of the animals and then returns. In CBI/L mice tumors present the three phases of immunoediting: elimination, equilibrium, and escape. Our aim was to evaluate M-406 tumor growth in conditioned media of mononuclear cells from CBI, CBI/L and CBI lines. M-406 cells (5x10⁴ cel/100µl) were cultured at 37°C and 5% CO₂ in conditioned media of mononuclear cells from different lines of animals that had not been in contact with the tumor: CBI (Mc-CBI), CBI/L (Mc-CBI/L) and CBI (Mc-CBI). As a control M-406 cells were cultured in RPMI 1640 + 10% BFS (Co), considered as 100% proliferation. After 24 and 48h incubation, cell proliferation was assessed using the WST-1 colorimetric assay. The results showed that after 24 hours, tumor cells proliferated more in Mc-CBI compared with Co (P<0.05) with no differences in the Mc-CBI/L and Mc-CBI. At 48h tumor growth showed the following behavior according to the medium: Mc-CBI>Mc-CBI/L>Mc-CBI (P<0.0001). It is concluded that mononuclear cells of CBI and CBI/L lines would release factors that stimulate tumor growth. The comparative study of different factors present in the conditioned media could offer some mechanisms that come into play when animals of these different lines were challenged with mammary adenocarcinoma M-406.

7. METABOLIC PROFILE OF FLEXOR DIGITALIS SUPERFICIALIS MUSCLE IN SELECTED MICE (Mus musculus)

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The aim of this research was to analyze variations in the metabolic profile in flexor digitalis superficialis muscle in selected mice lines. The selection use a biological index which combines body weight and skeleton length in an agonistic selection (CBI+: high body weight and long skeleton; CBI-: low body weight and short skeleton) and in antagonist selection (CBI/L: low body weight and long skeleton; CBI/C: high body weight and short skeleton). CBI control unselected line. Six adult males of each line were killed by overexposure to CO₂. Mm. flexor digitalis superficialis were identified, extracted and frozen in acetone cooling to -80°C. The muscles samples were cut in cryostat. The muscle oxidative capacity was assessed by means of NADH reaction. The effect of the genetic group on optical density (OD) measured with an image analyzer was analyzed with a one way analysis of variance. Selected lines showed higher OD mean values than controls. No significant differences were observed neither between lines within selective procedure (CBI- vs. CBI+; CBI/L vs. CBI/C) nor between lines selected for low body weight (CBI- vs. CBI/L). Lines selected for high weight differed in DO mean values (CBI/C > CBI+). It is concluded that independently of the direction of the selection pressure applied, muscles of selected lines exhibits a greater oxidative capacity than muscles of the control line. At the same body weight, mice with short skeleton show more oxidative muscles that those with long skeleton.

8. EGGSHELL CALCIUM AS A DIETARY SUPPLEMENT

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Calcium (Ca) content in the diet is usually deficient. Ca intake from dairy products is not often adequate and the supplementation with tablets has high cost and difficulties of adherence to treatment. The aim of this work was to study the mineral content of the egg shell, the bioavailability of Ca and ways of processing for the addition to foods. We evaluated 16 eggshells, which were dissolved with commercial vinegar and determined the mineral content as percentage of the dry weight of the eggshell (% mean±SD). We determined the content of Ca and strontium by atomic absorption spectrophotometry, phosphate by UV spectrophotometry, fluoride by direct potentiometry, sodium and potassium by flame photometry and sulfate by precipitation with barium. Carbonate was measured by the moles of CO₂ produced. Organic matter was determined by weight difference of the shell before and after incineration. The bioavailability was evaluated in adult Sprague Dawley rats. Ca was supplied with Ca carbonate (control) or eggshell powder (treated). Ca absorption was calculated with [(Ca intake - Ca in feces) x 100]/Ca intake]. Results: Organic matter: 16.2±4.6%; inorganic matter = Ca: 38.2±3.5, carbonate: 44.3±3.2, sodium: 0.51±0.09; phosphate: 0.44±0.06; sulfate: 0.32±0.07; potassium: 0.14±0.05; strontium: 0.14±0.02, fluoride: 1.8x10⁻⁶±1.1x10⁻⁶. A complete eggshell has 2.02±0.42 g of Ca and can be dissolved with 215±32 ml of vinegar in 48 hours. Ca absorption from the eggshell (58.3±16.9%) is not different from Ca carbonate (62.1±16.0%). We conclude that the eggshell is a good supplement of Ca, and easy to prepare.

9.

METRONOMIC CHEMOTHERAPY (MCT) WITH CYCLOPHOSPHAMIDE (CY) AND DOXORUBICIN (DOX) IN THE M-406 MOUSE MAMMARY ADENOCARCINOMA TUMOR MODEL. MECHANISM OF ACTION

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MCT consist of the chronic administration of low drugs doses, at regular intervals, without extended rest periods. DOX avoids DNA replication. CY, an alkylating drug, has toxic action on proliferating cells. We have demonstrated the inhibition of M-406 and M-234p mammary adenocarcinomas growth by MCT with CY + DOX. Our aim was to study the mechanism of action by which such an effect is achieved. Inbred female CBI mice were s.c. challenged with M-406 (day 0) and on day 10 distributed into 4 groups (G). GI: Control with no further treatment; GII: Treated with Cy 20mg/kg/day in the drinking water; GIII: Treated with DOX i.p. 0,5mg/kg de Dox 3 times/week; GIV: Treated as GII + GIII. Mice were weighted and tumor volume measured thrice weekly. On days 0 and 26 blood samples were taken for evaluation of hematologic toxicity, % of T regulatory cells (Treg; flow cytometry) and serum VEGF concentration (ELISA). Tumor apoptosis was determined by TUNEL. No hematologic toxicity was observed. The Tregs % did not differed among groups. VEGF concentration on day 26 was lower in the treated groups with respect to controls, showing GIV the lowest levels (Mean±SE, pg/ml; GI: 239,1 ± 65,8 vs GIV: 77,0 ± 6,64; P<0,05). The apoptosis level was higher in GIV than in GI (P=0,05). The inhibition of tumor growth observed in M-406 tumor in mice treated with MCT with CY + DOX could be due, at least in part, to tumor angiogenesis inhibition and apoptosis stimulation.

10.

VALIDATION IN SILICO, IN VIVO AND QUALITATIVE ANALYSIS OF A MATHEMATICAL MODEL OF THE HOMESTOSIS OF THE GLUCOSE-INSULINE SYSTEM

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Diabetes mellitus affects millions of people. New technologies for its control has led to the development of mathematical models. These models have a high number of parameters wich require complex methodologies for their estimation. In previous work we have presented a new simplified model of glucose-insulin system to the normal rat and a methodology for estimating and optimizing its parameters. This model has two differential equations and it has seven parameters, which represent the different organs and tissues. The objective of this study was to validate it in silico, *in vivo* and through mathematical analysis. In silico validation was performed with Simulink tool of MatLab. Expected variations were observed in plasma glucose and insulin levels when the parameters of insulin secretion, insulin dependent glucose uptake and glucose intake were changed. For *in vivo* validation Sprague-Dawley rats (n=3) were used and parameter of plasma insulin clearance was estimated by 2 different methods: using the model and by direct measurement of plasma insulin after an intravenous injection of the hormone. The values obtained showed no significant differences: for model: 0.176±0.148 min⁻¹ for insulin disappearance 0.104±0.132 (t-Student p=0,5). The qualitative analysis of the system revealed that the system in all cases has a single equilibrium point biologically consistent. We concluded that the mathematical model adequately reproduces the variations of blood glucose and blood insulin in normal rats, and it allows the simulation of system glucorregulatorio modifications.

11.

DEVELOPMENT AND VALIDATION OF A DEVICE FOR BIOMECHANICAL TESTS OF BONES

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Osteoporosis is a prevalent disease and the development of new therapies is constant. The verification of the efficiency of the therapy requires evaluation of bone strength. There is no national equipment to perform biomechanical tests. The aims of this work were to develop an equipment to study the mechanical properties of bone and the validation of the measurements. The equipment apply forces with a detection limit of 1N and can measure distances as small as 10 um. The system allows the calculation of biomechanical variables: ultimate load (N), fracture load (N), deformation (mm), stress (MPa), strain (µstrain), stiffness (N/mm), Young's modulus (MPa) and toughness (mJ). The fracture load (108±13) and stiffness (311±108) of femurs from 7-week-old rats were not different from values reported by others (fracture load:95±10, stiffness:300±25). Results are expressed as mean±SD and * indicate significant differences p<0,05 Student's t test. The device was tested with femurs of control rats and rats treated with 80 µmolNaF/100g bw that decrease the cross section of the shaft and the trabecular bone volume. As a consequence the resistance of cortical and trabecular bone is reduced in the NaF treated rats. Fracture load of trabecular bone of NaF-treated rats (18±10*) was lower than controls (52±28). On the other hand, the fracture load of cortical bone of controls (108±13*) was significantly higher than in NaF-treated rats (84±11), n=4/group. Conclusion: the equipment allows to measure variables for the interpretation of bone strength and the values obtained for these variables were consistent with reported values and with the expected values for the biological model tested in this work.

12.

TUMOR PROLIFERATION IN CONDITIONATED MEDIA OF MONONUCLEAR CELLS FROM CBI/L MICE IN THE DIFERENT PHASES OF TUMORAL GROWTH

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The M-406 mammary adenocarcinoma appeared spontaneously in a CBI female mouse and it is maintained *in vivo* in mice of the same line. Inbred CBI/L FS mice were artificially selected for body conformation from CBI mice. When CBI FS mice are s.c. challenged with M-406, they show exponential growth in 100% of the animals. When CBI/L FS mice are s.c. challenged with M-406 they show the three phases of the immunoediting theory: 51.6% in escape (ES), 18.5% in equilibrium (EQ) and 29.8% in elimination (EL). The aim of this study was to characterize the interaction of M-406 with the immune response of CBI/L animals. M-406 was cultured *in vitro* (5x10⁴ cells/100µl) in conditioned medium of mononuclear cells from animals with tumors in EL or ES, and from *naïve* animals. RPMI 1640+10% BFS was used as control. After 24h, proliferation was evaluated with WST-1 colorimetric assay. The proliferation percentage was higer in *naïve* mice followed by ES and then EL. No significant differences were found between control and EL. These results allows to conclude that there is any factor in the conditioned medium from *naïve* and ES that may stimulate tumor proliferation. Comparative study of the factors present in the conditioned medium could allow the identification of the mechanisms that are activated in the three phases of the immunoediting process when CBI/L animals are challenged with M-406.

13. ACUTE INFECTION WITH *Trypanosoma cruzi* IN IMMUNODEFICIENT RAG MICE WAS ASSOCIATED WITH INCREASED HPA AXIS ACTIVATION

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During acute infection with *Trypanosoma cruzi* (Tc) in C57BL/6 mice, corticosterone plasma levels (CT) were markedly increased. Mice deficient in recombinase activating gen (RAG) do not express antigen receptor and therefore have no functional T or B cell. We characterized the infection with Tc and the hypothalamic-pituitary-adrenal (HPA) axis response in mice deficient in this protein (RAGKO) and syngeneic controls (C57). Mortality was 100% in both groups, but RAGKO-Tc had significantly higher parasitemias (C57-Tc: 208.6 ± 50.3 vs RAGKO-Tc: 9948.6 ± 953.1 *), lost more weight and died earlier. Pro-inflammatory cytokines increased in both infected groups compared to controls, but IL-1β and IL-6 plasma levels were significantly higher in RAGKO-Tc (IL-6: C57-Tc: 254.9 ± 53.6 vs RAGKO-Tc: 6874.7 ± 1939.8 *, IL-1 β: C57-Tc: 7.8 ± 5.5 vs RAGKO-Tc: 61.6 ± 18 *). CT levels increased progressively in both groups and were higher in infected RAGKO-Tc (D18 pi: C57-Tc: 7.47 ± 1.02 vs RAG-Tc: 18.75 ± 2.73*). RAGKO only respond with innate immunity to infection and produce higher levels of pro-inflammatory cytokines in blood, which in turn activates a more forceful the HPA axis.

14. EFFECT OF HOST GENOTYPE AND SEX ON THE INFECTIVITY OF *Trichinella spiralis*

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Trichinella spiralis is a parasite infecting humans and a wide range of carnivorous and omnivorous mammals. The resulting disease, trichinellosis, is a zoonosis of worldwide concern. In spite of significant medical and technical advances, the prospect of eradicating this disease remains discouraging due in part to the complex relationship between parasite and host. Our objective was to analyze differences in the host-parasite interaction during the chronic phase of a primary infection with *Ts* in male and female mice of lines of the CBI-IGE stock with different resistance to the parasite. Young adult animals (95±5 days old) from lines CBI, CBI/L (L) and CBI/C (C) (n=15 per sex and line) were infected with a dose of 2 L1 larvae per g of body weight. Body weight was measured at the beginning of the experiment. Muscle parasite load in the chronic stage (32±2 days post-infection) was expressed as number of parasites per g of tissue (CPr) and the reproductive capacity index of *Ts*, ICRr, was calculated with the formula ICRr=CPr/infective dose. Body weight differs both between sexes within genotypes (p<0.001) and between genotypes within sex (p<0.0001), consequently the average infective dose for each group was different. No differences in CPr and ICRr were observed between sexes within lines, however, C males showed larger CPr and ICRr values than females (p=0.06). CPr was lower in L than in C and CBI (p<0.0001). L males had a smaller ICRr than C and CBI (p<0.0001) while L females only differed from CBI (p=0.0004). ICRr enabled classification of the hosts as susceptible or resistant irrespective of the infective dose. This variable also allowed us to detect a sex effect on the host-parasite interaction.

15. EFFECT OF QUERCETIN (NATURAL ANTIOXIDANT) ON CHANGES IN THE PROPERTIES OF THE ERYTHROCYTE MEMBRANE CREATED BY THE INTERACTION OF ALUMINUM WITH THE MEMBRANE

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Studies in our laboratory in red blood cells (RBCs) incubated *in vitro* with human AlCl₃ (Al) showed alterations in cell shape (CS) and osmotic fragility (OF), because of the oxidative stress generated by the metal. The flavonoids (eg quercetin (Qc)), polyphenolic compounds widely distributed in plant foods eaten by man, are an excellent source of antioxidants, that is efficiently captured by the RBC. We propose to study *in vitro* the possible prevention by the Qc of alterations in CS and OF in human RBCs caused by pretreatment with Al. Blood samples were obtained by venipuncture from healthy volunteers donors, over 21 years, from both sexes. CS was determined by microscopy and the OF by photometry at 540 nm. We reported: x50 (NaCl concentration which gets 50% of hemolysis) (n:10). Results: mean ± SEM. Statistical analysis: Anova. **Results:** X₅₀(mM) control 68,65 (+/- 2,20), Qc 66,51 (+/- 2,67), Al 81,84 (+/- 2,35), QcAl 69,43 (+/- 1,74). X₅₀ control, Qc y QcAl not differ between them but differ from Al (p<0,05). The biophysical alterations of RBC incubated with AlCl₃ can be counteracted by the antioxidant effect of preincubation with Qc. This allows us to propose that the incorporation of Qc at different levels of the lipid bilayer of the membrane prevents the interaction of Al with the polar heads of phospholipids thus preserving the discoid shape from the RBCs.

16. ANALYSIS OF LIPID PROFILE IN HIV (+) PATIENTS TRATED WITH DIFFERENT ANTIRRETROVIRALS OF HIGH EFFECTIVENESS. PHASE I

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The era of anti-retroviral treatment of high effectiveness (TAAE) began when proteases inhibitors were approved in 1996. The variability of lipid profile in HIV(+) patients with TAAE was analyzed. An observational study of 72 patients was carried out (2008 to 2011). Two treatment schemes with reverse transcriptase nucleoside inhibitors were developed. Patients' assignment: in Group I: zidovudine+lamivudine (AZT/3TC) 34 = T1: +efavirenz (EFV): 24, T3: + nevirapine (NVP): 10. In Group II: emtricitabine+tenofovir (FTN/TNF) 36 = T2: +EFV: 24, T4: + atazanavir+ritonavir: 10 and T5: + ritonavir+fosamprenavir: 2. Basal laboratory (lb) and post laboratory (lp), after 6 months of TAAE, were performed. A T-student test was applied to compare means in continuous variables (α=0,05). Results: They were expressed in mg/dL as mean±standard deviation for each treatment. T1: Total Cholesterol-tCol (lb)188±61; (lp)206±60. HDL (lb)38±12; (lp)42±9. LDL (lb)124±38; (lp)142±76. Triglycerides-TG (lb)136±72; (lp)155± 85. T3: tCol (lb)172±29; (lp)186±31. HDL (lb)34±11; (lp)38±10. LDL (lb)138±47; (lp)112±6. TG (lb)130±57; (lp)109±34. T2: tCol (lb)184±46; (lp)199±48. HDL (lb)39±32; (lp)34±8. LDL (lb)131±45; (lp)128±50. TG(lb)165±138; (lp)170±124. T4: tCol (lb)160±37; (lp)176±33. HDL(lb)30±6; (lp)31±9. LDL (lb)124±35; (lp)105±28. TG (lb)174±140; (lp)233±323. T5: tCol (lb)335±156; (lb)211±88. HDL (lb)29±6, (lp)37±12. LDL (lb)129±27; (lp)277±74. TG (lb)142±62; (lp)133±62. When providing AZT/3TC+EFV (T1) a significant increase of HDL was observed (p=0,02); no differences were seen neither in other laboratory results nor other treatments (p>0.05). Conclusion: After 6 months of TAAE, the studied patients show no changes in their lipid profiles, thus it could not be inferred that its variability has been due to TAAE.

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OSMOTIC FRAGILITY AND SHAPE OF RED BLOOD CELLS OF THE Hmb (b) LINE OF RATSRuiz MF², Casco C², Visconti M¹, Gayol M del C², Bollini A¹, Hernández G¹, Bazzoni G¹¹Cát. Física Biológica. ²Cát. Biología. Fac. Cs. Médicas. UNR.

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The inbreeding line b of rats was considered normal control of the line beta (obesity and diabetes model). Recently, the line b started presenting variability on glucose tolerance; therefore it was necessary to characterize it in different aspects. In 2009 we started their rheological characterization at 100, 200 and 300 days-old rats. The aim of this study is to compare at different ages inside a line and between the lines of rats the osmotic fragility (OF) of red blood cells (RBC), using photometry at 540nm which obtained the parameter x50 (NaCl concentration where 50% of hemolysis was obtained); and the erythrocyte shape (ES), by microscopy. U-Mann-Whitney test was used and the results were expressed as medians and ranges, considering significant if $p < 0,05$.

	100 days-old		200 days-old		300 days-old	
	b	beta	b	beta	b	beta
X50	74,76	67,05	74,20	81,34	73,56	86,80
nM	(58,06-84,19)	(58,25-88,16)	(67,72-81,67)	(78,09-86,41)	(70,96-78,48)	(79,41-98,77)
ES	stomatocytes 3, 4		stomatocytes 3, 4		stomatocytes 4, 3	

RBC of beta rats increased their OF ($p < 0,01$) with age but it does not happened with b line. At 100 days-old both lines presented the same OF; at the other ages RBC of beta line were more fragile ($p < 0,001$). No difference of the ES was found. The metabolic disorder in beta line influences the mechanical answer of the membrane of RBC but it does not modifies the ES, while glucose intolerance of b rats is not important enough to affect the RBC membrane.

18.

BREAST CANCER. MORTALITY IN ROSARIO BY DISTRICT (PERIODS 1996-2000 AND 2006-2010)

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Breast cancer (BC) is the leading cause of death from malignancy in women in most occidental countries, including Argentina. This work aimed to know the current mortality rates from this disease in Rosario, to compare with those obtained for an earlier period. We reviewed the certificate of deaths from BC of all women in the city. The periods 1996-2000 and 2006-2010 were studied. We calculated age groups specific mortality rates (AMR %₀₀₀) corresponding to the different districts and the city as a whole. We also calculated age standardized mortality rates (SMR %₀₀₀), applying the direct method. The SMR was 98.5 for the period 1996-2000 and 80.1 for the period 2006-2010. While in the North District, SMR were stable (69.7 and 60.2), it decreased in other districts (Center: 123.5 and 83.4, North West 72.0 and 43.5; South 78.9 and 41.7) and increased in others (South-West: 43.9 and 68.7; West: 50.8 and 104.6). AMR decreased in women older than 70 years was observed in Central, North and Northwest districts. In the South district, rates began to decline in women after 35 years old. Contrary, in the South-west AMR increased in women from 65 years was registered, and in the West rates increase in all age groups except 40-44 years old. In this district, the increase was found in the older age groups (65-69: 84.4 and 147.1, 70-74: 119.0 and 274.0, 75-79: 180.3 and 498.4, more than 80: 239.0 and 655.7). In conclusion, analytical epidemiological studies to detect factors that might indicate the reasons for the changes observed in different districts are necessary. Furthermore, the study of longer time periods would be very useful to evaluate the validity of these modifications.

19.

STUDY OF MINIMUM CONCENTRATION OF ANTIMICROBIAL FOR THE ERADICATION OF *Gardnerella vaginalis* BIOFILM ISOLATIONS

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The pathogenesis of many infections are related to the presence of microorganisms in biofilms (BF) *Gardnerella vaginalis* (GV) isolations are no exception. These BF are difficult to eradicate which is associated with antimicrobial (AM) treatment failures. The aim of this study was to determine the minimum concentration of: metronidazole (MTZ) and clindamycin (CLI) as AM capable of eradicating *in vitro* the BF formed by the GV (MCE). The test consists of: 1- Adherence and formation of the GV BF and 2-Observation and quantification of the BF eradication by the AM. We studied 5 strains of isolated GV from patients with bacterial vaginosis diagnosis (BV), confirmed by Amsel criteria and the Vaginal Content Balance study. All strains, once isolated and phenotipified as GV. Later we included a 100 µl of BHI+1% glucose (BHIg) in each well, in comercial 96 well microplates, and then added 100 µl of inoculum Gv (1.10^8 CFU/ml (D.O. 650). Incubation in anaerobic conditions at 37°C for 24hs. Later added 100 µl of CCCg and re-incubated for 24 hs. Then the wells were washed with 200 µl of PBS buffer three times, and we placed 200 µl (of decreasing dilutions) of MTZ (1024-1 µg/ml) and CLI (256-0.25 µg/ml) on each well. Again was reincubation for 24 hs at 37°C in anaerobic. After removing the contents from each cell were washed for three times with PBS and then dyed with 1% Safranin. The reading was at 450 nm. In 100% of the strains tested the (MCE) exceed 256 µg/ml for MTZ and 32 µg/ml for CLI. We propose an innovative methodology for *in vitro* study of an *in vivo* problem, thinking the GV as the main co-participant of BF in the BV. Given the results obtained, we intend to study a larger number of strains so we can link these new findings with failed treatments.

20.

SITUATIONS IN WHICH THE POPULATION SELF-MEDICATES WITH ANTIMICROBIALSLuciano M, Córdoba L, Braccia V, Carrión M, González Y, Hails I, Maurino, Paladini L, Pasquale J, Schwank S, Siffredi V, Gambandé T
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Introduction: antimicrobial (AM), used to treat infectious diseases are powerful tools in its eradication, but overuse is related to the selection of bacterial resistance mechanisms. This resistance is fueled by self-medication with AM. Objective: To present which symptoms cause people to self-medicate with AM. Methods: Closed surveys were conducted in the city of Rosario, which included 200 people, the data were statistically analyzed. Quantitative variables were described with means and standard deviations and percentages were calculated qualitative and frequencies. Test was used Chi-square Association. Results: Responses to multiple-choice question, which refers to situations in which self-medicate with AM, showed the following data: 39.5% (79) self-medicate with AM because of a sore throat, 36% (72) toothache, 28.5% (57) ear pain, 45% (90) fever, 40% (80) cold, 35.5% (71) urinary difficulties, 22% (44) skin infection and 10% (20) discomfort in the genital organs. Consumption in case of genital discomfort regarding sex of the respondent found significant differences ($p=0.03$). Discussion: We found that the more usual situation of consumption of AM is related to upper respiratory tract symptoms, which has usually viral etiology, so, their consumption is not useful, highlighting its benefits only as a placebo, favoring an increase in bacterial resistance and masking of symptoms. Regarding the discomfort in the genital organs, is more frequent the use of the AM is in male, due to the more pronounced symptoms of sexually transmitted infections (STIs) in men, being generally asymptomatic in women. Conclusion: From the perspective of both Microbiology and Health Education is important to collect and analyze data such as those offered in this study to incorporate behavioral changes in the population.

21. PREVALENCE OF UROPATHOGENS ISOLATED FROM PREGNANT

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The anatomical and functional changes that occur during pregnancy favor colonization of the Urinary tract (UT) by microorganisms which live in the perineal region, phenomenon known as asymptomatic bacteriuria. This increases the risk of acute pyelonephritis, premature rupture of membranes, preterm labor and early neonatal sepsis. Urine culture is the best method to diagnose it. It allows early and appropriate treatment, avoiding complications. We propose to collect epidemiological information on the prevalence of uropathogens in pregnant women, regardless the clinical situation that they present. A retrospective study of urine cultures records in female patients processed by our service in the period between 2004-2008 was realized. From 9633 urine samples significant bacteriuria was detected in 1650 (17.13%) of which 1366 (82.79%) were women, of which 136 (9.96%) were pregnant. Samples were obtained by midstream and kept refrigerated until processing. They were cultured and identified by conventional methods (ASM Handbook). The microorganisms identified and their prevalence were: *Escherichia coli* 111 (81%), *Klebsiella pneumoniae* 12 (9%), *Proteus mirabilis* 7 (4%), *Streptococcus agalactiae* 3 (2%), *Enterobacter aerogenes* 1 (1%), *Klebsiella oxytoca* 1 (1%), *Enterococcus faecalis* 1 (1%), *Proteus penneri* 1 (1%). There were no significant differences in the prevalence of uropathogens when compared with our population of nonpregnant women. While in the international literature *E. coli*, *Klebsiella pneumoniae* and *Proteus mirabilis* are the most frequent, the prevalence of this vary significantly from those presented in this study. The microbiological diagnosis of significant bacteriuria is a non-invasive, neither onerous procedure and prevents serious complications and even death.

22. CROSSOVER DESIGN FOR ASSESSING MOTOR SKILL IN PATIENTS WITH MULTIPLE SCLEROSIS

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There is some difficulty in planning clinical trials for rare diseases. Crossover designs are used in clinical trials as they provide good accuracy with few experimental units and allow evaluating changes in response over time. In this design experimental units receive different treatments in different sequences. Although studies in which an observation is obtained for each unit are the most used, it is possible to develop a design with repeated measurements. Mixed models are useful for the analysis of such designs. This work aims to select a model to explain the evolution of motor skills of the not dominant hand in patients with multiple sclerosis. The data used comes from an experience in which 8 patients are stimulated by transcranial stimulation technique (tDCS) and a variant of it called "Placebo". The first group received in the first period tDCS and then Placebo; the second group received in first period Placebo and then tDCS. The measurement of the skill is performed at three different times for each patient. In crossover design with repeated measures it is necessary to model both the mean and covariance structure. Since the data show a different behavior after the second measurement, the evolution of each response is modeled as a linear function of time using a spline model with random intercept. It fits the model with different covariance structures. Several models are compared using the likelihood ratio or the information criteria (AIC or BIC). The time evolution of the skill is different for both groups and periods ($p < 0.0001$). For the Group 1, when stimulated with tDCS, the average skill decreases and then stabilizes, while in the second period increases slightly. For Group 2 when starting with Placebo average motor skill decreases, but in period 2 increases slightly. It cannot conclude, in general, about the existence of beneficial effect duration of the stimulation.

23. CAMR-SA: NASAL CARRIAGE IN CLOSED GROUPS OF THE COMMUNITY

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During the last years, Community Associated Methicillin-Resistant *Staphylococcus Aureus* (CAMR-SA) emerged bringing the infections caused by the *Staphylococcus* genus to a new stage. Its characteristics are resistance to cefalotin and oxacillin, without accompanying resistance, and high grade of virulence. Its nasal carriage has been reported in closed groups such as assisted livings, sportsmen who participate in contact sports, prisons, and kindergartens. These strains have emerged in Argentina; nowadays, cases of severe infections caused by this microorganism have been reported. OBJECTIVE: to study the CAMR-SA nasal carriage in closed groups of the community such as assisted livings, and volley, hockey, football, water polo and rugby teams in Rosario city and neighbouring localities. MATERIALS AND METHODS: 224 nasal swabs were taken; they were seeded in blood agar and incubated at 37°C for 48 hours. The characteristic colonies were identified through biochemical tests. The antibiotic sensitivity testing was performed on those which proved to be *S. Aureus* according to the NCCLS standards, in Muller-Hinton agar. RESULTS: From 5 assisted livings, 123 nasal swabs were analysed. CAMR-SA was isolated from 3 samples of a single assisted living (2.43%). The colonized people average age was 70 years old. CAMR-SA nasal carriage was not found in the 101 high-level sportsmen analysed. If we consider all the samples obtained, the CAMR-SA strains represented a 1.34%. CONCLUSION: Detecting nasal carriage in healthy people, preventive measures could be established in order to control this new aggressive *Staphylococcus aureus* strain. Likewise, the number of severe infections reported caused by CAMR-SA would be reduced. We insist on the appropriate use of antimicrobials, since the incorrect and indiscriminate use of it creates ever more virulent strains and shortens the therapeutic range in an infection context.

24. MINIMUM INHIBITORY CONCENTRATION (MIC) TO ERYTHROMYCIN AND CIPROFLOXACIN OF *Campylobacter jejuni* ISOLATED IN HUMANS WITH ACUTE DIARRHEA

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Campylobacter jejuni is in our environment one of the main causes of diarrhea. While most are self-limiting cases, others require medication. Macrolides are considered the drug of choice for intestinal cases and fluoroquinolones have been proposed for the empirical treatment in adults, even for traveler's diarrhea. Since 1985 we have observed an increase in quinolone resistance in strains from human and chicken. In order to conduct resistance surveillance, 27 clinical isolations of *C. jejuni* were studied between 01/01/11 and 01/09/11, in two clinical centers. The MIC values by E test. The breakpoints for erythromycin are: S ≤ 4 and R ≥ 8 and ciprofloxacin are: S ≤ 1 and R ≥ 4 . We found an erythromycin resistance rate of 15%. It was determined that the MIC of that antibiotic of 90% of the susceptible strains was found between 0.5 and 2 $\mu\text{g} / \text{ml}$. With regard to ciprofloxacin, 79% were resistant. The MIC of susceptible strains was found between 0.06 and 0.25 $\mu\text{g} / \text{ml}$. We conclude that the empirical treatment with ciprofloxacin of acute diarrhea caused by *Campylobacter* in adults has no value. In contrast, erythromycin remains the drug of choice in pediatric patients, if we get a concentration between 0.5 and 2 $\mu\text{g}/\text{ml}$.

25. ACQUISITION OF CONTENTS AND ABILITIES ON BIOSECURITY PRINCIPLES IN BIOCHEMISTRY AND THE VALORIZATION OF MEDICINE 2ND YEAR STUDENTS

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Biochemistry is an annual subject of 2nd year of Medicine. A biosecurity workshop previous to a partial evaluation was carried out to contribute to the acquisition of contents, skills and abilities to identify risks and to determine suitable protective barriers. Objectives: To determine if the pedagogical intervention of the workshop on biosecurity principles affected the academic performance and to analyze the students' value judgments on biosecurity. Material and methods: The qualifications obtained in the question of biosecurity in 2009, 2010 and 2011 tests for a total of 166 students were gathered. A survey designed by the Area was provided to analyze the students' valuation of the 2011 workshop. An informed consent was given to 2011 students and 35 surveys were obtained. Pearson's Chi Square Test was applied with a 95% confidence level ($\alpha=0.05$). C contingency coefficient was used to determine differences in academic performance along different years. Results: Students' test approval: 14/69 in 2009, 31/49 in 2010 and 38/48 in 2011. A significant difference was obtained ($p>0.05$). Students valuations: students totally agreed with these items: the report assignment allowed them to acquire contents of basic biosecurity rules (7/10); the importance of the application of the rules in the laboratory (9/10); risks recognition of the students' assignment after the workshop (7/10); the application of biosecurity rules in laboratory work protected them and their pairs (8/10). Conclusion: The pedagogical strategy has a positive impact on the acquisition of contents, skills and abilities of basic biosecurity principles learning, observed in students' academic performance and valuations in the studied period.

26. SURGERY ASSOCIATED INFECTIONS IN A PRIVATE MEDICAL CENTER OF ROSARIO

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In 2001 the WHO said that world wide strategies to stop the antimicrobials resistance had to be used. Ten years later the need to formulate and update directions periodically to decrease hospital infections and adapt the treatment and antibiotic prophylaxis is emphasized. For this reason, it is important that each hospital has updated information of prevailing pathogenic agents. Goals: epidemiological check of the bacteriological agents found in the cultures of patients hospitalized in a private institution in Rosario. Material and Methods: Between January 2009 and January 2011, 154 cultures were processed bacteriologically from the following samples: subcutaneous fluid, abscess, abdominal collections, biliary, peritoneal and ascitic fluid, surgical wounds, different tissues and cysts. Results: 56(36%) cultures were negative. In the 98 (64%) positive cultures the bacteriological agents identifications made gave the following results: *Staphylococcus aureus*: 33,67% (33); *Escherichia coli*: 18,36% (18); Anaerobios (*Peptostreptococcus sp*; *Bacteroides sp*; *Clostridium sp*) 13,26 (13); *Pseudomonas aeruginosa*: 11,22% (11); *Staphylococcus coagulasa negativo*: 6,12% (6); *Klebsiella sp*: 5,1% (5); *Streptococcus viridans* 2,04% (2); *Acinetobacter baumani*: 2,04% (2); *Enterococcus faecalis*: 2,04% (2); *Morganella morganii*: 2,04% (2); *Citrobacter freundii*: 1,02% (1); *Enterobacter cloacae*: 1,02% (1); *Aeromonas trota*: 1,02% (1); *Providencia rettgeri*: 1,02% (1) Discussion : It is clear that the *Staphylococcus aureus* is the most frequently isolated agent. It is important to mention the prevalence of this bacteria in our research. This differs from the bibliography which says that Gram-negative bacilli are the most found (*Escherichia coli*, *Klebsiella* and *Pseudomonas*). Conclusions: It is important to know which infectious agents prevail within each health institution, specially for prevention and the right antibiotic treatment.

27. COMPARATIVE STUDY OF ERYTHROCYTE AGGREGABILITY (EA) IN RATS OF IIMb Y IIMFm/ β LINES

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Introduction: In previous work we showed hiperaggregability in line IIMFm / β (β) rat (model of Obesity and Diabetes). Line IIMb (b), originally eumetabolic, developed phenotypic instability with anomalous glucose intolerance and obesity tendency. **Objective:** compare the EA in lines b and β in animals of 100, 200 and 300 days. **Methodology:** We used male rats, line b and β of 100, 200 and 300 days (n = 10). The EA was determined by optical method in suspensions of red blood cells (RBs) Dextran 500 2% in saline (Htc: 40%) We determined: T (estimating the size of the aggregates) and V (estimating the initial rate). EA results were analyzed with t Student test and expressed as mean \pm SD. **Results:** (T) 100 days: β : 1.77 \pm 0.08, b: 1.76 \pm 0.08ns; 200 days: β : 1.76 \pm 0.09, b: 1.73 \pm 0.06 ns; 300 days: β : 1.72 \pm 0.06 b: 1.73 \pm 0.08 ns. V: 100 days: β : 0.49 \pm 0.18, b: 0.39 \pm 0.10***; 200 days: β : 0.53 \pm 0.14, b: 0.42 \pm 0.13***, 300 days: β : 0.48 \pm 0.18, b: 0.45 \pm 0.13 ns. T showed no significant differences between lines during animal's lives. V showed significant differences in 100 and 200 days animals but not at 300 ones. We can conclude that the line b developed changes in mechanical erythrocyte membranes tending to reproduce the EA behavior that exhibits the model β , probably related with progress in line b rat metabolic condition during their growth.

28. DIET AND GALLBLADDER DISEASE (GD) RISK. PRELIMINARY RESULTS

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The influence of diet on the risk of developing GD is not completely elucidated. To carry out this nested case-control study, 40 cases were contacted and 42 controls were randomly selected from subjects who participated in a previous prevalence study. Controls underwent a new abdominal ultrasound to rule out asymptomatic gallstones. We conducted a survey of socioeconomic conditions. In addition, a food frequency questionnaire (FFQ) was used to register diet consumed five or more years before GD diagnosis for cases, and usual diet for controls. A Photographic Atlas was used for quantifying food portions size. Total kilocalories mean daily consumption was significantly higher in cases than in controls (2729 \pm 133 vs. 2362 \pm 108; p=0,037). Adjusted Odds Ratios were statistically significant for protein (OR=0,96;95% CI=0,93-0,99;p=0,013), fat (OR=1,05;CI=1,00-1,11;p=0,039), dairy foods (OR=0,99;CI=0,991-0,998;p=0,005), vegetables (OR=0,994; CI=0,991-0,998;p=0,006), fruit (OR=0,67; CI=0,47-0,94; p=0,022), nuts (OR=0,99;CI=0,993-0,999;p=0,032) and fiber consumption (OR=0,91,CI=0,82-0,99;p=0,035). According to these preliminary results, a high dietary fat consumption increase GD risk. High intakes of protein, dairy products and fiber, as well as high intake of some sources of fiber (vegetables, fruits and nuts) would work as protective factor in preventing GD.

29. APPLICATION OF AGE-PERIOD-COHORT MODELS TO COMPARE COLON CANCER MORTALITY RATES BY SEX IN ARGENTINA

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For the last decades the age-period-cohort (APC) analysis has been focalized in the APC model fitting in order to estimate the effects associated to these three factors separately. Such model use information from a table containing rates classified by age groups and time periods. Frequently, the purpose of a study is to make pairwise comparisons between sets of rates, assuming a specific APC model for each one. In this way it is possible to contrast mortality due to a certain cause between two sub-populations such as men and women. Traditionally, descriptive plots are the most widely used method for comparative analysis. However they have some drawbacks because it is necessary to represent in the same plane three related time scales: age, period and cohort.

Given two sets of rates defined over the same age groups, time periods and cohorts, it is possible to construct linear hypothesis tests in order to determine whether the corresponding rates are absolutely proportional or proportional within any of the three factors: age, period or cohort. Primarily, an APC model was fitted to male and female colon cancer mortality rates, in order to perform a comparative analysis. A possible proportionality within age groups between both sub-populations was observed in graphical comparisons. Then, this hypothesis was evaluated using a Wald Chi-Square test. It provided a value of 163.09 ($p < 0.0001$), indicating that the rates of colon cancer in women and men are not consistent with this type of proportionality. Even when proportionality does not hold, the APC model can help to organize and clarify comparative analysis.

30. DIAGNOSIS OF VERTICALLY TRANSMITTED INFECTIONS IN PREGNANT WOMEN IN THE FIRST LEVEL OF ATTENTION

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According to Latin American Center of Perinatology (CLAP) it is considered as accurately studied women who have one control of Chagas disease and Hepatitis B (HBV) before 20 weeks gestation and one of Syphilis, HIV and Toxoplasmosis before and after this limit. Objectives: to evaluate CLAP compliance care serology, and relationship between educational level, residence place and controls number with serology. Methods: 334 women perinatal cards from Rosario and nearby area that delivered their babies in Provincial Hospital from 12/1/2010 to 2/28/2011 were included. Education: incomplete primary, complete primary and incomplete secondary, complete secondary and others. Irwin Fischer and Chi-square Tests were applied. Results: 5% cards accomplished with CLAP regarding reactions number and realization time. In 60% at least one of each serological reactions were found, but they were not consistent with the required number of reactions. In 32% they lacked at least one of the determinations. The 7 women who did not have reactions done were women with 1, 6 ± 0, 5 controls as an average. HBV test was the least checked, missing in 28% cards followed by HIV in 11%. Toxoplasmosis was not studied in 5%, Chagas disease in 4% and Syphilis in 3%. A significant association between education level and serological controls ($p = 0,002$) was found. There were no statistical differences between living place and serological controls. Conclusions: Serology was incomplete in a high number of cards according to CLAP; women without serology also had an inadequate controls number. Education higher level was linked with more appropriate serological studies in number and date of completion.

31. NEW AND OLD TOXICITY IN PATIENTS WITH HIV NUCLEOSIDE

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Objetivo: To evaluate the toxicity of 3TC/AZT vs TNF/ETB. Material and Methods: quantitative study, observational, prospective patients carried HIV (+) under HAART from 06-08 to 06-11. They followed two groups of patients were treated with the AZT/3TC group 1 and group 2 Tenofovir/Emtricitabine more NNRTI or IP. How to recruit responds to a non-probability sampling rate and sample within it discriminated against, with inclusion and exclusion criteria and signing of the IC. Categorical variables were analyzed using Chi-square and Wilcoxon rank. Significance level $\alpha = 0.05$. Results: The sample of 102 patients. The sample was limited to 72 patients who had second laboratory information in addition to the baseline. Group I 32. HB g/dl 14.02±0.31 basal/14.14±0.29 second, Hto% 54.58±13.21 basal/52.55±10.05 second, HDL mg/dl 37.38±4.94 basal/ 35.13±1.68 second, basal creatinine mg/dl 0.81±0.03 and second 0.84±0.03. No significant differences were detected between laboratories. Group II 40. Hb g/dl 13.30±1.6495 basal /13.52±1.527 second, Hto% 40.05±4.80 basal/40.20±5.211 second, HDL mg/dl 34.83±60.628 basal/38.05±46.44 second, basal creatinine mg/dl 0.73±25.667 and second 0.84±0.03. No significant differences were detected in Hb, Hto and creatinine, and if there are differences in HDL. Viral load decrease is statistically significant by Wilcoxon ($p < 0.0001$) for both groups. There was no lipotrophy. Conclusion: Both treatment regimens were shown to be safe and effective from the immune toxicity could be found only in group II with regard to alteration of lipids precisely HDL.

32. PARAMETERS OF OXIDATIVE STRESS IN LIVER AND KIDNEY, AND HISTOLOGICAL STUDIES IN RATS TREATED WITH MONOSODIC GLUTAMATE (MSG)

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Our objective was to determine the oxidative state in liver and kidney in rats of both sexes treated with MSG and to examine possible histological alterations. Worked with four groups (n = 6): M-MSG and H-MSG (males and females respectively with neonatal subcutaneous administration: 4 mg/g weight), M-C and H-C (male and female controls treated with saline solution). In liver and kidney we made histology: and oxidative stress: lipoperoxidation (LPO), glutathione (GSH) level and activity of the enzymes Glutathione peroxidase (GPx), glutathione reductase (GR), superoxide dismutase (SOD) and catalase (CAT). Nor changed GSH in liver. In kidney, M-MSG reduction of GSH ($\mu\text{mol/g tej}$): M-C 2,8±0,15, M-SMG 2,2±0,19*, with increase in GPx (nmol/min mg prot): M-C 48,2±2,5, M-MSG 54,3±1,5* and GR (nmol/min mg prot): M-C 19, 8±1,9, M-MSG 27,6±1,7*, without changes in SOD and CAT, indicating an increase in the turnover of GSH. H-MSG has not modified any parameter. In liver: M-MSG there was an increase in GPx and CAT with reduction of SOD. H-MSG showed an increase in GR with reduction of SOD. Histology showed degenerative changes at the proximal convoluted tubules level, cytoplasm vacuolized. Cylinders were observed in distal and cortical tubules light. This could be linked to increased urinary excretion of proteins in the treated rats. We observed in liver only steatosis characterized by fat cytoplasmic vacuoles. MSG affected parameters of the oxidative state with a sexual dimorphism in the response, which would also differ according to the affected organ.

33. EVALUATION OF DIFFERENT ENERGY SOURCES ON THE REARING OF HOLSTEIN FRIESIAN CALVES

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The objective was to evaluate the performance of Holstein Friesian calves fed different energy sources. The experiment was carried out using 20 male calves of 60 days of age. Animals were allocated to two different groups according to their live weight. Following an adaptation period, calves were fed a diet containing 20 % of alfalfa hay and soybean meal+ground sorghum grain (TS) or soybean meal+Hominy feed (TG). Diets were isoenergetic and isoproteic. The following variables were measured twice a month: live weight, height, thoracic perimeter and dry matter intake. Means were compared using the “t-student” test. Results are presented in table 1: Mean±SD for treatments

	TS	TG
ILW, kg	73,1± 11	73,3±12
FLW, kg	138,5±25	136,3±19
LWG 0-28, kg	0,75±0,1	0,72±0,1
LWG 28-54,kg	0,97±0,1	1,06±0,1
LWG 54-68,kg	1,00±0,2	0,81±0,2
LWG 0-68, kg	0,93±0,2	0,90±0,2
DMI 0-68, kg	4,45±0,3	4,13±0,5
FCE 0-68,kgDM/kgLW	4,97±0,3	5,06±0,5
Pthorax 0-68, cm	18,1±4.9	16,1±4.1
Height, cm	14,7±2.4	13,5±2.3

ILW: initial live weight, FLW: final live weight, LWG: live weight gain, FCE: feed conversion efficiency, Pthorax: thoracic perimeter. DMI: dry matter intake. There were no significant differences between treatments. It is concluded that animals performed well in both treatments using different energy sources.

34. COMPARISON OF TWO MODELS OF SYNDROME METABOLIC INDUCED WITH MONOSODIUM GLUTAMATE (MSG) IN FEMALE RATS

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Our objective was to determine whether there are differences with the metabolic alterations observed during chronic administration of oral MSG, and whether such changes may affect renal function. We worked with 4 groups of female rats (n=7): MSGn: postnatal subcutaneous administration of MSG (4 mg / g), Cn: control treated with saline, GMSo: treated orally 3 g/100 g food and 1% drinking water daily for 5 months from the 2nd month of life, Co: NaCl. Weight was monitored weekly, naso-anal length and tail length (growth parameter), obtaining the Lee index (IL). Metabolic changes (weight gain, IL, retroperitoneal fat, tolerance test glucose and insulin and lipid profile) and renal function were studied at 7 months of age. It was observed in both models, weight gain (g) induced significant GMS from weaning in neonatal and in the period of 5 months in the oral: Cn: 220 ± 5, GMSn: 241± 2 *, Co: 181± 12, GMSo:218 ± 10 *, as well as an increase of Lee index of obesity indicator. Only in the neonatal group was found a reduction of tail length (cm) Cn: 18.9± 0.3,GMSn: 14.6 ± 1.5 *, Co: 25.2 ± 0.9,GMSo: 24,3 ± 0.8. In both groups increased retroperitoneal fat that was accompanied by a reduction in HDL cholesterol GMSn and an increase in total cholesterol GMSo. These changes were accompanied by impaired glucose tolerance (K% / min): Cn: 1,5 ± 0,05, GMSn: 0.81± 0.03 *, Co: 1.66 ± 0.1 and GMSo: 1,15 ± 0.05 * and insulin resistance. No changes in kidney function, but there was a decreased ability to concentrate urine in GMSn. Both models produce metabolic syndrome abnormalities coinciding with the exception of growth retardation.

35. HEMORHEOLOGICAL ALTERATIONS INDUCED BY AEROBIC TRAINING IN RATS

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Different studies show that persons who practice physical activity regularly present a minor mortality than sedentary persons and that exercise is especially effective to improve health conditions in situations such as ischemic cardiopathy, hypertension, obesity, diabetes mellitus, etc. The aim of this work was to evaluate the possible hemorheologic alterations induced for aerobic training in rats. That work is part of a project financed by UCES. A group of 12 male medium age (9-10 months) Wistar rats, provided by the biotery of UCES, corresponding to constituted by 6 sedentary (CS) and 6 with aerobic training (EA) was studied. Blood samples were extracted by cardiac puncture, anticoagulated (EDTA) analyzed in 24s. Plasmatic and whole blood viscosity was measured in a Viscosimeter (Brookfield DV-II), viscoelastic parameters were obtained using an Erythrodeformeter, and aggregation kinetics with an Erythroaggregameter. Results showed that plasma and whole blood viscosity do not differ significantly (p>0.2, p>0.3) to low shear rates in the samples EA with regard to the CS. However, plasma viscosity was slightly increased to high shear rate (p<0.1) in the samples EA, indicating a probable increase of the plasmatic protein concentration. Significant differences are not observed in aggregation kinetics between the EA and the CS (p>0.1) but if a significant increase is observed in the RBC surface viscosity and elastic module (p<0.02, p<0.001) due to the aerobic training. Results indicate that the aerobic training in rats would induce an increase in the RBC elasticity and surface viscosity without modification in RBC deformability.

36. PAGET’S DISEASE IN A PRE-HISPANIC TIBIA IS CONFIRMED BY HISTOLOGICAL SECTIONS AND RADIOCARBON DATE

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In 1947 in the “Cerro Grande” of Los Marinos Island, Río Paraná, Argentina, was recovered a pre-Hispanic pagetic tibia. Radiocarbon date was obtained of this human bone. The age obtained is very young: 590 +/- 60 BP. This shows that the bone belongs to an individual who lived prior to the European contact. Radiograph, CT Scan, Macroscopical and Histological examinations were performed. Differential diagnosis were considered, and present-day etiological theories (viral, genetics) are being discussed. New fields of paleopathological analysis could allow the study of biological relationships between different populations who lived along the Southern Paraná River.

37. EVALUATION OF HEMATOLOGIC VARIABLES IN CALVES COMPARING DIFFERENT GENOTYPES

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The objective was to compare hematologic variables on three different genotypes. Thirty male (castrated) calves were used: 10 American Holstein (AH), 10 Holstein Friesian (HF) and 10 crossbreds (F1) Holstein x Jersey (HxJ). Each group was allocated to a different lot. Animals were fed a TMR, consisting of 68% of ground corn grain, 20% of soybean meal, 10% of alfalfa hay and 2% of a mineral supplement. Calves were reared from 80 kg up to 330 kg. Blood samples were extracted at: 80, 180 and 320 kg of live weight and analyzed for: hematocrit, white blood cell count, and count of each type of white blood cell. Data were analyzed using the Mixed procedure comparing treatments, sampling periods and their interaction. Results are shown in Table 1: Mean±SD of hematologic variables in different genotypes.

	AH	HF	HxJ	p value		
				Tt.	Per	Tt*Per
Hto	31.2 ± 5.0	33.8 ± 4.7	32.5 ± 3.3	0.028	0.0001	0.082
Leu	11050±4284	10775±4317	10973±3676	NS	NS	NS
N	3364±2440	2572±1516	2476±1288	NS	0.021	NS
B	22 ± 44	34 ± 76	44 ± 57	NS	0.029	NS
E	124 ± 208	94 ± 135	54 ± 83	NS	0.068	NS
M	724 ± 590	696 ± 490	1088 ± 818	0.010	0.0001	0.02
L	6731±2514	7200±3635	7242±2572	NS	NS	NS

Hto.: Hematocrit %, Leu.: Leukocytes total/mm³, N: Neutrófilos/mm³, B: Basófilos/mm³, E: Eosinófilos/mm³, M: Monocytes/mm³, L: Linfocitos/mm³.

It is concluded that there are differences between genotypes for hematocrits and monocytes cell count.

38. FATTENING CALVES FROM MILKING HERDS COMPARING THREE GENOTYPES

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The objective was to compare live weight gain, dry matter intake, feed conversion efficiency and carcass yield of three different genotypes when fed a TMR. The trial was conducted using 45 male (castrated) calves: American Holstein (AH, n=15), Holstein Friesian (HF, n=15) and crossbreds (F1) Holstein x Jersey (HxJ, n=15). At the age of 75 days, animals were allocated to three different lots and fed a TMR consisting of 68% of ground corn grain, 20% of soybean meal, 10% of alfalfa hay and 2% of a mineral supplement. Feed intake and live weight were measured every two weeks. Animals were slaughtered at 330 kg and carcass weight was registered. Data were subjected to analysis of variance (ANOVA) and the Tukey's test was used to test differences between means. Results are shown in Table 1: Mean±SD for the three genotypes

	AH	HF	HxJ
ILW (kg)	80.7(±9.3)a	60.5(±7.3)b	62.6(±8.5)b
FLW(kg)	336.1(±34.7)a	331.5(±21.3)a	333.3(±23.0)a
LWG(kg/day)	1.35(±0.51)a	1.12(±0.52)b	1.15(±0.54)b
DMI(kg/day)	5.70(±2.19)a	5.18(±2.26)b	5.02(±2.10)b
FCE(kgDM/kg LW)	4.20(±1.79)a	4.45(±1.97)a	4.30(±1.81)a
Days to slaughter	186	229	229
Carcass yield (%)	55.3(±2.2)ab	56.3(±1.8)a	54.1(±1.9)b

Means within rows that do not have a common superscript letter differ at P<0.05. ILW: Initial live weight; FLW: Final live weight; LWG: live weight gain; DMI: dry matter intake; FCE: feed conversion efficiency.

39. MEMORY WORK IMPAIRMENT IN NEONATE WISTAR RATS EXPOSED TO VANADIUM THROUGH LACTATION

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We have demonstrated that vanadium exposure through lactation alters the physical and neural development of the offspring and its CNS myelination. Heat shock protein (Hsp 70) activation and astrogliosis in hippocampus (Hc) and cerebellum (Cer) were also observed in those pups. The aim of this work was to perform a memory work study in neonate Wistar rats exposed to sodium metavanadate (NaVO₃) through lactation. 8 Wistar rat litters were randomly distributed in Control and Treated groups. Treated dams received 3mg of NaVO₃/kg bw/day i.p from the 10th to 21st postnatal day. Control dams received the same volume of saline. The behavioral test used was adapted from Ennaceur and Delacour (1988) one. An open box made of wood 65x45x45 cm was used. The objects to be discriminated were pyramids or cubes of different colours made of glass. Each session was comprised of 2 trials of 3 min with a 30 min intertrial interval. In the first trial the rat was placed in the box were 2 identical objects were placed in each of two corners. In the second trial the rat was placed in the box with a familiar object in one corner and a new one in the other. In both sessions the time spent by the rat exploring each object were recorded. In NaVO₃-exposed rats, the preference % for the novel (NOP) (55.2±3.0) wasn't different of 50% while it was significantly higher (63.7±2.6, p< 0.01) in control ones. The deficit in visual recognition observed in the present work could be related to the altered myelination detected in those animals.

40. HISTOLOGICAL AND CELL PROLIFERATION STUDIES ON PARTIAL HEPATECTOMISED RAT LIVER

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We correlated histological changes with cell proliferation index (CPI) after partial hepatectomy (PH) performed in rats. Wistar male adult rats were anesthetized (i.p., Ketamine-Xilazine) and PH was performed. Rats were sacrificed at: 24 h, 48 h and 72 h and at 7 and 15 days, after surgery (n=3). Non hepatectomised rats were used as controls (C). After the sacrifice, liver tissues were fixed in 10% formaldehyde and histological processed for paraffin embedded. Some samples were stained with H&E and others were used to determinate CPI immunohistochemically by PCNA technique, at the different times mentioned above. H&E: abundant micro vacuoles were observed in hepatocytes at 24 h and 48 h, diminishing at 72 h. At 7 and 15 days no vacuoles were seen. Organized hepatocyte cords and sinusoidal morphology were found in all groups. At 72 h isolated eosinophilic groups of hepatocytes (IEGH) were seen, without extracellular matrix or sinusoids inside. CPI obtained with PCNA: C=1.7±0.3; HP24=26.9±1.1; HP48=37.9±5.5; HP72=32.0±1.4; HP7D=11.0±4.2 y HP15D=5.6±0.2. The greatest amount of cells in phase S of cell cycle was obtained at 24 h (absolute value=2.8±0.8; %=10.3±2.9) and the greatest value in phase M was obtained at 48 h (absolute value=5.6±1.2; %=14.8±1.1). We still observed significant higher values of CPI compared with C at day 15. We suggest that a great number of cells are maintained in the cell cycle up to 15 days after PH. The peak of mitosis at 48 h could explain IEGH found at 72 h that may represent *de novo* generated hepatocytes.

41.

IN VITRO MODELING OF ARTERIAL HYPERTENSION BY MEANS OF TREATMENT WITH TRYPSIN

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Rheological studies of erythrocyte membrane (RBC) are useful to analyze and model microcirculatory alterations observed in vascular pathologies as human arterial hypertension (HTA). Previous studies have demonstrated that the surface electric charge (CAE) decrease in RBCs affecting intercellular interactions. An *in vitro* modeling was carried out by mean of comparative analysis between the action of trypsin and the alteration in hypertensive patients to understand their mechanism of action. The *in vitro* modeling consisted of quantifying the effect of different concentrations of trypsin on RBC from healthy donors. The CAE was measured applying the distribution in a two-phase watery system technique and by means of spectrophotometric analysis using the polycationic Alcian Blue dye. The time needed to attain 50% of final aggregation ($t_{50\%}$) was obtained using an Erythrogameter, aggregation shape parameter (ASP) by means of image analysis. Results show a decrease in CAE from RBC treated with trypsin ($p < 0.05$) and from HTA ($p < 0.005$) with respect to the controls. The CAE and $t_{50\%}$ values from RBC treated with 2 mg/mL were similar to RBCs from hypertensive patients. The trypsin treated RBCs presented altered ASP as did RBCs from HTA. Then, *in vitro* modeling was satisfactory to reproduce the alterations in RBC from HTA, being established that the 2mg/mL trypsin treatment produces similar alterations to those found in this pathology.

42.

REFERENCE RANGE FOR URINARY EXCRETION OF ALBUMIN AND β -N-ACETYL GLUCOSAMINIDASE IN THE FIRST TRIMESTER OF PREGNANCY

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Preeclampsia is a multiple system disease of unknown cause and one of the leading causes of maternal and perinatal morbimortality. Since the pathophysiology includes endothelial damage, several biochemical markers, which are indicative of it, such as microalbuminuria (MAU) and N-acetyl β glucosaminidase (β NAG), have been proposed. In order to determine the predictive value of these parameters in preeclampsia, the first objective was to establish the reference range of MAU and β NAG in the first trimester of pregnancy. This study included 19 pregnant women without associated diseases, gestational age: 10-15 weeks, prior clinical evaluation by a tocogynecologist and signature of informed consent. MAU (immunoturbidimetric method), β NAG (colorimetric method) and creatinine levels (CrU, enzymatic method) were determined in the first morning urine. The results were (media \pm SD): MAU: 10 \pm 6 mg/l; β NAG: 4.3 \pm 3.5 U/l; CrU: 1.2 \pm 0.6 g / l; MAU / CRu: 10, 5 \pm 7.5 mg / g Cru; β NAG / CRU: 4.1 \pm 3.2 U/g CrU. The reference ranges obtained (media \pm 2SD) for MAU/CRU and β NAG/CRu were: 0 - 25.5 mg/g CrU and 0 - 10.5 mg/g CrU, respectively. This allows studying the possible predictive value of the MAU and β NAG as early markers of preeclampsia.

43.

ANALYSIS ON THE VARIABILITY OF THE URINE ALBUMIN/CREATININE RELATION ACCORDING TO TIME AND TEMPERATURE OF PRESERVATION

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The objective was to determine the stability of the relation between microalbuminuria (MAU) and urine creatinine (CrU) in samples preserved at different temperatures ($^{\circ}$ C) for a longer period of time than recommended by the commercial technique. The urine samples (first of the morning) were collected from 23 pregnant women, in their first or second trimester, with no associated pathologies. MAU (mg/l; immunoturbidimetric method) and CrU (g/l; kinetic method) were determined in the fresh samples and in 2 aliquots preserved at -20° C and -70° C for 90 days. No significant differences were found in the MAU levels: time zero, 90 days at -20° C and -70° C (media \pm SD: 17,1 \pm 13,0; 16,5 \pm 11,9; 17,6 \pm 11,1 respectively; $p=0,6017$). However, there were significant differences in the CrU concentrations in the differently preserved samples ($p < 0,0001$). After 90 days, CrU value was significantly higher than in time zero, both at -20° C and -70° C (media \pm SD: 0,77 \pm 0,40; 0,93 \pm 0,50; 0,94 \pm 0,50 respectively), there being no significant difference between these last two ($p > 0,05$). To conclude, the CrU analysis must be performed on a fresh sample, whereas the MAU can be determined in urines preserved even up to 90 days at -20° C or at -70° C, which is considerably useful for research as it allows to optimize the human and economic resources available.

44.

COMPARATIVE STUDY OF THE METHOD OF 4-AMINOFENAZONA WITH TRADITIONAL METHODS APPLIED TO FORENSIC PRACTICES

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The preliminary step in the forensic practices for to detect blood are the orientative tests, their positividad affects to other studies. The orientative tests are based in the activity peroxidasa of the hemo group that in witnesses of H₂O₂ and organic reagents it produces coloration or luminiscencia. The Adler Method (M1) uses as oxygen acceptor to the bencidina in acetic acid and their positividad is manifested with blue-greenish color. The Luminol Test (M2) uses 3-amino-ftalhidrazina in carbonate of sodium means and their positividad is a blue luminiscencia that it observes in the darkness. In the 4-aminofenazona method and fenol (M3) react with the hemo group giving rosy color. Our objective was to evaluate in supposed stains of blood, the results of the reaction M3 with the conventional methods (M1, M2). 20 suspect samples were processed: 10 hyssops with substances of the place of the fact, 5 knives and 5 dark clothes. An aliquot was extracted of the hyssops and the methods M1 and M3 were applied. Aliquots with hyssops absorbed in physiologic were taken of the knives and clothes. M2 was also applied to the clothes. Results: 8 positive hyssops applying M1 and M3; 1 hyssop was weakly positive for M1 and negative for M3 ($p=0.008$). In the samples of knives 3 were positive and 2 negative identical with M1 and M3. The clothes were 3 results positive with the 3 methods (M1, M2, M3); 1 clothe was negative in the 3 methods and 1 clothe was weakly positive with M1 and negative with M2 and M3. The χ^2 test showed significant statistically association between the conventional methods (M1, M2) and it proposed (M3) ($p < 0.001$). The results show that the method 4-animofenazona-fenol can substitute to the traditional methods applied to forensic practises.

45. SPERM APOPTOSIS AND LOCATION OF IMMUNE RESPONSE IN HUMAN SEMEN

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Apoptosis play an important role in human reproduction. Anti-sperm antibodies (ASAs) have been related to infertility problems. The aim of this work was to investigate the relationship between sperm apoptosis with the occurrence of ASAs and its location in human semen. A retrospective study was made of 230 men that consulted for infertility in the Human Reproduction Service. Detection of ASAs in seminal plasma and/or in sperm membrane was performed with indirect and direct TAC II technique respectively. 43 semen samples with positive ASAs were selected (agglutinated sperm > 25%). The apoptosis was evaluated by the Tunel method. The apoptotic index (IA= N° apoptotic sperm x 100/ N° total cells) was calculated. Samples were classified in three groups: G1) ASAs only fixed on sperm membrane; G2) ASAs free in seminal plasma; G3) ASAs fixed and free simultaneously. The Spearman coefficient rS was calculated. There was no correlation between the IA and ASAs in G1 (rS = 0,218; P = 0,183, n=39), G2 (rS = 0,095; P = 0,747, n=14), neither G3 (rS = 0,146; P = 0,682, n=10). Apoptosis appears not to be related with the presence of ASAs located in seminal plasma neither on the sperm membrane. A combination of assays that studies the sperms alterations would be offer better and more complete information for the study of male infertility.

46. PREVENTION AND CONTROL OF BRUCELLOSIS AND LEPTOSPIROSIS: ZONOTIC DISEASES AS A CURRENT ISSUE IN THE COMMUNITY

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The distribution of diseases in a population is not limited to one level but it recreates a multidimensional and dynamic image of the health-disease process. The aims of this work were to develop community strategies for the prevention of zoonoses, to determine the serum-reactivity rate to *Leptospira interrogans* and *Brucella canis* in canine sera, and to characterize the associated risk factors. An immune-epidemiological study was carried out in Casilda city, Santa Fe. Ninety-seven houses and 131 dogs were randomly chosen (1:3/ house). The seropositive rate to *Leptospira interrogans* was determined through MAT serological test with 8 serovars. Sera with an agglutination of 50% or more live leptospira were considered positive in relation to a control, to a 1:100 dilution employed as cutting point. The diagnosis for canine Brucellosis was performed with the fast sero-agglutination technique in plaque with 2ME. A survey was carried out with the owners. Health education workshops were organized together with an interdisciplinary team. Results were 26% (34/131), for MAT, while 6%(3/45) of dogs were seroreactive to SAR 2-ME. The survey sample analysis showed: a) waste accumulation sites: 39%(38/97), b) no garbage collection service area: 18%(17/97), and no current water service 54%(20/97), c) surveyed people who did not show awareness of zoonosis nor prevention measures: 94%(91/97). Communication between the health team and with the community was organized and awareness and community organization for zoonosis prevention were promoted.

47. CONTROL AND PREVENTION URBAN ZONOSSES ACTIVITIES IN STRAY DOGS BELONGED TO SAN JOSE DE LA ESQUINA DOG SHELTER

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Zoonotic diseases are those transmitted from animals to humans. By the term of "urban zoonoses" are designated diseases naturally transmitted from animals, but also considered urban zoonoses those have been established between the people who inhabit the periphery of the city. The aim of this study was to establish the diagnosis of zoonotic disease situation and intestinal parasites in stray dogs, confined to shelters. In 2010 we studied 139 stray dogs housed in the shelter of San José de la Esquina, took out the diagnosis of brucellosis, dirofilariasis (serum) and intestinal parasites (faeces). For brucellosis were used microagglutination RSAT-agglutination or antigen-BPA-buffered, using indirect ELISA to confirm the results and dirofilariasis was determined by ELISA. Stool examination for parasites were processed by direct and enrichment techniques. Were surveyed in total 139 animals, we determined the existence of 45 (32.4%) brucellosis sera including 5 (3.6%) had clinical signs related. The filarial ELISA was negative in all animals. Analyzed fecal samples were found 46% of positive samples (at least one parasite). The most frequent parasites were *Ancylostoma caninum* (26%) and *Giardia* spp. (15%). This reflects that some stray dog disease could come from the street. Diffusion is important to the people of the risks associated with zoonoses and their preventive measures.

48. USE OF PROSTAGLANDIN F2 α IN POSTPARTUM DAIRY COWS AND ITS EFFECT ON SUBCLINICAL ENDOMETRITIS

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Subclinical endometritis (SE) decreases reproductive efficiency in dairy cows. The prevention and treatment of this pathology is important for improving the reproductive and productive performance of dairy cattle. Several authors described the use of prostaglandin F2 α or its analogues, based on the effective results on clinical endometritis, for SE. The aim of this work was to evaluate whether the use of PGF2 α in postpartum reduces the presence of SE. Were examined 271 Holstein dairy cows between 21-56 days in milk (DIM), of 6 farms in the southern Santa Fe. Two of them using 2 applications postpartum of PGF2 α , at 30 DIM and other at 45 DIM (with PGF2 α). Farms remaining do not use of PGF2 α (without PGF2 α). To calculate the % of neutrophils were used endometrial samples obtained by cytobrush. Cows in "healthy" (without SE) and "SE+" (with SE), were grouped.

	With PGF2 α	Without PGF2 α	Total
Healthy	164 (90.11%)	76 (85.39%)	240 (88.56%)
SE+	18 (9.89%) a	13 (14.61%) a	31 (11.44%)
Total	182	89	271

Was applied Fisher test. There were no significant differences (P<0.3095) on the presence of SE between groups. We concluded that use of PGF2 α in postpartum, not decreases SE, in relation to farms that do not use. It required further studies of therapeutic measures for the control and treatment of SE.

49. SEARCH FOR EVIDENCE OF LEPTOSPIRA INFECTION VICTORIA, ENTRE RÍOS

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Leptospirosis is an infectious disease caused by *Leptospira interrogans*. It affects animals and the infection is transmitted to humans through the urine. In April 2011, cases of leptospirosis were confirmed in adult patients, engaged in rural and fishing in the coastal zone and islands. The objective was to: look for evidence of leptospiral infection serological and bacteriological techniques in areas of Victoria, facing the islands. We obtained 28 serum samples of dogs from Victoria's municipal shelter located near the coast, which were analyzed using the Microscopic Agglutination Test (MAT), which is the reference test. Paired samples were analyzed in positive dogs. In the vicinity of the shelter were hunted red 2 rats (*Holochilus brasiliensis*), and the same blood serum was analyzed by MAT and seeded organs obtained at necropsy. In the coastal area, facing the islands, water samples were collected in ditches and stagnant pools, where pH measurements were found close to 8 with Merck strips. All samples were grown in special media for leptospira. The seeded samples were observed in dark field microscope and observed spirochetes compatible with *L. interrogans* only in water samples from ditches with pH 8. We also analyzed the soil pH and observed alkalinity. Planted samples, so far no development of leptospire was obtained, although not ruled them until after 6 months. Of the total samples analyzed dog sera, five (17.8%) tested positive for MAT. The prevalent serovar Castellonis and the title was highest was 1:200. Sera from mice were negative. The results showed that ecological conditions in Victoria, are favorable for the survival of leptospire. The evidence found in this study are consistent with those found in dogs in the area south of Santa Fe.

50. NUEVA ROMA "C" NEIGHBORHOOD'S ANIMAL HEALTH COMMUNITY CLINIC. PROFILE OF THE DOGS AND CATS CONSULTS IN ONE YEAR

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Nueva Roma "C" is a neighborhood of minimal economic resources habitants, located in Casilda, Argentina. The aim of this study was to elaborate the profile of the animals that came to the clinic. The period of study was from August 25, 2009 to August 25, 2010. In this period 413 animals were brought to consult; 368 of these animals were dogs. The arithmetic mean of the dogs and cats age was 19 and 20 months respectively. There were few animals spayed at the moment of first consult (2.7% of dogs and 6.8% cats). However 12% of the dogs and 61% of the cats solicited this surgical intervention. It was observed a deficiency in the population control. The number of owners that wanted to spay their dog was very low in comparison with cats. These results shows that the owners wanted to spay their pets, and that they weren't able to do it previous the community clinic was open. It is concluded that is necessary to reinforce the education to assure the compromise of the owners with their animals' welfare and environment. It is also necessary to try to determinate why there are so few older dogs and cats in the neighborhood.

51. SURVEY OF PET FOOD FOR ADULT DOMESTIC FELINES IN MAINTENANCE. METABOLIC ENERGY EVALUATION

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In Argentina, the elaboration and commercialization of pet food for adult cats is regulated according to the dispositions of AAFCO (American Association of Feed Control Officers). They recommend the cat food to have a minimum value of metabolic energy (ME) of 4000 kcal/Kg of dry matter. The aim of this work was to make a survey of the whole dry cat food available for adult cats in maintenance and to determinate if they are meeting the ME specifications. Fourteen products were studied. The ME was calculated from the information gathered of crude protein, lipids and carbohydrates. The ME was determinate with the NRC 1985 formula: $EM (Kcal/100g) = (3,5 * CP) + (8,5 * lipids) + (3,5 * carbohydrates)$ and for the products that specifies in their packages, the ME was calculated with the NRC 1974 formula. The ME of each product obtained was compared with the AAFCO specifications. The average value of ME was 3600 Kcal/Kg of dry matter. Only three products (21%) were above the value recommended. The high percentage (79%) of product with a caloric concentration below the minimum value demonstrates that in some cases it may occur an energetic deficiency. It is concluded that most of the products are not meeting the specifications and that the ration has to be regulated.

52. SURVEY OF PET FOOD FOR ADULT DOMESTIC FELINES IN MAINTENANCE. CHEMICAL COMPOSITION

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The American Association of Feed Control Officers recommends that the products elaborated for the feeding of adult cats must have values of crude protein (CP) and lipids above 24 and 10% respectively and carbohydrates to be below 40%. The aim of this study was to do a survey of the whole dry pet food for adult cats available in Argentina and to establish if they are meeting the nutritional specifications. Fourteen commercial dry foods were studied. The chemistry composition available in the package was considered and compared with the specifications established by AAFCO. The arithmetical mean and range was calculated. The mean values and range (in % of dry matter) were: CP, 29.3 (24-36); lipids, 11.5 (8-21); carbohydrates, 35.8 (27-42,5). One and six products provided inadequate quantities of CP and lipids respectively. Three commercial foods provided excessive amount of carbohydrates. It is concluded that not all the pet food available for adult cats meets the nutritional recommendations; therefore it would be convenient to carefully evaluate the composition of the product before offering it to a cat.

53. NUEVA ROMA "C" NEIGHBORHOOD'S ANIMAL HEALTH COMMUNITY CLINIC. DOGS AND CATS DISEASES FREQUENCY DISTRIBUTION IN ONE YEAR

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Nueva Roma "C" is a neighborhood of limited economic resources habitants, located in Casilda, Argentina. The aim of this study was to establish the diseases frequency distribution observed in dogs and cats in the first year of this clinic. The period of study was from August 25 2009 to August 25 2010. There were 566 consults taken; 42% of the animals had skin problems, 22% had gastrointestinal affections. The rest of the medical problems were respiratory, traumatic, nervous diseases, etc. The great amount of patients with skin diseases was mostly because of ectoparasite diseases. It was difficult to have a rigorous control of the animals in most of the cases, because only the 37% of, the owners returned to the clinic. It is concluded that the animals of this neighborhood have mostly diseases related to poor sanitation practices, lack of anti-parasite treatment and deficient nutrition.

54. EFFECT OF HEPARIN ON THE SPERM-ZONA PELUCIDA INTERACTION IN PIGS

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Heparin is a sulfated glycosaminoglycan (sGAG) extensively used *in vitro* to study the role of GAGs secreted by the female reproductive tract. In cattle, heparin is thought to promote capacitation by removing proteins (spermadhesins) that are adsorbed to the sperm (e) and would inhibit capacitation. In pigs, some of them are also removed; others remain attached interacting with the zona pelucida (ZP) and belong to the heparin binding proteins group (HBP). To determine the role of heparin on e/ZP interaction, porcine ovaries were processed to obtain ZP, aliquots of it were applied to coverslips. The samples were inverted on slides containing capacitated sperm suspension from 5 adult boars, with and without heparin, incubated in humidified chamber during 15, 30 y 60 min. Coverslips without ZP were used as control. Attached sperm were counted under light microscope. Statistics: ANOVA. The number of attached sperm was significantly lower with heparin compared to those incubated in its absence at 30 min of incubation ($p < 0,0001$). In untreated coverslips, the number of sperm attached was minimal and did not vary significantly throughout the incubation process in all cases. This result could be explained by considering that the zona pellucida contains certain polysulfated domains that possibly compete with heparin and could be involved in the pre-selection of sperm contributing to the prevention of the polispermia.

55. VARIATION FOR SINGLE SEQUENCE REPEATS (SSR) POLYMORPHISM IN A TOMATO SECOND CYCLE HYBRID (SCH) AND ITS PARENTS IN RESPECT TO THE UNIFORM GENOTYPES FROM WHICH THEY WERE DERIVED

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Forty nine SSR were applied to a tomato SCH and its parents, two Recombinant Inbred Lines (RIL18 and RIL1) derived from a cross among the cultivated *Solanum lycopersicum* cv. Caimanta (C) and the wild *S. pimpinellifolium* LA722 (P). These original genotypes together with the interspecific F₁(CxP) had been previously characterized with the same SSR. In the first group of genotypes (RIL18, RIL1 and SCH), 41 SSR amplified. Just 26 of them were polymorphic, 22 being codominant (as expected for SSR) and 4 being dominant. In addition, 10 of the polymorphic SSR amplified two bands (6 in RIL18 and 4 in RIL1), which is not expected in homozygous genotypes. Five of the 15 monomorphic SSR had also been monomorphic in the second group of genotypes: C, P and F₁(CxP). The remaining 10 monomorphic SSR had been polymorphic in the second group and represent genomic regions fixed by artificial selection during the selfing generations that allowed obtaining the RILs. Also, 6 of the SSR showing two bands in the first group had been monomorphic in the second group, these results suggesting that chromosomal rearrangements occurred during the selfing generations. Though a less levels of polymorphism was found in the first group in respect to the second one, an adequate number of SSR was selected to characterize the segregant generations of the SCH in order to validate QTLs detected in early generations derived from F₁(CxP).

56. ADVENTITIOUS SPECIES OF THE FAMILY BRASSICACEAE IN THE SANTA FE PROVINCE

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The Family Brassicaceae (=Cruciferae) is classified in the Order Brassicales (*sensu* APG-III). It consists of annual or perennials herbs, with basal leaves in rosette and alternate stem leaves. Flowers tetramerous, inflorescence in raceme. Fruit siliqua or silicula, dehiscent or indehiscent. The objective of this work is to begin the taxonomic study of the Brassicaceae Family and the distribution of adventitious taxa for the Santa Fe province. We worked with specimens preserved in the herbaria UNR (Rosario), SF (Esperanza) and SI (Darwinion, San Isidro); together with bibliographical revision, field observations and lab work to confirm the specimen's identity. There are only 16 genera present in Santa Fe. Of these, only four are native: *Lepidium* L., *Descurainia* Webb & Berthel., *Exhthalmolobos* Al-Shehbaz & C.D. Bailey and *Rorippa* Scop. The rest are adventitious and usually weedy. Genera with only one species: *Capsella* Medik. (*C. bursa-pastoris* (L.) Medik.), *Diploaxis* DC. (*D. tenuifolia* (L.) DC.), *Eruca* Mill. (*E. vesicaria* (L.) Cav.), *Hirschfeldia* Moench (*H. incana* (L.) Lagr.-Foss.), *Iberis* L. (*I. amara* L.), *Neslia* Desv. (*N. paniculata* (L.) Desv.), *Rapistrum* Crantz (*R. rugosum* (L.) All.), *Nasturtium* R. Br. (*N. officinale* R. Br.). Genera with two species: *Brassica* L. (*B. nigra* (L.) W.D.J. Koch and *B. rapa* L.), *Camelina* Crantz (*C. alyssum* (Mill.) Thell. and *C. sativa* (L.) Crantz), *Raphanus* L. (*R. raphanistrum* L. and *R. sativus* L.) and *Sisymbrium* L. (*S. irio* L. and *S. officinale* (L.) Scop.). Taxonomic information, illustrations, a distribution map and keys for the identification of taxa based on morphological characters are provided.

57. INTRODUCTION TO THE STUDY OF THE FAMILY SAPOTACEAE IN THE PROVINCE OF SANTA FE (ARGENTINA)

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The Family *Sapotaceae* is classified in the order *Ericales* (*sensu* APG III); it consists of 53 genera and 1100 species of pantropical distribution. This family is represented in Argentina by 3 genera and 7 species. They consist of trees or shrubs, with white latex; simple leaves, entire, alternate or sub opposite, with or without stipules, pinnate venation, and an indument of malpighian hairs in T form. Perfect or imperfect flowers, solitary or in axillary fascicles. Calyx with 4 or 5 persistent sepals, free or partially united in the base. Corolla with united petals; 4-5-(6) lobes. Stamens 4-5-(6) opposite to the petals. Superior ovary, 2-5-(6)-locules. Fruit usually a berry. The objective of this work is to begin the taxonomic study and the geographical and ecological distribution of the *Sapotaceae* Family for the Santa Fe province. We worked with specimens preserved in the herbaria UNR (Rosario), SF (Esperanza) and SI (Darwinion, San Isidro); together with bibliographical revision. Our preliminary results show that the family is represented in Santa Fe by 3 genera, 4 species: *Chrysophyllum gonocarpum* (Mart. & Eichler) Engl.; *C. marginatum* (Hook. & Arn.) Radlk.; *Pouteria gardneriana* (A. DC.) Radlk. y *Sideroxylon obtusifolium* (Roem. & Schult.) T.D. Penn. Taxonomic information, illustrations and a distribution map are provided.

58. INCIDENCE OF FUNGIC PATHOGENS IN GRAINS OF MAIZE FROM A FERTILIZATION NITRO-SULPHURED CROP

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The object of the present work was to determine the incidence of fungic pathogens in grains of two cultivars of maize, with different levels of nitro-sulphured fertilization. The cultivars used were ACA 2000 and ACA 417 RR2. The treatments consisted of an arrangement factorial of three levels of N (N_0 : control; N_{75} : 75 kg N.ha⁻¹ and N_{150} : 150 kg N.ha⁻¹) and two levels of S (S_0 : 0 kg S ha⁻¹ and S_{40} : 40 kg S. ha⁻¹). To determine the fungic incidence the grains incubated on 2% APG with light alternation (ISTA, 2009). A total of 200 seeds / hybrids were analyzed. The percentage of each pathogen were determined. The statistical was in BCAD with 3 repetitions / hybrids. The information was analyzed by ANOVA and Tukey-Kramer. The pathogens present were *Aspergillus flavus*, *Diplodia* spp., *Fusarium* spp. and *Penicillium* spp. *Aspergillus flavus* did not present differences between levels of N, level of S, and interacción NxS, in each cultivar. In ACA 2000 *Diplodia* were significant differences (p <0,05) between N_0 (14%) and N_{75} (5%), and between N_0 (14%) and N_{150} (2%) (p <0,01). The differences (p <0,05) between levels of S (S_0 9% and S_{40} 4%). For *Fusarium* and *Penicillium* were differences between levels of S (p <0,10) (S_0 24% - S_{40} 36% and S_0 23% - S_{40} 52% respectively). In ACA 417 RR2 for *Diplodia* were differences (p <0,05) between N_0 (11%) and N_{75} (3%), and between N_0 (11%) and N_{150} (1%) (p <0,01). For *Fusarium* there were differences on N (p <0,05) (N_0 18% and both doses of N 32%). For *Penicillium* were differences for S_0 18% and S_{40} 6%. The *Diplodia* incidence was reduced with N fertilization in both hybrids. The choice of the fertilizer is an important factor on the managing of the culture.

59. SOYBEAN: LATE SOWING DATES AND THE RISK OF EARLY FROST IN ZAVALLA, ARGENTINA

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Field trials were carried out in Zavalla, Argentina (33° 01' S) in two cropping seasons (CS). Sowing dates (SD) were 18 January 2007 (CS1) and 14 January 2008 (CS2), respectively. In CS1 the cultivars (cvs) were: DM 3100; DM 3700 (MG III); DM 4200; DM 4600; DM 4870 (MG IV); RA 514; A5766RG (MG V); A6411RG; N Andrea 66 (MG VI); A7321RG; A 7336RG (MG VII). The same cv were used in CS2, except the cv A7336RG, which was substituted by TJs 2170 (MG VII). Phenology, daily minimum temperature, rainfall, were recorded throughout the CS. Experimental design was a randomized complete block with 3 replications for each CS and SD. Seed yield, adjusted to 13.5% humidity, plant height, main stem node number, were determined. Data were analyzed by analysis of variance and LSD test was used to compare means. In both CS, the occurrence of the first frost were at the end of may and during june, so the early cv (MG III and IV) could complete their cycles before frost happened, except DM4870 in CS1. Late cvs (MG V, VI and VII), were affected by frost and they couldn't complete the filling period. The cycles of the cv which completed their growth had varied between 94 -101 days (d) and 80 - 93 d in CS1 and CS2, respectively. Number nodes and yield had low values because of the late SD. DM 3100 and DM 3700 in CS1 and DM 4200, DM 3100, DM 4870 and DM 3700 in CS2 showed the higher yield, without any difference among them (P<0,10). The means values were 1070 kg/ha and 1671 kg/ha in CS1 and CS2, respectively. In order to avoid damage frost, is very important to know the behaviour of cvs when sowing date was extremely delayed.

60. EVALUATION OF CORN HYBRIDS (ZEA MAYS L.) UNDER DIFFERENT DENSITIES

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The objective of this trial was to evaluate yield and its components in corn hybrids having different characteristics and sown at different densities. The trial was conducted in Zavalla (60° 53' W, 33° 01' S.), Argentina in 2011. The hybrids sown were: I 880 MG (H1) , KM 3601 MG RR2 CL (H2), SRM 565 MG (H3), Ax 852 MG (H4), NK 880 TDMax (H5), NK 910 TDMax (H6), M 510 Hx (H7), and 2879 TDMax (H8). The experimental design was a randomized complete block with 4 replications for each hybrid. The treatments consisted in two densities: low (D1) 6 pl m⁻² and high (D2) 11 pl m⁻². The variables used were: number of spikes (NS), yield (Y), weight of 1000 grains (WG), grains per plant (GPP), grains per square meter (GPM2) and yield per plant (YP). The data were analyzed by ANOVA and Duncan's test. A statistically significant difference (p<0.05) between the densities was found in NS for H8 (D1:1.2 spikes.pl and D2:0.9 spikes.pl), H6 (D1:1.0spikes.pl and D2:0.8 spikes.pl), and (p<0.10) for H5 (D1:1.0 spikes.pl and D2: 0.87 spikes.pl). A significant difference (p<0.05) in WG was found between the two densities for H4 (D1:367g and D2:331g) and (p<0.10) for H8 (D1:363g and D2: 377g). The GPP and YP values showed a significant difference (p<0.05) between the densities for all hybrids, being 50-60% lower under D2 than under D1. The GPM2 variable showed no significant differences between densities for none of the hybrids. H4 varied significantly in its Y (D1:9922 kg ha⁻¹ and D2:8213 kg ha⁻¹), WG (D1:317 g and D2:330 g) and YP (D1: 153 g.pl and D2: 80.2 g.pl) when density increased, therefore showing less plasticity in density choice. The other hybrids showed compensatory mechanisms between yield components allowing better tolerance to changes in the densities studied. The crop density adjustment should be made taking into account the behavior of the different varieties available in the market.

61. PROTEOLYTIC ACTIVITY IN HEPATOPANCREAS OF THE EURYHALINE CRAB *Neohelice granulata*: RESPONSE TO HYPERREGULATION AND FEEDING

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Hyperegulation is a physiological status with enhanced energy demands. Biochemical adjustments in extrabranchial organs (i.e. modulation of digestive enzymes in hepatopancreas) could occur in response to hyperegulation and could be affected by feeding status. However, integrative studies on these adjustments are lacking. The aim of this work was to determine proteolytic activity (Prot) at different times after feeding in hepatopancreas of *N. granulata* from Mar Chiquita coastal lagoon acclimated to different salinities (S) (35‰S:osmoconformation; 10‰S:hyperegulation). Adult male crabs exposed for 10 days to 35 and 10‰S (feeding regime: 3 times a week), were unfed for 5 days (preingesta=PI) and then individually fed. Prot was determined in PI, immediately (t0) and at 1, 8, 24, 48, 72, 120 h post-ingesta. Prot (enzyme units \times h \times 1 \times mg protein⁻¹) was assayed by measuring azocasein (1% ρ /v) hydrolysis in Tris-HCl, 100 mM, pH 7.5. PI and t0 values were similar both in 35 and 10‰S. In 35‰S, Prot was higher between 8 (8.23 \pm 0.2) and 72 h (8.9 \pm 1) post-ingesta than in PI (5.04 \pm 0.95). In 10‰S, no differences occur between PI (4.2 \pm 0.87) and any time post-ingesta. No differences were found between osmoconformation and hyperegulation conditions in either PI or at any time post-ingesta (p=0.001). The results shows a different Prot responses to feeding in 35 and 10‰S suggesting a relationship between post-feeding adjustments and osmoregulatory status.

62. OSMOREGULATION AND Na+K+ATPase ACTIVITY IN GILLS OF *Neohelice granulata* FROM BAHÍA BLANCA

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Although flexibility in osmoregulation is a key strategy for the occupancy and using of habitat in euryhaline crabs, studies on the role of branchial Na+K+ATPase activity (NKA) in hyporegulation and comparative intraspecific studies on osmoregulation pattern in individuals from different geographic areas are lacking. The aim of this work was to determine osmolality of the hemolymph (HL) and NKA in anterior 1-5 (AG) and posterior 6-8 (PG) gills in adult males of *N. granulata* from Bahía Blanca estuary exposed to 10, 35 and 45‰ salinity (S) to establish the occurrence of osmoregulatory flexibility, possible adjustments in NKA and comparative aspects with crabs from different areas. Osmolality (mOsm \times kg⁻¹) in HL and the medium (M) was determined with cryoscopic osmometer. 2500 \times g supernatants (homogenization medium: 0.25M sucrose/0.5mM EGTA-Tris pH 7.4) were used as enzyme extracts. NKA (nmol Pixmin-1 \times mg prot⁻¹) was assayed by measuring 5mM ATP hydrolysis in (mM): 20 imidazole /100 NaCl/30 KCl/0.5 EGTA, pH 7.4; control: without KCl, 1 ouabain. t-test was used as statistical analysis (p<0.05, n=5). Crabs hyperegulate in 10S (M:316 \pm 5;HL:659 \pm 42), osmoconform in 35S (M:908 \pm 21; HL:825 \pm 33) and hyporegulate in 45S (M:1192 \pm 12;HL:870 \pm 24). Upon osmoconformation, NKA was AG: 84 \pm 18 and PG: 220 \pm 61. NKA in AG was not affected by S. In PG was higher in 10 (468 \pm 79) than in 35S. In 45S, NKA in PG was similar to that in 35S. Similarly to crabs from Mar Chiquita lagoon, the results show flexibility in osmoregulation pattern and differential modulation of NKA in PG suggesting the occurrence of differential adjustments associated to hyperegulation.

63. ATPases ACTIVITIES IN GILLS OF CRAB *Neohelice granulata* FROM CONTRASTING HABITATS OF MAR CHIQUITA COASTAL LAGOON: DIFFERENTIAL RESPONSE TO HYPOREGULATION

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Differential adjustments in key biochemical parameters (i.e. enzymes involved in osmoregulatory responses in euryhaline crabs) could be related to successful occupancy of contrasting habitats within a same area. The aim of this work was to determine ATPase activities in anterior (1-5) (AG) and posterior (6-8) (PG) gills in male crabs from mudflat (M) and saltmarsh (SM) exposed for 10 days to 35 (osmoconformation) and 45‰ salinity (S) (hyporegulation). 2500 \times g supernatants were used as enzyme extract (homogenization medium: 0.25M sucrose/0.5mM EGTA-Tris pH7.4). ATPase activity (nmol Pixmin-1 \times mg prot⁻¹) was assayed by measuring 5mM ATP hydrolysis in (mM): 20 imidazole pH7.4/130 NaCl/0.5 EGTA/ 1 ouabain/1 NaN₃ without (Basal ATPase activity: BA) or with bafilomicin A1 (1 μ M). The difference of both assays represents V-H+ATPase activity (VHA). t-test was used as statistical analysis (p<0.05, n=5). In 35S BA in AG of crabs from M was 1096 \pm 202, being similar in SM. In 45S, BA in AG was similar to that in 35S in crabs from both areas. In crabs from SM, BA in PG was similar in 35 (430 \pm 41) and 45S (678 \pm 101). In crabs from M, BA in PG was higher in 45S (1091 \pm 253) than in 35S (439 \pm 58). About 30% of BA in PG of crabs upon hyporegulation corresponds to VHA. The results suggest a differential role of VHA in PG upon hyporegulatory conditions and the occurrence of differential adjustments in PG associated to hyporegulation in relation to habitat.

64. NMDA RECEPTOR IN THE MARGINAL SPHINCTER CONTRACTION OF THE SEA ANEMONE *Phymactis clematis*

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The aim of this work is to determine the existence of a locus PCP present in ionotropic NMDA receptor (iNMDAR) on the marginal sphincter of *P. clematis*. Once dissected, the sphincter was placed in an isolated organ chamber connected to a polygraph. Sea water in controlled conditions was used. Three successive assays were made and expressed as dose-response curves (DRC): 1) at increasing concentrations of KCl (20-200mM); 2) KCl+Glutamate (Glu) (10-5M) and 3) Glu(10-5M)+ketamine (21mM). ANOVA test were used. P values< 0.05 were considered statistically different. The results show that the maximal response obtained was: 1) 1.46g \pm 0.17 (n=27) with 180mM KCl; 2) 1.74g \pm 0.14 (n=18) with 140mM; 3) 0.83g \pm 0.35 (n=9) with 180mM. Previously different concentrations of Glu alone were tested, without response in any of them. But the contraction increased when the membrane was depolarized with KCl, suggesting a higher calcium influx. The channel ligated to the NMDA receptor has a locus PCP phencyclidine and other non-competitive antagonists binding site. To determine the existence of this locus, DRC were made with the addition of ketamine, which has more affinity for this site than other antagonists, resulting in a significantly lower maximal response than the other experiments. The presence of ketamine in the medium would be annulling the effect of Glu as well as the one caused by calcium that flows through the iNMDAR and other Ca²⁺ channels. These results reinforce previous work in which was postulated that the iNMDAR is involved in the regulation of the calcium influx that causes marginal sphincter contraction of sea anemone *P. clematis*.

65. DIGESTIVE FLEXIBILITY AT THE BIOCHEMICAL LEVEL IN THE EURYHALINE CRAB *Neohelice granulata* FROM MAR CHIQUITA COASTAL LAGOON: EFFECT OF AN ABRUPT CHANGE OF SALINITY ON LIPASE AND PROTEOLYTIC ACTIVITY IN THE HEPATOPANCREAS

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Studies about possible digestive adjustments at the biochemical level associated to osmoregulatory flexibility of euryhaline crabs are lacking. The aim of this work was to study the effect of an abrupt salinity change on lipase (Lip) and proteolytic (Prot) activities in hepatopancreas. Male crabs maintained for 10 days in 37‰ salinity (hyporegulation conditions) were transferred (To) to 10‰S (hyperregulation conditions). Lip and Prot were determined at To and at 2, 4 and 24 h after transfer (Controls: Crabs maintained at 37 and 10‰S throughout the experimental period). 10000xg 15 min supernatant was used as enzyme extract (homogenization medium: Tris-HCl 0.1M, pH 7.4) (4 ml x g tissue⁻¹). Lip (μmoles pNP x min⁻¹ x mg protein⁻¹) was assayed by measuring pNPPalmitate hydrolysis (0.7 mM) in 50 mM Tris/HCl 50mM pH 8.5 at 37°C. Prot (enzyme units×h⁻¹×mg protein⁻¹) was assayed by measuring azocasein hydrolysis (1% p/v) in Tris-HCl, 0.1M, pH 7.5. At 2 and 4 h, Lip (2 h= 40.6 ± 12.8; 4h= 40.1 ± 10.9) and Prot (2h= 4.8 ± 0.9; 4h= 4.1 ± 0.6) were similar to To (Lip= 38.6 ± 15.1; Prot= 4.5 ± 0.4)(n= 5; Anova, p<0,05). After 24 h, Lip (17.7 ± 5.1) and Prot (2.5 ± 0.5) were lower than activities at To (n= 5; Anova, p<0,05). In controls, Lip and Prot remained constant. The results shows a modulation of lip and prot activities in hepatopancreas upon an abrupt salinity change suggesting that differential adjustments in digestive capacity could be occurring in relation to changes in osmoregulatory status.

66. GLUCOSE HOMEOSTASIS IN THE EURYHALINE CRAB *Cyrtograpsus angulatus* FROM MAR CHIQUITA COASTAL LAGOON: REGULATION BY DOPAMINE

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Glucose (Glu) constitutes the main monosaccharide present in the haemolymph of crustaceans. Glu homeostasis would be tightly controlled by endogenous and exogenous factors (i.e. salinity). In crustaceans, biogenic amines (i.e. dopamine) (Dopa) are involved in the regulation of several key physiological processes. However, in euryhaline crabs, the knowledge about the regulatory role of biogenic amines in glu homeostasis upon different ambient salinities is still scarce. The aim of this work was to study the effect of Dopa on glu homeostasis in adult male crabs of *C. angulatus* acclimated for 10 days in 35‰ and 10‰ salinity (S) (osmoconformation and hyperregulation conditions, respectively). Crabs were injected with 100μl of saline (400mM NaCl, 10mM KCl) without (control) (35‰S: n=6; 10‰S:n=9) or with 10-4M Dopa (35‰ S: n=12; 10‰S: n=14). After 30 min, a sample of hemolymph was taken and hemolymphatic glu (mg/ml) was determined with Wiener Lab AA glycemia kit, by reading the developed red quinoneimine complex at 505 nm. In both 35 and 10‰ S, individuals treated with dopa exhibited a higher glu concentration (about 38 and 34%, respectively) in hemolymph, (35‰S:control=198±17, Dopa=318±, t=-2,19 p<0.047; 10‰S: control=191±36, Dopa=291±32, T=118, p=0.025). The results suggest that dopamine up-regulates hemolymphatic glucose in *C. angulatus* acclimated to different osmorregulatory conditions.

67. RESPONSES TO SALINITY IN *Cyrtograpsus angulatus*: OSMOREGULATION AND ENERGY STORES

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Studies on osmoregulatory flexibility and associated metabolic adjustments in various ecological relevant euryhaline crabs are lacking. The aim was to determine osmolality (O) and glucose levels (Glu) in hemolymph (HL) and glycogen content (G) in anterior (AG) and posterior gills (PG), chela muscle (M) and hepatopancreas (H) in male individuals (n=6-8) of *C. angulatus* from Mar Chiquita coastal lagoon exposed for 10 days to 40, 35, 10 and 6 ‰ salinity (S). O (mOsm x kg⁻¹) and Glu (mg glu x ml HL⁻¹) were determined by cryoscopic osmometer and with Kit (Wiener Lab AA), respectively. G (mg x g tissue⁻¹) was assayed in homogenates from AG, PG, M(sucrose 0.25M/EGTA-Tris 0.5mM, pH7.4) and H (Tris-HCl 0.1M, pH 7.4) by hydrolysis with α-amylglucosidase (0,2ml/ml). ANOVA was used for statistical analysis (p<0.05). O in HL was different to Me at 40, 10 and 6‰S (40‰S=981±14, Me=1096±23); (35‰S=894±38; Me=964±4) (10‰S=644±27, Me=273±1), (6‰S=609±17; Me=146±7). Glu in HL was similar in all salinities (6‰S=108±53; 10‰S=55±9; 35‰S=77±19; 40‰S=70±20). G in BA was lower in 40‰S (11±2), but similar in 6 and 10‰S (17±5.5 and 29±6, respectively) to 35‰S (38±0.8). G in BP, M and H was not affected by salinity: BP(6‰S=14,8±4; 10‰S=14,1±3; 35‰S=15±4; 40‰S=13,4±3), M (6‰S=29,9±9; 10‰S=36±9; 35‰S=39±5; 40‰S=58,3±24); H (6‰S=1,1±0,3; 10‰S=1,1±0,6; 35‰S=2,2±0,9; 40‰S=1,5±0,1) (p>0.05). The results show that *C. angulatus* behaves as hypo-hyperregulator and suggests the occurrence of differential metabolic adjustment in relation to osmoregulatory status.

68. MICROBIOLOGICAL QUALITY OF LIQUOR WITH AND WITHOUT MILK

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The aim of this work was to determine the microbiological quality of craft liquors made with different natural materials with and without milk in their composition. Was analyzed 10 samples of liquor without milk: A y B industrial, artisanal (8) from: C to I and J home manufacture. The artisanal with milk were 6, from K to P. Spread on surface was carried out (0.1ml) by triplicate, with H&L for moulds (incubated 10 days at 25°C) and for mesophilic aerobic bacteria (BAM) with PCA and incubated to 37°C for 48 h., the colonies were characterized macro and microscopically. Results: BAM: A y B: < 10, C: 35x10; D: <25x10; E: 13x103; F:<25x10; G: <10; H: <10; I: <10; J: <10. K 10,4 x 102, L <10; M 80 x 102, N 3 x 102, O 19 x 102 y P 5 x 102. Industrial samples A y B and artisanals E, F, G, I, J do not show molds. In C and D was identified *Penicillium* spp., In H and O *Moniliella* spp. In L, *Cladosporium* spp. The industrial liquor don't have microorganisms. In the craft liquor BAM is present, but the counts are low with the exception of E. In the liquor with milk all samples except L, showed growth of BAM. The highest growth was observed in the Irish Cream. The liquor are not inherently dangerous products for health, so in our country, are not legislated microbiological limits for these. Taking into account these results, is important that the manufactures of these products comply with GMP thus preventing microbiological contamination and growth in the product.

69. AUTOLOGOUS IMMUNOGLOBULIN BINDING TO ERYTHROCYTE ASSOCIATED WITH AGING

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The binding of autoantibodies to senescent cell-specific autoantigens (SCA) would be the main mechanism for removal of senescent red blood cells (SeRBC). The aim was to estimate the concentration of membrane-bound autologous immunoglobulin (IgG) in young red blood cells (YRBC) and SeRBC. We studied peripheral blood samples of volunteer donors with negative serology (n=8). SeRBC and YRBC were separated by preformed Percoll gradients. Aliquots were hemolyzed in 5 mM phosphate buffer (5P8) and centrifuged at 20000 g for 20 min at 4°C. The pellets were washed with 5P8, resuspended in 10 mM Hepes buffer, 130 mM NaCl pH 7.4 (buffer A) and incubated at 37°C with fluorescein isothiocyanate labeled goat anti-human IgG antibodies. The suspensions were centrifuged at 30000 g for 20 min at 4°C and washed twice with buffer A. The pellets were solubilized with buffer containing 1% 20P8 poly (oxyethylene-8-lauryl ether). The determination of IgG was performed on a Varian Fluorescence Spectrophotometer Cary Eclipse, using 495 nm λemisión and 519 nm λexcitación. Membrane proteins were assessed spectrophotometrically and expressed in mg of protein fluorescence. The average fluorescence/mg protein was significantly higher (p<0.01) in SeRBC (71.85±9.25) compared to YRBC (59.95 ± 5.90). The results show an increase of membrane-bound IgG and SCA associated with aging.

70. MOLECULAR BASIS OF D NEGATIVE PHENOTYPE IN THE POPULATION OF ROSARIO

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The D- phenotype is caused by the complete deletion of the RHD gene in Caucasians. However, specific RHD regions are present among D- individuals from different ethnic groups. The Amerindian and African genetic component varies in the distinct social strata of our population. The aim of this work was to study the molecular basis of the D- phenotype in individuals from Rosario. Blood samples from D- patients from the Hospital Provincial del Centenario (HPC) (n=653) and a private laboratory (LP) (n=661) were studied. The Rh phenotype was determined using monoclonal antibodies. The presence of the RHD gen was investigated using a multiplex PCR strategy. Samples RhD-/RHD+ were studied by PCR-SSP, microarray and sequencing. The percentage of RhD-/RHD+ samples was significantly higher in HPC (p<0,05) (HPC: 2,45%; LP: 1,21%). The RHDψ allele was exclusively found in 1,17% of the HPC samples with cccc phenotype. In samples with C or/and E antigens the following alleles were found: r's (HPC: 10, 91%; LP: 1, 69%), RHD-CE(3-9)-D (HPC: 3,64%; LP: 3,39%) and RHD(M295I) (LP: 1,82%). New alleles were also identified: RHCE(1-2)-RHD(3361del11-10) (LP: 1,69%), RHD(581insG) (LP: 1,69%), RHD(46T>C) (HPC: 1,82%; LP: 3,39%). The variants found in HPC patients reveal the contribution of the African etnia to their genetic pool. The alleles found in LP samples are characteristic from Europeans. The molecular study of RH will help to develop reliable strategies for blood selection in Blood Banks and prenatal RHD genotyping.

71. PARTICIPATION OF HLA ALLELES IN THE THERAPEUTIC RESPONSE TO INTERFERON ALFA AND RIBAVIRIN IN HCV INFECTION

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At present, it is possible to cure about half of patients with Hepatitis C (HC) treated with the best therapeutic option available: Pegylated Interferon and Ribavirin (PEG-INF/RBV). Therefore, almost half of patients will be nonresponders, showing advanced fibrosis and insulin resistance. The mechanisms that regulate resistance to interferon therapy in HC are not well known. Viral factors have been implicated, metabolic and genetic factors. The aim of this study was to determine the involvement of HLA-DRB1 alleles in the response to combination therapy with PEG-INF/RBV of HC. Patients were divided into responders (n = 8) and no responders (n = 10). We performed molecular typing of HLA-DRB1 alleles by PCR-SSP. To determine the association of HLA-DRB1 alleles with the response to treatment with PEG-INF/RBV, we analyzed the frequency of these alleles in patients with HC. The results indicate that although the DRB1*13 allele has a > frequency alleles in responders (25%) than non-responders (5%), the difference is not statistically significant (p>0.10). These preliminary findings do not allow an association of HLA-DRB1 alleles with response to combination therapy in the patients studied. We believe it is necessary to analyze a greater number of individuals in order to determine the presence of association of these genetic markers with response to treatment in patients with HC PEG-INF/RBV.

72. PHAGOCYTOSIS PARAMETERS ASSESSMENT IN SENESCENT RED BLOOD CELLS

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Senescent red blood cells (SeRBC) expose cryptic antigens that are recognized by cells of the mononuclear phagocyte system. It is proposed that autologous IgG accumulation in the erythrocyte membrane could activate the binding of C3b, promoting phagocytosis of SeRBC. The aim of this study was to determine the presence of autologous IgG and C3b in SeRBC and Young RBC (Y) by flow cytometry using whole blood. We analyzed 20 samples of peripheral blood from volunteer donors. 100 µL of RBC were washed with phosphate buffered saline solution (PBS) and 0.2% suspensions prepared in PBS. Aliquots were incubated with: I) anti-human IgG Alexa 488 and II) anti-C3b plus anti-mouse APC. Cytometry was performed on the cytometer FACS Aria II. Data were analyzed with the program FACSDiVa. Given that FSC represents the cell size and SSC internal cellular complexity, consider that the population SeRBC are located in <FSC and> SSC, while YRBC are located in the region of > FSC and <SSC. The averages of the percentages of cells positive for IgG were SeRBC: 1.50 ± 0.7 and YRBC: 0.16 ± 0.4. The median percentage of C3b positive cells were SeRBC, 2.2 ± 1.4 and YRBC: 0.2 ± 0.55. The percentages of cells positive for both markers tested were significantly higher in SeRBC (p <0.0001). These findings indicate an increase of IgG and C3b on the membrane SeRBC. These results demonstrate that RBC of different ages can be analyzed by this methodology, without performing a physical separation of cells.

73.

STUDY OF ABH ANTIGEN EXPRESSION AND CD44 MARKER IN ORAL LESIONS

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The aim of this study was to investigate the relationship of changes in ABH antigens, the expression of adhesion molecule CD44 and the degree of malignancy in patients with oral lesions. We worked with samples included in paraffin plugs from patients (n=50) to investigate ABH antigens and saliva samples from the same patients to analyze CD44 expression. ABH antigens were investigated by the modified specific immunoadherence technique. Saliva samples were incubated with anti-CD44 antibody and then with a secondary antibody labelled with allophycocyanin. Results were semiquantified from strongly positive to negative adherence. A weak ABH antigen expression in atypical areas and a total deletion in the areas affected with neoplasia was observed in 17 of the 50 samples, belonging to patients with malignant oral lesions. Conservation of antigen expression was observed in 2 patients with oral cancer, while ABH antigen deletion was observed in 12 of the 50 samples analyzed belonging to patients with benign lesions. A conserved expression of ABH antigens was observed in 19 samples from patients with benign oral lesions. Results obtained by confocal microscopy showed varied fluorescence intensity corresponding to the presence of CD44 protein in samples from patients with diagnosis of cancer and pre cancer, coinciding with that observed in the deletion in paraffin plugs. Our studies indicate that loss of ABH antigen expression may be associated with malignant oral lesions. Furthermore, overexpression of CD44 molecule observed in these patients could be considered as a risk marker in these individuals.

74.

VASCULAR FLORA OF THE PROVINCE OF SANTA FE: FAMILY TILIACEAEDi Sapio O¹, Galetti L², Oakley L², Prado D²¹Farmacobotánica, Fac. Cs Bioquímicas y Farmacéuticas, UNR, Rosario. ²Botánica, Fac. Cs Agrarias, UNR, Zavalla.

The Family Tiliaceae, currently included in the Order Malvales, (now part of the subfamily Tilioideae, according to the system APG-III), is cosmopolitan and comprises about 47 genera and approximately 700 species that inhabit tropical and subtropical regions in both hemispheres. In Argentina, the family is represented by four genera, of which only two appear in the Province of Santa Fe: *Corchorus* L. and *Luehea* Willd. This contribution is an introduction to the knowledge of this family and its geographical distribution in the departments of the province of Santa Fe. The methods consist of a bibliographical review, consultation of national herbaria with important collections of the province (SF: Esperanza; SI: San Isidro; UNR: Zavalla), field work experience of the authors and lab work to confirm their identity. The preliminary results show that the genus *Corchorus* L. is represented in Santa Fe by only one species: *Corchorus argutus* Kunth, a native shrub distributed in the northern departments and lives frequently in low fields, floodplains and gallery forests. The genus *Luehea* Willd. is represented by the sun-loving tree *Luehea divaricata* Mart. ('azotacaballo', 'Francisco Alvarez'), cited in national catalogs for the province but without a voucher specimen. However, we observed and photographed its presence in Paraje Hardy, Dept. Gral. Obligado. The bark and leaves of this species are used in vernacular medicine for its tonic and antiarrheal properties. Taxonomic information, distribution map and illustrations are provided.

75.

STUDY OF THE ACTION OF L-ASCORBIC ACID ON THE GENOTOXICITY OF THE SULFATHIAZOLE-NITRITE MIX IN ACIDIC MEDIUM WITH THE ALLIUM TESTPontoriero A¹, Sánchez Y^{1,3}, Giulidori C¹, Hure E¹, Mosconi N¹, Rizzotto M^{1,2*}¹Área Química General, Facultad de Ciencias Bioquímicas y Farmacéuticas, UNR; ²CIC-UNR, Rosario. ³Facultad de Ciencias Exactas, Químicas y Naturales, Universidad de Morón.

DNA can be damaged by several exogenous and endogenous substances. Between the last are notably the N-nitroso compounds, which are formed by the reaction of nitrogenated substances -like sulfonamides- and nitrites in the stomach. L-ascorbic acid can react with nitrite diminishing or removing the nitrosation risk, fact that was previously proved by us for this system with the Ames test. *Allium cepa* L (the common onion) is a natural material frequently used in bioassays, being considered the standard organism for quick tests, since it shows a high correlation with mammalian test systems. The aim of the present work was to determine the possible inhibitory action of L-ascorbic acid on the genotoxicity of the sulfathiazole-NaNO₂-HCl reaction mixture with the *Allium* test. Fito and genotoxicity were evaluated by means of *Allium* test, exposing the onion bulbs (7 bulbs per each concentration of analyzed substance) to increasing doses of the systems in study (the reaction mixture in presence and absence of increasing doses of L-ascorbic acid, sodium sulfathiazole, NaN₃ as positive control). Length and morphology of the roots were evaluated as macroscopic parameters. Mitotic index (MI, as the sum of meta, ana and telophase, evaluated over 5000 cell. Chromosome staining: Schiff reagent plus acetic orceine) as microscopic parameter. With the sulfathiazole-nitrite acidic mixture the MI diminished, being telophase the most affected one. This action was not reversed by the L-ascorbic acid in the assayed range of concentration.

76.

BLUE LIGHT PERCEPTION BY *Xanthomonas axonopodis* pv. *citri* AND ITS RELEVANCE IN BACTERIAL VIRULENCE

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Light is a major environmental stimulus. In living organisms light perception is carried out through photoreceptor proteins that belong to different families based on the structure of their light-absorbing molecules. LOV (Light, Oxygen, Voltage) domains are blue-light sensory modules that were originally described in plant phototropins. Data obtained from bacterial genome sequences revealed the presence of multiple photosensory proteins, whose physiological role are just beginning to be elucidated. *Xanthomonas axonopodis* pv. *citri* (Xac) is a gram negative bacterium responsible for citrus canker, a severe disease that affects the Citrus genus. The Xac genome sequence contains a gene, *lov*, encoding a LOV-protein. We constructed a *lov* knock-out mutant strain of Xac which showed alterations in processes related with the bacterial colonization of host plants. These include flagella synthesis, exopolysaccharide production and adhesion to plant tissues. Moreover, the symptoms developed in orange (*Citrus sinensis*) leaves inoculated with the *lov* mutant strain of Xac resulted temporally and phenotypically different than those generated by the wild type strain. These results demonstrate the novel involvement of a photosensory system in the regulation of the pathogenic process of Xac and constitute the first report of a functional LOV-protein in *Xanthomonas* spp.

77. INVOLVEMENT OF BIFUNCTIONAL CATALASE KatG FROM *Xanthomonas axonopodis* pv. citri IN THE OXIDATIVE STRESS RESPONSE

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Xanthomonas axonopodis pv. citri (Xac) is a Gram-negative obligate aerobic bacterium that infects citrus plants. Pathogenic bacteria are usually exposed to hydrogen peroxide (H₂O₂) produced either by normal aerobic metabolism or as part of the plant defense response against microbial invasion. In order to survive and colonize plant tissues Xac must overcome H₂O₂ toxicity, and catalases are the main enzymes employed for its detoxification. We have previously shown that Xac develops an adaptive response to H₂O₂, and the level of induced protection correlates with the bacterial ability to induce catalase activity during the adaptation treatment. Three catalase genes have been found in Xac genome, being one of them, katG, the one that codes for a bifunctional catalase-peroxidase enzyme. In this study, a XackatG mutant strain was generated by insertional mutagenesis. The effect of katG disruption on the catalase pattern was assessed by native gel electrophoresis and staining, demonstrating the absence of the two fastest-migrating bands present in wild-type cells. In contrast to wild-type bacteria, no induction of total catalase activity was observed in XackatG mutant after treatment with sub-lethal concentrations of H₂O₂, revealing the role of KatG in the adaptive response of Xac to this oxidant. During the interaction with host plants the mutant strain produced typical canker lesions to a similar extent of wild-type cells, indicating that katG disruption does not affect bacterial pathogenicity.

78. ANALYSIS OF HOST, BASAL AND NON-HOST *Citrus sinensis* RESPONSES AGAINST XANTHOMONAS

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Citrus are the most important fruit crops in the world. They are exposed to pathogens that produce important economic losses. The aim of this work was to study *Citrus sinensis* (orange) responses against bacterial phytopathogens of *Xanthomonas* genus. Infections with *X. axonopodis* pv. citri (Xac, canker agent), allowed to analyze the host compatible interaction, while Xac type III secretion system (TTSS) mutant (Mut), the basal response or changes caused by TTSS effectors. Citrus treatment with *X. a. pv. vesicatoria* (Xav), a pepper and tomato pathogen, was used to study the non-host incompatible response. The interactions were physiologically characterized and orange transcriptome determined at 8 h post-treatment (pt) using microarrays. The experiments were performed in triplicate using leaf tissue. No differences were observed during host and basal responses at early time pt, whereas necrotic lesions, bacterial death, production of reactive oxygen species and disruption of cell membranes similar to a hypersensitive response (HR), were evidenced at 24-48 h pt with Xav. During Xac disease 137 transcripts were differentially expressed ($p < 0.05$), of which 11 changed in comparison with Mut, candidate genes to be regulated by TTSS effectors. Also, 81 transcripts were similar between Xac and Mut, probably involved in basal defenses. During orange-Xav interaction, 1387 transcripts were differentially expressed when compared with Xac ($p < 0.01$), which be involved in orange HR. These results may help to understand the bases of citrus response to pathogens.

79. DIAGNOSIS OF *Neospora caninum* IN DOGS OF INFLUENCE AREA, FACULTAD DE CIENCIAS VETERINARIAS, UNR

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The protozoan parasite *Neospora caninum* has been found in dogs, cattle and other species in the world. It causes serious injury to the dog (type neuromuscular) and abortions and neonatal death in cattle. Phylogenetically close to *Toxoplasma gondii*, and misdiagnosed in many occasions, must be differentiated. The objective of this study was to determine the presence of antibodies to *N. caninum* in sera of dogs in the influence area, Fac. de Cs. Veterinarias, UNR. Teachers and students from three area schools conducted health professionals caring practices involving diagnosis, prevention and control of zoonoses in animals, in the period December 2010 to March 2011. It covered 131 dogs (64 males and 67 females) whose ages were less than 1 year (12 dogs), up to 5 years: (93) and over 5 years (26). Dog's blood and serum were processed in Area Parasitología, Fac de Cs. Bioq. and Farm. We performed ELISA with recombinant antigens of *N. caninum*: NcrSAG1 and NcrGra7. NcrGra7 has greater sensitivity to recent infections, whereas no significant difference NcrSAG1. Sera were tested at a dilution 1/100. ELISA for Toxoplasmosis (T) was also performed (differential diagnosis). Of the total samples (131), 13 sera (10%) had antibodies against NcrSAG1, NcrGra7 (not reactive to T). Animals may be checked by extending a subclinical form of the disease. It is important to prevention and control activities to prevent transmission in humans, the possible zoonotic potential of parasite.

80. BENZNIDAZOLE INDUCES A G0/G1 ARREST IN CELLS DERIVED FROM ACUTE MYELOID LEUKEMIA

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Previous studies from Pascutti et al. first reported a cytostatic effect of the trypanocidal drug Benznidazole (BZL) in epithelial cell lines. This finding prompted us to evaluate the effect of the drug on the growth of the leukemic cell lines, THP1 and OCI/AML3. We have found that BZL inhibits proliferation without inducing cell death by apoptosis or necrosis. Therefore, our aim was to evaluate in which cell cycle phase BZL stops the proliferation of THP1 cells. Results: THP1 cells treated with 1mM BZL for 24 h had a higher number of cells in G0/G1 phase compared to the control (66.9 ± 5.6 vs. $49.3 \pm 4.1\%$) and lower percentage of cells undergoing S (27.1 ± 1.5 vs. $41.3 \pm 3.4\%$), respectively ($p < 0.05$). Similar results were observed with 48 h of treatment. By immunoblot we observed increased expression of cyclin D1 both at 24 and at 48 h of treatment with BZL; while the protein levels of CDK2 and cyclins E, A and B do not show significant differences from control. In synchronized cells previously treated with 1mM BZL, we confirmed the cell arrest in G0/G1 phase with a consequent decrease in S phase ($p < 0.05$). Conclusion: BZL decreases THP1 cell proliferation by arresting the cell cycle in G0/G1 phase. This is accompanied by an increase expression of cyclin D1 suggesting that the arrest is in the restriction point before the G1 phase output.

81. INHIBITORY EFFECT OF BENZNIDAZOLE ON THE DEVELOPMENT OF SARCOMA E100 (SE100)

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Studies performed at the Immunology Institute have showed that the trypanocidal drug Benznidazole (BZL) inhibits the production of proinflammatory cytokines and the proliferation of cell lines through inhibition of NF-kappaB.

Our aim was to evaluate the antiproliferative capacity of BZL in an animal tumor model, SE100, both in its development *in vivo* and *in vitro*. Materials and Methods: 12 adult male rats of the line "m" were inoculated with SE100 (100% of implant and death) and were randomly divided into: control group received orally 1% carboxymethylcellulose (CMC) and treated group, received 200 mg/kg BZL in 1% CMC, administered daily and for 15 days after the tumor appeared. Results: BZL group showed smaller tumor size than control group ($p < 0.05$), and the immunoblot showed weaker expression of cyclin B and MDM2 (p53 inhibitor) ($p < 0.05$). There was a reduction of cell proliferation by BZL treatment in the *in vitro* cultures at 48 h ($p < 0.005$), without increased in LDH and metabolic activities comparing with control cultures. Conclusion: BZL inhibits the development of the SE100 in the *in vivo* studies associated with lower expression of MDM2, a scenario that could lead to proliferative arrest possibly by increasing the expression of p53 (cell cycle inhibitor). Concordantly in the *in vitro* studies BZL inhibits cell proliferation of SE100 with no evidence of cytotoxicity.

82. COLAGENAZA TYPE II: DENTINAL PULP COMPLEX OF MICE WITH TWO TYPES OF DIET AND GERMS DETECTION N DENTINE

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The conjunctive tissue of dental pulp and dentine mineralized is difficult to visualize in joint form. Caries, disease characterized by tooth hard tissue demineralization and destruction exposes the dentinal surface to mouth and germs causing infectious complex inflammation. Our aim was to apply a technique to visualize the joined dentinal pulp complex and to identify germs in dentine during carious evolution. Healthy and decayed teeth of 50 mice "P", 70 days age, Medical Sciences Faculty bioterium were studied. A half (G1) was fed by usual diet and the other one (G2) by saccharose solution 10%, for 5 weeks. In a sequential form 5 mice were sacrificed after having finished every week, occlusal caries were detected by Rodamina, magnifying glass and explorer. Maxillae were surgically extracted, fixed in formaldehyde 10%, demineralized in nitric acid 8%, fibres were eliminated with Type II collagenaza, included in paraffin, cut 5µ thickness, colored with Gram's technology Jensen's modification and examined with light microscope to 450 and 1000 increases. Decayed teeth were 61% in G1 and 81% in G2. Teeth with germs in dentine were 82% in G1 and 95% in G2 ($p = 0.002$). Germs Gram+ presence was $p = 0.50$, prevailing in dentine deep third G1 32% and 42% G2. Gram's- proportion was 17% in G1 and 43% in G2 ($p = 0.0003$), were distributed in 10% in G1 dentine superficial third and 31% in G2 superficial and average thirds. This technique allowed observation of dentinal pulp complex in joint form, and made possible to detect germs in mineralized components. In G1 minor proportions of caries and germs were observed. In both groups there was not difference in Gram+ location. Gram- were scanty and superficial in G1, prevailing in G2 in dentine superficial and average thirds.

83. DISEQUILIBRIUM FROM JENSEN SHANNON FOR COMPARING ERYTHROCYTES INCUBATED IN GLUCOSE MEDIUM

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The erythrocytes membrane is a viscoelastic material constituted by a protein cytoskeleton and a phospholipids bi layer. The high levels of erythrocyte membrane protein glycosilation on diabetic patients induced alterations on the erythrocytes viscoelasticity which could alter the microcirculation. Using the Erythrodeformeter, based on diffractometric technics, we could measure the fluctuations produce on the long axes of the elliptical diffraction pattern, when the cells are subjected to well controlled shear stress. In this work, we studied the relation between Jensen Shannon quantifier corresponding to samples from healthy donors and the alterations produced when are incubated *in vitro* with different glucose concentrations. The erythrocytes have been washed and resuspended in PBS on 4 aliquots: one without treatment and the other 3, incubated in glucose solutions (1%,2%,5%). These experimentals data where stored in time series (256 datos), and where process: a) we applied wavelet transform, choosing spline cubic as mother wavelet, and the constants for each level that is a, b are diadic, b) the values of disequilibrium (Qjs), where used for comparing the distances between probability distributions. The average results: $\langle Qjs(\text{con}) \rangle = 3509,218 \times 10^{-4}$, $\langle Qjs(1\%) \rangle = 3511,202 \times 10^{-4}$, $\langle Qjs(2\%) \rangle = 3515,571 \times 10^{-4}$, $\langle Qjs(5\%) \rangle = 3527,268 \times 10^{-4}$. This statistic measure could be used for comparison parameter on studying the reological properties of erythrocytes, improving the understanding of the alterations on blood microcirculation on diabetic patients.

84. INFLAMMATORY RESPONSE IN EXPERIMENTAL MODEL OF EXTERNAL ROOT RESORPTION

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The root resorption is a pathological process initiated by specific clastic cells that remove the organic and mineral components of dental hard tissues. The etiology of the different types of root resorption requires two phases: mechanical or chemical injury for protection tissues and stimulation by infection or pressure. The objective was evaluated the presence of inflammation in an experimental model of external root resorption in the rat molar. 30 male rats line "e" of 8 weeks of age were used. Rats were anesthetized with an intraperitoneal injection of a ketamine/xylazine solution. An intraoral pocket incision was made below both first lower molars. The cervical third of mesiovestibular root of first lowers molars was worn away with a round bur #1/2 involving dentine and cement. In the orifice there was not placed any filling. It was irrigated with sterile saline solution and the soft tissues were held in place. The animals were sacrificed at 7, 30 and 60 days to observe short and long-term inflammatory response. The dissection of the jaw was made. The samples were fixed with formol 10% and processed to make histologic studies. The images were classified based on the located inflammatory reaction found in the adjacent site to the wearing down: 0) no detectable inflammation; 1) disperses inflammatory reaction; 2) located inflammatory reaction; 3) diffuse, intense inflammatory reaction. No differences in the degree of inflammation between the 3 studied groups were observed: 7 días: 0.45 ± 0.605 ; 30 days: 0.20 ± 0.41 ; 60 days: 0.16 ± 0.375 . The results are expressed like average \pm standard error. These findings indicate that it is a valid model to evaluate different materials that could be used in the treatment of external root resorption since the made surgical maneuvers do not cause additional inflammation in the study site.

85. BACTERIOLOGICAL EVALUATION OF THE EFFECTIVENESS OF FLUORINE AS ANTIMICROBIAL THERAPY IN A DENTAL COMMUNITY PREVENTIVE PROGRAM

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Dental caries is of multifactorial etiology, *Streptococcus mutans* (Sm) has been implicated as an etiologic agent. It has been suggested that fluoride topical gels would be able to decrease the amount of (Sm) and would therefore be an important preventive maneuver. In previous studies a collection and processing technique of stimulated saliva in children was set-up. The aim of this study was to evaluate the effect of an acidulated phosphate fluoride topical gel 1.23% (APF) in the number of colony forming units (CFU) of Sm in saliva from children 6 to 8 years old. 44 children participating in a community prevention program were randomly selected. The approval and parental consent were obtained; a sample of stimulated saliva was gotten by chewing a piece of latex tube (2 cm) for 1 minute. Saliva was aspirated with a 3 ml sterile Pasteur pipette, and placed in a sterile centrifuge tube with screw cap, then packed in ice at 4°C for transport and processing; then were cultured on blood agar, Sm was identified by standard biochemical tests and the CFU were measured, (baseline sample). Four weekly applications of APF gel were carried out. Saliva sampling was repeated on the fifth week to assess the effect of treatment. The number of CFU in the baseline sample was between 2 x 10³ and 3 x 10⁵ and after treatment was between 3 x 10² and 6 x 10⁴. The reduction in the number of CFU was statistically significant (p=0.001). APF topical gel is a recommended maneuver in a preventive program, but should evaluate if its effect is beneficial over time.

86. PENTOXIFYLLINE AND SILDENAFIL EFFECT IN GASTRIC MUCOSAL INJURY MEDIATED BY STRESS IN RATS

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Pentoxifylline (PEN) and sildenafil (SIL) produce phosphodiesterase inhibition, and then an increase of the AMPc and GMPc. They produce vascular wall relaxation. Stress models of immobilization (IN) and immersion (in) produced catecholamines release, with vasoconstriction, therefore altering gastric mucosal defense. Aim: evaluate PEN and SIL action in gastric mucosa of rats submitted to stress with IN-in. Method: Randomized Female Wistar rat Groups (n = 7 each group), 350 g, 24hr fast, water ad libitum. Experiments: Group 1) 2 ml orogastric saline (OG), 60 min wait (control). 2) PEN 85mg/kg, OG, 60 min; 3) SIL 7.14 mg/kg, OG, 60 min; 4) PEN + SIL, OG, 60 min. Later rats were submitted at stress IN-in 18°C water 6 hrs. Rats were sacrificed by ether overdose, laparotomy and gastrectomy were performed, gastric necrosis area was tabulated by planimetry. Histology: (H.E.). Statistics: Student's t test and ANOVA. Macroscopical gastric area percentage: Results: 1) lesional area was of 50% ± 8.7 histology: hemorrhagic necrosis in 2/3 of the surface of gastric mucosa. 2) 18% ± 3.0 (p < 0.05) erosion area, little necrosis covering half the thickness mucosal, without hemorrhage. 3) 32% ± 7.2 (p < 0.01) necrosis compromises 1/3 of the surface of the mucosa with little bleeding on the surface. 4) 3% ± 1.5 (p < 0.001) only patches of mild edema on the surface. Conclusions: the addition of the effect using both drugs protects gastric mucosa against stress, more than each one alone.

87. IMPACT OF SATURATED FAT ACIDS IN BIOMASS AND GLUCOLIPIDIC PROFILE IN TWO ANIMAL MODELS

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Present-day diets mainly made of saturated fat acids (AGS), (AG) trans and (AG) Ω 6, have contributed that obesity and diabetes reach epidemic proportions. It was supplemented the animal habitual diet with grated coconut as vegetal source of AGS, increasing the impact in metabolism variables. Male b rats (insulinresistant later obese) and β (obese later diabetic) and eumetabolic Wistar (W) were fed from weaning (n: 5/group) with: habitual food: (BC), (bC) and (W), habitual food+AGS: (BS) and (bS). At 50, 100 and 200ds biomass (g), Gli0 and Gli120' and triglyceridemia (Tg) (g/l) were analyzed (ANOVA/Bonferroni) (media \pm SEM). At 50ds b expressed normal values. At 100ds diets with AGS have biomass higher values (p:0,0001), lower than Gli120' (p:0,0001) and TG (p>0.05). At 200ds the tendency of 100ds is kept, for biomass [β S: 482,2 \pm 8,7; bS: 469,3 \pm 10,1; β C: 452,4 \pm 11,4; bC: 436,9 \pm 1,8; W: 457,6 \pm 13,1] (p:0,0362), for Gli0 [β S: 1,14 \pm 0,09; bS: 1,16 \pm 0,02; β C: 1,76 \pm 0,03; bC: 1,32 \pm 0,03; W: 1,08 \pm 0,02] (p:0,0001), Gli120' [β S: 1,91 \pm 0,14; bS: 1,34 \pm 0,07; β C: 2,31 \pm 0,06; bC: 1,76 \pm 0,06; W: 0,87 \pm 0,07] (p:0,0001), for Tg [β S: 2,42 \pm 0,33 vs \hat{a} C: 2,64 \pm 0,42] (p>0.05); [bS: 0,98 \pm 0,09 vs bC: 0,88 \pm 0,07] (p>0.05)], W: 0,55 \pm 0,01. High intake of AGS of vegetal origin, affects the lipidic metabolism producing obesity, but not necessarily affects the glucidic metabolism, having decreased the diabetic progression in b. The different metabolization of these AGS, result of the different length of its chains, could be an explanation of different AGS glucotoxicity.

88. FAT ACIDS: THEIR INFLUENCE IN THE HEPATIC FUNCTION OF TWO OBESE DISMETABOLIC MURINE MODELS

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Evidences associate obesity with low grade chronic inflammation as origin of non alcoholic hepatic steatosis. Habitual diet of obese and dismetabolic b and β male rats was supplemented from weaning to 200ds with vegetal sources of fat acids (AG) (n:6/group): habitual food(C), habitual food+chia seeds(CH) [Ω 3], habitual food+sunflower seeds(G) [Ω 6], habitual food+high oily sunflower seeds(GO) [Ω 9] and habitual food+grated coconut(CO) (saturated AG). Eumetabolic Wistar (W) received habitual food. Hepatic enzyme levels were evaluated as indirect inflammatory indicators. At 100 and 200ds were analysed (ANOVA/Bonferroni) alanine aminotransferase (ALAT), aspartate aminotransferase (ASAT), alkaline phosphatase (FAL) (mUml). ALAT 100ds: β C: 75 \pm 1.6; bC: 51 \pm 0.6; β CH: 72 \pm 0.8; bCH: 61 \pm 0.2; β G: 74 \pm 4.9; bG: 57 \pm 3.5; β GO: 68 \pm 3.3; bGO: 59 \pm 1.5; β Co: 76 \pm 1.5; bCo: 55 \pm 2.8; W: 47 \pm 1.9 (p:0.0001). ALAT 200ds: β C: 50 \pm 0.5; bC: 56 \pm 3.4; β CH: 58 \pm 1.1; bCH: 62 \pm 2.3; β G: 49 \pm 2.2; bG: 86 \pm 6.8; β GO: 49 \pm 4.0; bGO: 66 \pm 1.9; β Co: 49 \pm 2.1; bCo: 60 \pm 2.8; W: 48 \pm 1.3 (p:0.0001). At 200ds FAL has higher obstruction at β with all treatments (p:0.0001). β has hepatic inflammation at 100ds decreasing at 200ds. b has an increase at 200ds. This and other studies evidence an inflammatory peripuberal process in β that start in adipose disfuncional tissue that resound at systemic level (liver). Instead, in line b, the process possibly start with a hepatic postprandial inflammation that would later resound systematically.

89.

EFFECT OF INSATURATED FAT ACIDS IN THE GLUCOLIPIDIC PROFILE OF IIMb LINE RATSOgusuku M¹, Gutiérrez S¹, Alle G¹, Coria M¹, Echenique C², Gayol MdC¹¹Cát.Biología. Fc. Cs. Médicas, ²Área Parasitología. Fc. Cs. Bioq. y Farm. UNR. E-mail: orinchi@hotmail.com

Diet insaturated fat acid (AG) metabolism produces eicosanoids involved in inflammatory processes, depending on the modulating effect of its origin. It was evaluated the influence of AG Ω 3, Ω 6 and Ω 9 in the IR of inbred IIMb (b) line rat, supplementing the habitual diet. Since weaning it was given (n:5/group) habitual food (C), habitual food+sunflower seeds (G), habitual food+chia seeds (CH) and habitual food+high oily sunflower seeds (GO). Eumetabolic Wistar rats (W) were given habitual food. At 50, 100 and 200ds (g/l) Gli0 and Gli120', triglyceridemia (Tg) and cholesterolemia (Col) were analysed. ANOVA is expressed as (media \pm SEM). At 50ds normal values were expressed. 100ds: the inflammatory effect of the supplements is observed. CH has Gli0 and Gli120' lower values (p:0.001) but Tg higher values (p:0.001) with probable effect over IR action that would lead to a lower triglyceride grasping of adipose tissue. At 200ds, 100ds tendency is considered: Gli120 [G:1.94 \pm 0.19, CH:1.24 \pm 0.01, GO:1.81 \pm 0.16, C:2.06 \pm 0.11, W:0.85 \pm 0.03] (p:0.001) and Tg [G:0.65 \pm 0.10, CH:1.44 \pm 0.09, GO:0.67 \pm 0.12, C:0.88 \pm 0.06, W:0.57 \pm 0.05] (p:0.001). Col does not differ among diets. Biomass (g) is higher in G and GO (anabolic effect) [G:487,1 \pm 13,4, CH:429,5 \pm 10,2, GO:475,1 \pm 15,7, C:435,2 \pm 1,4] (p:0.01). Supplement inflammatory modulating potencial lies in the metabolites, so the eicosanoid effect derived from the AG Ω 6 would increase the inflammation (proinflammatory diet), the derived from AG Ω 3 would have attenuating effect (anti-inflammatory diet) while the derived from AG Ω 9 would have an intermediate effect (inflammatory diet).

90.

MULTIDIMENSIONAL ANALYSIS OF VARIABLES RELATED WITH PRODUCTION IN DISMETABOLIC LINES eSS, BETA AND eSMT RATSTorres N, Gayol MdelC, Martínez SM, Montenegro S, Tarrés MC
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The reproductive sphere is often affected by diabetes and obesity, having done important contributions studies done with murine models. This work has the aim to characterize multidimensionally rats of eSS, beta and eSMT dismetabolic lines and build typologies that group the analysed cases according to their affinities in variables associated with productivity. The sample included 874 registers (22% eSS, 66% beta and 12% eSMT) that performed all the inclusion criteria. Birth took place in 57% of cases. The mean \pm SD of the considered variables were: maternal age (MA) = 132.9 \pm 50.1 days and paternal (PA) = 157.15 \pm 67.43 days, maternal weight (MW) = 214.52 \pm 42.35 g and paternal (PW) = 339.3 \pm 79.64 g, days from mating to birth (d) = 29.31 \pm 8.84, litter size at birth (n) = 7.73 \pm 3.29 and at weaning (s) = 6.8 \pm 3.5 pups, fertility coefficient (FC=7n/21+d) = 1.15 \pm 0.5 and of survival (SC=s/n) = 0.86 \pm 0.29. Data were analysed through main components, choosing as active variables MA, PA, MW, PW and FC and as illustrative the genetic line, presence or absence of partition, d, n, s and SC. The two first components explained the 78% of the total variance, principally connecting factor 1 with parent age and weight and factor 2 with mating fertility and the pup survival. Through classification technics, five clusters were defined, according to the variables that made significant contribution. The space configuration showed better fertility in eSS and eSMT diabetic lines, while beta lower productivity could be connected to their "diabesity".

91.

EVALUATION OF ADIPOSITY BY MURINOMETRICS INDEX IN SPONTANEOUSLY DIABETIC 12 MONTH-OLD eSS AND eSMT RATS

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eSS and eSMT rats spontaneously develop complex type 2 diabetic syndromes with higher expression in males. In both lines, ageing and body weight, witch is influenced by the nutritional environment, are risk factors of their metabolic disturbances. In order to find reliable indicators of visceral fat accumulation, a transversal study was carried out in eSS and eSMT male and female rats kept under the usual conditions of breeding. At 12 month of age, body weight (BW), length of tail (LT), naso-anal length (NL), total length, abdominal circumference (AC) and thoracic circumference (TC) were registered and Lee index (LI:BW/NL) and BMI (BW/NL) were calculated. Student's t-Test for independent samples (between sexes and lines) was used. Body weights were similar in eSS and eSMT females while AC, TC, LT and BMI were higher in eSS. Higher values of AC, TC and LT were verified in the eSMT males. In eSMT, all variables were positively correlated with sex (p=0.000); in contrast, no differences in AC (p=0.975) and TC (p=0.135) were detected between male and female eSS rats. It is interesting to note that up 12 months of age, while in both sexes weight did not differ between lines, the body conformations were quite different. BMI was a better indicator than LI of the highest visceral obesity in the males, consistent with the characteristic greater resistance to insulin in this sex in both lines, probably because the influence of the long of tail in LI.

92.

WEIGHT, CORPORAL FAT AND GLUCOLIPIDIC PROFILE ASPECTS IN CBI/01 AND CBI/02 FEMALE MOUSE SUBSTRAINSGayol MC^{1,2}, Chapo G¹, Menoyo Inés³, Zubillaga Patricio², Gayol MdelC², Tarrés MC^{2,4}¹Bioterio Central. ²Cátedra de Biología. ³Cátedra de Química Biológica. Facultad de Ciencias Médicas. ⁴CIUNR.UNR. E-mail: mgayol@hotmail.com

The mostly used animal species in scientific experimentation is the mouse (*Mus musculus*) and the association of the peculiarities fixed in each, generates an individuality related to their qualities, that must be taken into account when choosing the model for a research work. Phenotypic aspects of mouse CBI/01 and CBI/02 glucolipidic profile, coming from the same line and kept as reproductive closed colonies and in independent breeding rooms were characterised. At 240ds of age, in 25 females of each subline, it was registered: weight: W g, noseanal length NAL cm, tail length: TL cm and the Body Mass Index was calculated BMI: W/ NAL². It was dosed, in fasting, glucemia: G, triglyceridemia: TG and total cholesterolemia: COL (mg/dl). The animals were euthanased, and the Related Abdominal Fat (AFr %) was calculated. The mouse handling followed the international bioetic recommendations. The results were expressed as (mean \pm SEM): (CBI/01 vs CBI/02) W (56,8 \pm 4,6 vs 53,09 \pm 6,5) (p<0,05) BMI (0,42 \pm 0,03 vs 0,34 \pm 0,03) AFR (8,9 \pm 1,9 vs 4,39 \pm 2,3) and COL (116,1 \pm 14,3 vs 94,96 \pm 13,03) (p<0,001). CBI/01 has shorter length than CBI/02 (p<0,001). Both strains present fasting hiperglucemia (>126mg/dl) and normal lipidemias. The correlation with BMI, was direct in CBI/01 : r BMI /COL=0,401 (p=0,0524), being the rest not significant. These aspects, provide phenotypic information according with the "Mouse Phenome Database Project" as possible models to understand the complex human biology and pathology.

93. PHENOTYPIC STABILITY OF VARIABLES RELATED TO FERTILITY AND LACTATION IN TWO MOUSE CBI/01 FILIALS

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The experimental results can be influenced by the analysis units, so biological property knowledge of the animal used is required. Due to this, it was interesting to value if in albino female mice belonging to CBI/01 substrain population, inbred in Bioterio Central, Facultad de Ciencias Médicas, UNR, the fertility and lactation characteristics were kept stable or varied (with the homocigosis increase owing to consanguinity). Two filials distant 8 years were analysed: F58 (10female, 2003) and F81 (9females, 2011). Polygamic mating was joined (2 or 3 females x male) and being pregnancy verified, the future mothers were placed in individual birth trays. At birth time it was registered: days from mating to parturition (d); total number of born individuals (n), separated by sex (nM) and (nF) and litter weight (LW) (g). The offspring were weaned at 21 ds of age. Determination of sex was done, registering individual weight, and litter total weight (WTW). Fertility (FC=7N/21+d) and Survival Coefficients (SC=S/n) were calculated. At puberty, 49 days of age, the individual female and male weight was registered. (PFW and PMW). Results: expressed as mean±SEM: (F 58 vs F81): d, n, nM, nF, FC, SC: (p>0,05). WLW (19,28 ± 1,06 vs 22,84 ± 1,2) (p<0,05), WTW (137,7 ± 10,6 vs 174,1 ± 10,7) (p<0,05). PMW (36,9 ± 0,28 vs 40 ± 0,3) and PFW (32,6 ± 0,24 vs 34,3 ± 0,32) (p<0,001). The fertility value variables, and their stability among generations, would produce a good reproductive aptitude of CBI/01 substrain. The present generation higher mouse weight could be attributed, to modifications in the environment, not discarding dispersive process actions.

94. DIFFERENCES IN THE CORPORAL CONFORMATION OF MICE IN CBI/01 AND CBI/02 SUBSTRAINS

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CBI/0 mouse lines, began in 1980 in Bioterio Central de Facultad de Ciencias Médicas, UNR. In 1983, the base population was divided, producing two colonies that are bred in an independent way and in different rooms. They can be considered sublines, named CBI/01 and CBI/02 having at present more than 80 generations. To investigate if the substrains keep similar corporal conformation phenotypic characteristics, 25 females of each subline coming from matings produced in simultaneous way and of litters in a number between 8 and 12 offspring were evaluated since weaning. Weight (Wg), noseanal length (NALcm) and tail length (TLcm) at weaning (21 ds), puberty (49ds) and adulthood (90ds) were registered. Results were expressed as mean±SEM:

In W21 and 49, CBI/01 vs CBI/02 (p>0,05), and W90 (43,22±2,4 vs 39,54±6,6)(p<0,001). CBI/02 is longer at the 3 ages: NAL 21(p<0,05), 49 and 90(p<0,001), TL 21, 49, 90 (p<0,001). The correlation between NAL and TL were direct and significant at 21ds in both sublines (p<0,0001), at 49 and 90ds, they did not show any correlation in CBI/01 (p>0,05) and were positive in CBI/02 (p<0,001). The Body Mass Index (W/ NAL²) did not differ at 21ds (0,25±0,02 vs 0,24±0,01) (p<0,05) and it was higher in CBI/01 at 49 (0,336±0,02 vs 0,313±0,02) and 90 ds (0,36±0,03 vs 0,31±0,02)(p<0,001). The four variables increased with age (repeated measurements ANOVA: p<0,0001). CBI/01, of higher BMI, is of compact contexture and CBI/02 is of long structure, differences possible connected with the deriva phenomena not neutralized for genic flow among population (migration absence).

95. RED BLOOD CELLS AGGREGABILITY IN SYSTEMIC ERYTHEMATOSUS LUPUS

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Red blood cells alterations that affect its aggregation can be demonstrated by measuring into an aggregating environment from certain polimer. The way red blood cells are aggregated shows their intrinsic capacity to aggregate and its called aggregability. The aim of this study its to analyze the aggregability of red blood cells in Sistemic Eritematous Lupus patients. Red blood cells from Sistemic Eritematous Lupus patients (n=30) and from healthy people (n=30) were washed a resuspended to 40% Hto in dextran 500 (aggregating polimer) in 2% physiologic solution. Erythrocyte aggregation was estimated by measuring variations of time using light trasmitted through a whole blood sample, obtaining: s0/n0 meaning mean aggregation size, and 2k2n0 meaning initial process rate. "t de Student" was considered for the stadistical analisis. Findings show Sistemic Eritematous Lupus patients have significantly higher values from healthy controls: s0/n0 (1.92±0.05 vs 1.80±0.07, p<0.002), 2k2n0 (1.61 ±0.71 vs 1,18±0.61, p<0.001). Increased erythrocyte aggregation in Sistemic Eritematous Lupus patients should be explained by the higher aggregability of red blood cells, thus these findings could be related with the increased rate of thrombosis in Sistemic Eritematous Lupus patients

96. STUDY OF THE CORRELATION BETWEEN ERYTHROCYTE DEFORMABILITY AND MEMBRANE LIPIDIC FLUIDITY IN PATIENTS WITH SYSTEMIC LUPUS ERYTHEMATOSUS

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The aim of our work was to study the membrane lipid fluidity and its relationships with the erythrocyte deformability in red blood cells from Systemic Lupus Erythematosus patients compared with red blood cells of a group of healthy patients. The erythrocyte deformability was estimated by its inverse, the rigidity index (RI) obtained by the time required for the filtration of a suspension of erythrocytes through pores of 5µm in diameter. The lipid fluidity was measured by fluorescence polarization with TMA-DPH (Trimethylammonium-Diphenylhexatriene) as a marker. The anisotropy (r) is inversely related with the fluidity. Statistical analysis used was "t Student" and the Pearson correlation coefficient. The results showed that women with SLE had significantly higher values compared to control women: IR (10.03 ± 3.89 vs 7.11 ± 1.26, p<0.01) and r (0.18 ± 0.15 ± 0.03 vs 0.01, p<0.001). It was also found a statistically significant correlation between IR and r (r = 0.57, p<0.02). It is likely that lipid peroxidation of the erythrocyte membrane present in patients with Systemic Lupus Erythematosus causes loss of membrane lipid fluidity and decreased erythrocyte deformability.

97.

EFFECT OF LIPID PEROXIDATION ON ERYTHROCYTE AGGREGATION IN PATIENTS WITH SYSTEMIC LUPUS ERYTHEMATOSUS (SLE)Torrente J¹, Zilli M¹, Svetaz MJ², Leroux B³, Dimattia J¹, Carrara P¹, Petrelli D¹, Spengler MI¹¹Cátedra de Física Biológica, Fac. de Cs. Médicas, ²Sección Inmunidad Celular, Dpto de Bioquímica Clínica, Fac. de Cs. Bioq. y Farm., ³Médica Dermatóloga. Universidad Nacional de Rosario. E-mail: isabelspengler@hotmail.com

The goal of this work was to study the erythrocyte aggregation and lipid peroxidation of red cells in a group of 30 women with SLE and correlate both variables comparing them with a group of healthy women. Both parameters of erythrocyte aggregation process were determined by optic method in the EDTA anti-coagulated blood part: s0/n0 estimates the size of the aggregates, and 2k2n0 estimates the initial velocity of the process. In the Heparin anticoagulated part erythrocyte membrane lipid peroxidation was determined measuring the concentrations of Malonildialdehido (MDA) by TBARS. The results showed that women with SLE had higher levels, comparing them with the group of healthy women, of: MDA (nmol /ml of erythrocyte package) (3.64 ± 0.71 vs 2.95 ± 0.61), s0/n0 (1.89 ± 0.06 vs 1.80 ± 0.10 , $p < 0.001$) and 2k2n0 (1.02 ± 0.71 vs 0.70 ± 0.49 , $p < 0.02$). Also, a significant correlation was found between MDA concentration and s0/n0 ($r = 0.48$; $p < 0.05$) and between MDA concentration and 2k2n0 ($r = 0.49$, $p < 0.05$). The increment of lipid peroxidation is a sign of structural membrane damage. The changes in the lipid membrane by free radicals determine the increment of the erythrocyte aggregation parameters. These modifications could show that the free radicals, which produce peroxidation of the lipid membrane, may alter the physical and chemical properties of the erythrocyte membrane, modifying its aggregability.

98.

STUDY OF OSMOTIC RESPONSE IN RED BLOOD CELLS WITH SYSTEMIC LUPUS ERYTHEMATOSUS AND ITS RELATIONSHIP WITH DEFORMABILITYZilli M¹, Torrente J¹, Svetaz MJ², Leroux B³, Parente F¹, Dimattia J¹, Petrelli D¹, Spengler MI¹¹Cátedra de Física Biológica, Facultad de Ciencias Médicas, ²Sección Inmunidad Celular, Departamento de Bioquímica Clínica, Facultad de Ciencias Bioquímicas y Farmacéuticas, ³Médica Dermatóloga. Universidad Nacional de Rosario. E-mail: isabelspengler@hotmail.com

We study the osmotic response and the deformability of red blood cells in patients with Systemic Lupus Erythematosus (SLE) and compare the values we found in healthy control patients with the ones obtained from Lupus patients in order to find out the relationship between both variables. We study 30 women (40+/- 5) with SLE and we repeat the study in 30 healthy controls of similar age. All patients and healthy controls have signed the informed consent. The erythrocyte deformability was measured by the filtration of erythrocytes with polycarbonate membranes with 5mm pores and it was estimated using its inverse: the rigidity index (RI) obtained from filtration. Osmotic fragility was determined using red blood samples anticoagulated with heparin into ClNa solutions from 0 to 0,145 M. We calculated the concentration of ClNa in which we obtained the 50% of hemolysis (x50). The results showed that SLE patients have significantly higher values compared with healthy controls of: IR (9.84 ± 4.78 vs 7.11 ± 1.26 ; $p < 0.02$) and x50 (g/dl) (0.381 ± 0.030 vs 0.357 ± 0.036 ; $p < 0.05$). We also found a significant statistical correlation between IR and x50 ($r = 0.81$, $p < 0.005$). A decrease in flexibility of red blood cells membrane cannot resist changes in cellular volume.

99.

EFFECT OF IMMUNOGLOBULINS IN ERYTHROCYTE AGGREGATION OF PATIENTS WITH SYSTEMIC LUPUS ERYTHEMATOSUS (SLE)Dimattia J¹, Svetaz MJ², Leroux B³, Petrelli D¹, Spengler MI¹¹Cátedra de Física Biológica, Facultad de Ciencias Médicas, ²Sección Inmunidad Celular, Departamento de Bioquímica Clínica, Facultad de Ciencias Bioquímicas y Farmacéuticas, ³Médica Dermatóloga. Universidad Nacional de Rosario.

In this work we set out to study the association between plasma concentration of immunoglobulins and the parameters characterizing erythrocyte aggregation in the blood of female patients with SLE. Venous blood was obtained from 30 female patients with SLE and 30 healthy control women of similar age. All had signed written informed consent. The plasma concentration of immunoglobulins (Igs) was determined by radial immunodiffusion. The two parameters of erythrocyte aggregation process: s0 / n0 which estimates the average size of the aggregates and 2k2n0 estimates the initial velocity of the process, were determined by optical method. The results showed that women with SLE had significantly higher values with respect to control women, to: Immunoglobulin plasma concentration (mg / dl) (1803 ± 521 vs 1411 ± 430 ; $p < 0.005$), s0/n0 (1.89 ± 0.06 vs 1.80 ± 0.10 , $p < 0.001$) y 2k2n0 (1.02 ± 0.71 vs 0.70 ± 0.49 , $p < 0.02$). It was also found a statistically significant correlation between Igs and s0 / n0 ($r = 0.45$, $p < 0.05$) and between Igs and 2k2n0 ($r = 0.49$, $p < 0.05$). The increase in plasma concentration of immunoglobulins could favor the increase of intercellular bridges between erythrocytes and so increase erythrocyte aggregation. This increase would be capable of inducing a decrease of flow, primarily in the microcirculation, causing damage to tissue perfusion.

100.

METABOLIC EFFICIENCY RELATED TO DIFFERENT SOURCES OF DIETARY CALCIUM IN ADULT β RATS

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Obesity is a multifactorial disorder. Genetic and environmental factors contribute to its development. Dietary calcium has been proposed to have beneficial effects on body weight control. A critical point in calcium supplements is their bioavailability.

The aim of this study was to evaluate the effects of two high calcium diets differing in the source of the mineral on metabolic efficiency of obese β line rats. Twelve 200 days old obese male β rats, were fed a diet containing 1.2 g% of calcium provided either by non-fat dry milk (group L) or CaCO₃ (group H) for 45 days. Both diets were isolipidic and isocaloric. Animals body weight and food intake were measured every other day. Metabolic efficiency was estimated by food efficiency conversion, which was calculated as the ratio total food intake / body weight gain. Comparisons between groups were performed using Student's t test and differences were considered significant when $p < 0.05$. Data are presented as mean \pm standard deviation. Results: total food intake (g): L: 629.40 ± 24.17 vs. H: 833.20 ± 33.22 ($p < 0.01$); body weight gain (g): L: 8.92 ± 6.26 vs. H: 57.96 ± 10.76 ($p < 0.001$); food efficiency conversion (%): L: 1.43 ± 1.03 vs. H: 5.90 ± 1.12 ($p < 0.001$). Conclusion: lower metabolic efficiency of group L (beneficial effects on obese syndrome management) might be due to a higher bioavailability of calcium in dairy products. It is also probably that other bioactive compounds present in milk, such as lactose, peptides and fatty acids have synergistically acted.

101.
SKELTAL GROWTH OF β RATS FED A LOW CALCIUM DIET

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Most of epidemiological studies that related calcium intake to bone health are focused on evaluating the quality or mineralization grade of the bones but not on their growth in length.

The aim of this study was to compare the effects of a low calcium diet (h: 0.2 g %) versus a normal one (N: 0.6 g %) on the skeletal growth of obese β line rats. Animals were placed on h diet (n: 11) or N diet (n: 15) since 21 (weaning) till 51 days old and tail length were measured every other day. The von Bertalanffy growth curve ($L_t = A \cdot (1 - b \cdot e^{-k \cdot t})$) parameters "A" and "k" were calculated. L_t : tail length at time t. A: asymptotic tail length value, it indicates media tail length of mature animals. k: tail length maturation ratio. Comparisons between groups were performed with Student's t test and differences were considered significant when $p < 0.05$. Data are presented as (mean \pm standard deviation). Results: inicial tail length (cm): h: 8.64 ± 0.92 vs N: 7.98 ± 0.55 (ns); final tail length (cm):: h: 14.23 ± 1.12 vs N: 14.11 ± 1.07 (ns); A: h: 19.89 ± 1.81 vs N: 19.38 ± 2.15 (ns); k: h: 0.035 ± 0.011 vs N: 0.035 ± 0.010 (ns). Conclusion: calcium deficiency in diet did not limit the skeletal growth of β line rats and the maturity rate.

Although dietary calcium exerts an important role on osseous tissue density it seems that it has not the same great importance in the length that bones reach.

102.
SKELTAL GROWTH OF β RATS FED A HIGH CALCIUM DIET

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Post weaning skeletal growth depends on genetic and environmental factors such us nutrition and physical activity. Most of epidemiological studies related to calcium intake it are focused on bones mineralization but not on their growth in length.

The aim of this study was to evaluate the effects of a high calcium diet on the skeletal growth of obese β line rats.

Weaning male β rats were fed an AIN diet containing 0.9 g% of calcium (group H, n: 13) or 0.6 g% of calcium (group L, n: 15) for 30 days. Animals tail length were measured every other day. The von Bertalanffy growth curve ($L_t = A \cdot (1 - b \cdot e^{-k \cdot t})$) parameters "A" and "k" were calculated. L_t : tail length at time t. A: asymptotic tail length value, it indicates media tail length of mature animals. k: tail length maturation ratio. Comparisons between groups were performed with Student's t test and differences were considered significant when $p < 0.05$. Data are presented as (mean \pm standard deviation). Results: inicial tail length (cm): H: 9.15 ± 2.17 vs N: 7.98 ± 0.55 (ns); final tail length (cm):: H: 14.28 ± 1.65 vs N: 14.11 ± 1.07 (ns); A: H: 19.76 ± 1.95 vs N: 19.38 ± 2.15 (ns); k: H: 0.031 ± 0.007 vs N: 0.035 ± 0.010 (ns). Conclusion: higher calcium intake by feeding H diet, did not modify bone length of male β rats nor their maturity rate. Maximum bone growth is strongly influenced by genes and factors related to nutrition might modify the skeletal growth just in case nutrients (e.g.: calcium) are present in a low level but not when they are in excess.

103.
CHARACTERIZATION OF HALOTHANE ANESTHESIA IN 100 AND 200 DAYS OLD CBI/L MALE MICE

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Experimental procedures can lead to distress in research animals. Anesthesia administration might modulate this response, and must be adjusted to each individual. To characterize sedoanalgesia, clinical variables were registered in mice selected for large body and low weight. 17 CBI/L male mice were anesthetized with halothane, at the age of 100 and 200 days. Halothane was vaporized at atmospheric pressure in 100% oxygen, at 6 l/min for 40 min. Variables were registered in a specially designed anesthetic chart. Sedoanalgesia was reached when animals showed simultaneously: negative response to tail clamping, no response to handle and they were in a state of sleep. Optimal sedoanalgesia was obtained in less than 5 minutes after halothane administration. Recovery features were also characterized. Data are presented as median (min-max) and analyzed by using non parametric tests. $P < 0.05$ was considered significant. Optimal sedoanalgesia was observed in all 200 days old (DO) animals and only 17% in the 100 DO group ($p < 0.001$). Sedoanalgesia was longer in 200 DO: 43 (35-45) min. than in 100 DO: 45 (41-50) min. mice; $p < 0.001$. Also, 100 DO mice showed a delay to reach sedoanalgesia: 7 (3-10) min, than 200 DO: 3 (2-5) min. No differences in the recovery period were observed. We conclude that halothane anesthesia was optimal for 200 DO mice but not for 100 DO group, who need more time to achieve sedoanalgesia. These facts might be due by diminished neuronal population or less unspecific binding sites for anesthetics in 200 days old mice.

104.
PASTEURIZED SKIMMED COLD MILK. BEST METHOD OF CONSERVATION OF THE PERIODONTAL LIGAMENT IN DENTAL AVULSION

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In the avulsed tooth, no element of the oral mechanism is so tied to the functionality, aesthetics and emotions as the state of dentition. Therefore, the requirement is the recovery and restoration of broken teeth or avulsed and the supporting bone, knowing the healing capacity of each cell type of tissue compartments. The method of storage before replantation is a factor that determines the root resorption in the future. Objectives :1-perform clinical and pathological data obtained from extracted teeth in dental services and offices 2-analyze and recognize the viability of the periodontal ligament in various means of preserving avulsed tooth recognize the best 3-way preserve the tooth for later reimplantation. Material and methods: total: 120 teeth with closed apices, extracted. Transport: dry solution, saline, water, pasteurized milk cold. Times the levels of 5, 10, 15, 20, 30 and 60 minutes, with subsequent immersion in 10% formalin with routine processing. Preliminary Results: Given the difficulty of collecting the teeth were processed analytical conditions received 80 pieces of 63 were studied to date, of which 30 were kept in cold pasteurized milk 15 in saline, 8 in tap water , 5 dry at room temperature and 5 in 10% formalin (as controls) Partial Conclusions: Milk 5': active fibroblasts, vascular tissue, without inflammatory cells. Milk 10': active fibroblasts, vascular tissue, the presence of inflammatory cells. Milk 15': active fibroblasts, inflammatory cells and edema. 20 milk means milk the same 15'. Milk 30': ditto. 60 The decline in milk active fibroblasts and vessels. Salina: 5.15 and 30 min. A large number of inflammatory cells and edema. The presence of bacterial colonies. Running water: a large number of colonies of bacteria, inflammatory cells, tissue necrosis. Was quantified with a microscope characteristics of the cells of the grid. Dry at any time provided there are no viable fibroblasts.

105.
EFFECT OF SALINE STRESS ON GERMINATION OF
Lycium cuneatum

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In the Forest Wedge of Santa Fe, Argentina, inside the Oriental Humid Chaco, native forest has constantly been degraded; which has produced widespread erosion and an increase of salinization. The aim of this paper was to determine the effect of different saline concentrations on germination of *Lycium cuneatum*. A harvest of these seeds was done (march, 2010) in a "quebrachal" of the Centro Operativo Experimental Dr. Tito Livio Coppa, "Las Gamas", province of Santa Fe (29° 28' S, 60° 28' W 58 m.s.n.m). The seeds were placed in plastic boxes with moistened sand to field capacity. Four treatments were made, each one with four repetitions of 50 seed. NaCl at 0.2; 0.25 y 0.3 Molal were used for the treatment; the witness was moistened with H₂O. Seeds were disinfected with sodium hypochlorite during two minutes. These boxes were taken to a chamber with 12 hours of photoperiod and 25°C. The germination speed index was calculated (IVG) and the average time of germination (TMG). The results were analyzed with Anova, a then with a Multiple Range Test to identify significant differences. These were found between the witness and the treatments. What follows that salt presence affects germination of *Lycium cuneatum*. Therefore a slight increase in salt in surface will affect negatively regeneration of this specie in anthropized environments.

106.
X-Ray: CHARACTERIZATION OF SEEDS AND FRUITS OF
NATIVE FOREST SPECIES OF FOREST WEDGE,
ARGENTINA

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The X-Ray technique is greatly known and used in medicine, biology and industries and it is very useful in studies of seeds and fruits quality by its anatomy and morphology. The objective of this work was to create radiographic patterns that allowed to characterize woody native and cultivated seed and fruit species by its morphological and anatomical characteristics, physical damages and abnormalities in the maturation process. Radiographic plates of fruits and seeds were obtained with the X Ray Equipment RUTAX and traditional revealed of the plates were made. There were taken 100 radiographic plates of the fruits of each one of the shrubs and forest species studied. This radiographic images were compared with their respective digital photographic images. The patterns were made with radiographic images and its corresponding digital photographic images considering: whole seeds and seeds completely developed; seeds not completely developed; empty seeds; seeds damaged by insects and broken seeds. Forest species generally have seeds with hard teguments and a great quantity of empty seeds. The X-Ray Rutax equipment allowed to make an efficient and quickly characterization of seeds and fruits of forest species.

107.
LIQUID-CRYSTALLINE EMULSIONS WITH TERBINAFINE
SULFATE FOR TOPICAL APPLICATION

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Liotropic liquid crystals proved to be adequate systems for the controlled release of drugs, which is why it is expected for the liquid-crystalline emulsions a similar behaviour. The liquid-crystalline emulsions presented, according with Susuki, Takei and Yamazaki, greater stability. In this work we studied conventional emulsions (not liquid-crystalline) and liquid-crystalline emulsions, with and without added of sulfate of terbinafine, an antifungal drug which is forming mainly part of the oily phase at the pH of the emulsions. It was tested four different emulsions: two prepared by a conventional method without the formation of liquid crystals, of which one was taken as a reference (sample 1) and the other was added 0.1% of terbinafine sulfate (sample 2); and two other emulsions prepared by a method for formation of liquid crystalline interphases, the first was taken as a reference (sample 3) and the other was added 0.1% of terbinafine sulfate (sample 4). Terbinafine sulfate was added to the aqueous phase formed by a mixture of triethanolamine and methyl paraben. The alkalinity of the aqueous phase made that terbinafine becomes the oily phase in the form of base. The formation of liquid-crystalline structures was found using a polarizing microscope Carl Zeiss Axiolab fitted with digital camera Olympus SP 35. The emulsions (1) and (2) did not show liquid-crystalline structures and emulsions (3) and (4) presented liquid-crystalline structures. The proposed methodology allows to obtain liquid-crystalline emulsions with the active substance incorporated both in the oily phase and in the liquid-crystalline structure.

108.
PERCEPTIONS AND ATTITUDES OF OCCUPATIONAL
RISKS IN DAIRY FARMERS

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Livestock and agriculture are among the economic activities with the highest number of occupational accidents (OA). Our objectives were to estimate risk perception, OA and safety practices in dairy farming. A census was carried out on 75 farmers associated to a dairy cooperative in Paraná, Entre Ríos using a structured questionnaire in a cross-sectional design. Statistical analysis included χ^2 and Spearman correlation. Sixty percent milked less than 47 cows, 56% had suffered at least one OA, 84% required medical care, 22% hospitalization and 81% had to stop working (78.4±145.1 days). Although 42% identified brucellosis as a zoonoses, 58% were never tested to detect the disease. Higher risks were perceived in main road transit, agrochemical and animal management. Use of tractors, machinery, tools and grinders were inter-correlated, r from 0.40 (p=0.0032) to 0.81 (p < 0.0001). Risk perception during abortion and parturition manipulations were correlated (r=0.71 p<0.0001). Sixteen percent wore gloves, 12%, mask and 11% specific clothing when working with herbicides, insecticides and pesticides. Glove usage during parturition (41%), abortion (36%) and dead animal handling (24%) were inter-correlated (r= from 0.66 to 0.81 p < 0.0001). Wearing seat belts during urban, rural and main roads transit were associated (p<0.005). No associations were found between perceived risks and adoption of safety practices. Eighty three percent had never received training on occupational risks, highlighting the need of such task to increase adoption of safe management practices.

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