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1. REMEDIATION OF CHROMIUM CONTAMINATED WATERS BY D-GALACTURONIC ACID POLYMERS. PRELIMINARY STUDIES

Bellú S, González JC, Ceruti MJ, Mamana N, García S, Brambilla L, Signorella S, Sala L.

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Cr(VI) is known to be a carcinogenic and mutagenic agent; and Cr(III), although an essential micronutrient, can be oxidized to Cr(VI) by hydrogen peroxide under physiological conditions. There is increasing interest to develop new methods for the removal of total Cr from industrial waters using cheap substances able to reduce Cr(VI) to Cr(III) and to retain Cr(III) into a solid matrix. With this in mind, the ability of D-Polygalacturonic acid (D-pGal) to act as reductant of Cr(VI) and trapper of Cr(III) was evaluated. The redox reaction was found to be: D-pGal + Cr(VI) = D-pGal(oxidized) + Cr(III) + formic acid, and the reaction products were detected and quantified by HPLC, using an ion exchange column.

Spectrophotometric measurements of a reaction mixture at 350 nm, in heterogeneous phase, at pH = 1.0 and 60°C, showed that 1 g of D-pGal was able to reduce 0.18 g of potassium dichromate in 9 h. At the same time, the absorbance intensity at 570 nm did not show the expected increment, pointing out that Cr(III) remains retained into the solid matrix of D-pGal. Additional studies on D-pGal and Cr(III) mixtures at pH 1 and 12, in heterogeneous phase, showed that D-pGal was capable to bind Cr(III).

These results indicate that the D-galacturonic acid polymers are able to reduce and retain chromium into their matrix.

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2. PHASE SPACE CORRELATION ON THE ANALYSIS OF THE ERYTHROCYTES VISCOELASTICITY

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The main object to apply time series analysis, is to obtain fractal parameters that could reflect the different dynamical properties from erythrocytes belonging not only from healthy donors but also from α and β thalassemic patients. In this work the problem of characterise the cell behaviour was made on the basis of fractal approximation for ordinary (OBM) and fractionary Brownian motion (FBM), because the data set contains deterministic and stochastic components, so it could be modelled as a system of bounded correlated random walk. Here we report studies on 20 donors: 3 α /thalassemic, 7 β / thalassemic and 10 healthy donors non smokers and non alcoholic individuals. The photometrically recorded series were obtained by a process of creep and recovery over several millions of cells, using for this purpose a home made device called Erythrodeformeter. We have the time series but we have not the differential equations for the process so it could be modelled by a trayectory in phase space, using delay series we generate two different phase space with creep and recovery process to be used in the correlation. The results suggests that the time series from β thalassemic the correlation decrease with the steps, while the series from healthy donors and also from α thalassemic it is not function of the step. This information, which can not be found by a linear approach, could be used not only in analysing these nonlinear parameters, but also in evaluating clinical aspects of erythrocyte rheological properties. Further studies with larger and well defined patient populations are in process.

3. REACTIVE OXYGEN SPECIES (ROS) PRODUCTION IN HEPATOCYTES CELL CULTURE FROM SHAM AND HEPATECTOMISED RATS

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In a previous work, we demonstrated that 1, 5 and 24 hours after partial hepatectomy there is an increase of reactive oxygen species in the remnant liver. The aim of this work was to characterize ROS production in hepatocytes cell culture from sham and hepatectomised rats. Male Wistar rats were subjected to partial hepatectomy (PH) of 65% or simulated surgery (Sham, Sh) (n=5 of each group). One hour after surgery, hepatocytes were isolated by collagenase digestion. The viability of all cell suspensions used was higher than 85%. Hepatocytes were seeded in Petri dishes (1,5x10⁶ cell/dish). ROS production was determined at 1, 3 and 24 hours of culture. To quantify ROS production, the culture was loaded with DCFH-DA (5 μ M) for 30 min. at 37°C 5% of CO₂. Fluorescence formation was monitored at an excitation wavelength of 488 nm and an emission wavelength of 525 nm. The results obtained were: Sh(1h)=555,92 $\pm$ 99,97; PH(1h) =601,69 $\pm$ 65,01; Sh(3hs)= 532,34 $\pm$ 19,82; Sh(24hs)= 292,91 $\pm$ 49,88^{*}; PH (3hs)= 567,25 $\pm$ 45,58; PH (24hs)=566,54 $\pm$ 85,46^{*} *p<0,05 vs. Sh(24hs); [#]p<0,05 vs Sh(3hs). There were no differences between Sh and PH at 1 and 3 hours of culture, this could be due to the stress produced by the isolation technique. At 24 hours of culture we observed an increase of ROS in PH, suggesting that at this time of culture hepatocytes showed priming by PH. In conclusion, a 24 hours culture could be appropriate to study liver regeneration process in isolate hepatocyte.

4. STUDY OF C-ERBB-2 AMPLIFICATION IN BREAST CANCER FROM PRE-MENOPAUSAL PATIENTS

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The c-erbB-2 oncogene is amplified in up to 30% of breast carcinomas. The presentation of breast cancer at early age is usually associated with bad prognosis of the disease. The aims of the study were to investigate c-erbB-2 amplification in breast cancers from pre-menopausal women and to correlate the results with other prognostic factors, such as the histological grade (HG), the tumour size (TS), presence of estradiol (ER) and progesterone (PR) receptors and metastatic axillary nodes (MT). The samples were obtained from breast tumour tissues mounted in paraffin from pre-menopausal patients (n=46) up to 45 years-old. The amplification of c-erbB-2 was investigated by differential PCR. The results were expressed as c-erbB-2 amplification positive or negative, presence or absence of ER/PR and MT, TS <5 cm or TS $\geq$ 5 cm, HG I/II or III. The data were analyzed by means of the exact test of Fisher. The results indicated that 9% of the patients presented c-erbB-2 amplification in the tumour. Significant correlations between the TS with MT and HG, MT with ER and PR, and HG with ER were observed. The frequency of amplification of c-erbB-2 found was not higher than that one reported in patient older than 45 years in a previous study. The loss of association of c-erbB-2 amplification with other factors of bad prognostic could suggest that the amplification of the c-erbB-2 would not be a characteristic factor of the breast cancer in the group of patient up to 45 years-old studied. *This work was supported by a grant "Ramón Carrillo - Arturo Oñitvia" from the Ministry of Health of Argentina, 2003.*

5. PRELIMINARY STUDIES ON ABH ANTIGEN IN ASCARIS LUMBRICOIDES EXTRACTS

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Many blood group antigens have been found in numerous parasites. The clinical significance of this fact is not clearly established. Previous experiences have shown the same ABO and P System antigens in *Ascaris lumbricoides* extracts and in their hosts. The aim was to investigate ABH antigen in *A. lumbricoides* extracts (AE) from O Group children. We worked with 9 AE from O Group patients and 17 AE from No O Group children (A, B and AB Group). The AE were prepared by surgical remotion of the cuticle and refrigerated mechanical rupture. Agglutination Inhibition Tests (AI) were used with the AE. The tests were carried out facing the AE against two anti H monoclonal antibodies of well-known concentration and Ig class and Ulex europaeus lectine in optimal concentration. O Group red cells were used as revealing system. The AE from O Group children were facing too against anti A, anti B and anti AB monoclonal antibodies (anti A, 2.23: Ig M- < 0,7 µg/ml; anti B, 2.54: Ig G₃- < 0.5 µg/ml; anti B, 2.62: Ig M- < 0,5 µg/ml; anti AB, 2.39: Ig G₃- < 0.5 µg/ml). No AE from No O Group patients inhibited the agglutination of anti H and lectine with O red cells. One AE from O Group child inhibited the agglutination of anti H 2.72 with O red cells. The semiquantitative AI Test was made to confirm the presence of H Antigen in this AE. Two series of anti H 2.72 dilutions (1/2 to 1/1024) were prepared. Physiological solution was aggregated in each dilution of the first series and AE in the second one. Both series were revealed with O Group erythrocytes. The titre of the first series was 512 and the titre of the second one was 16. No AE from O Group children inhibited the agglutination of anti A, anti B and anti AB with isogroup erythrocytes. Conclusions: 1) AE from O Group children can present H antigen but it isn't present in AE from No O Group patients. 2) The epitope selectivity have been shown because the AE only neutralized anti H 2.72. 3) 5 titres difference between the two series have determined a significative amount of H antigen in the AE. 4) A and B epitopes absence was corroborated in AE from O Group children.

6. EVALUATION OF SOME QUALITY PARAMETERS IN PARACETAMOL TABLETS OF THE LOCAL MARKET

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Among the pharmaceutical dosage forms, tablets are those characterized by being the most widely used due to their practicality, and because they can be easily manipulated, thus assuring accurate and precise dosification. In a generics-based health system it is important to assure the interchange suitability of products having the same dose of a given drug. Aiming to verify the exchangeability of paracetamol (acetaminophen) tablets (500 mg), some of their quality parameters such as friability, disintegration, dissolution, uniformity of dose units and assay were assessed in 10 brands of locally marketed paracetamol tablets, according to the USP 24. It was observed that all but one of the tested brands complied with the assays, being the most remarkable non-compliance observed a failure in the friability test, leading to broken tablets; in addition, when dissolution profiles were constructed, it was observed that in spite that the different brands complied with the dissolution assay, the profiles were not the same. In conclusion, despite a certain degree of equivalence among the tested brands, there are minor variations which, without leading to non-compliance, demonstrate slight differences among the brands.

7. MATHEMATICAL MODEL OF THE BEHAVIOR OF A BACTERIAL STRAIN EXPOSED TO VANCOMYCIN

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Many scientific studies of kinetic of bacterial death emphasize the importance of mathematical models to the aims of giving rigor to research on bactericidal effect of antimicrobial agents. *Staphylococcus spp.* with heterogeneous resistance to vancomycin (VAN) exhibit a peculiar behaviour when they are exposed to the antibiotic, while susceptible sub-population of cells decreases due to bactericidal effect of VAN, the small fraction with intermediate resistance is not affected, resulting in a clear cell-proliferation within hours. The curves obtained by plotting the decimal logarithm of CFU per millilitre against time do not respond to any of the existing mathematical models. The present study intends a model that describes the curves obtained when a heterogeneously VAN resistant strain is exposed to different VAN concentrations. The strain is a coagulase-negative staphylococci isolated from blood of a newborn infant. The proposed model consists of a sum of two logistic functions, which bear witness to a nonlinear dependence of log(CFU/ml) with time. The unknown parameters, k₁, k₂, k₃ and k₄, were estimated by nonlinear regression. The ordinary least-squares criterion was used to fit the model to the data. k₂ and k₄ are tie with the rate of death and growth respectively, whereas k₁ and k₃ are reciprocally related to the size of the resistant sub-population. The model allows an accurate description of the underlying dependency of parameters and the concentration of VAN in the growth medium.

8. STUDY OF GLYCOPEPTIDE RESISTANCE IN COAGULASE-NEGATIVE STAPHYLOCOCCI (CNS) ISOLATED FROM NEWBORN INFANTS

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Infections due to CNS are common among very low birth weight infants and they have become a serious therapeutic problem when they express methicillin resistance. Glycopeptides are drugs of choice in that cases. In order to determine whether vancomycin (VAN) resistance is emerging among clinical strains we have examined 106 CNS isolated from newborn infants. All of them were identified up to the species level by conventional methods. Methicillin resistance was evaluated by detecting the *mecA* gene using PCR amplification. They also were tested for susceptibility to VAN and other antibiotics by the reference NCCLS disk diffusion method. The VAN MICs were determined by agar dilution. We also performed several screening tests to determine reduced susceptibility to VAN. Although we didn't find any VAN resistant strain, we found 4 strains heterogeneously VAN resistant, which were confirmed by the Population Analysis Profile method. These strains contained sub-populations of cells that were able to grow in VAN concentrations as high as 8 µg/ml. These four isolates were negative when tested for the enterococcal *van* genes by PCR. Thin-sections examined by transmission electron microscopy showed a thicker cell wall in the resistant subclone than did the parent strain. These heterogeneously VAN resistant strains are a preliminary stage that allows development into resistant strains upon exposure of the organism to constant level of VAN. For that reason, surveillance of glycopeptide resistance emergence is crucial for patients in care areas under VAN usage.

9.

MUTAGENIC ASSAYS OF SULFATHIAZOLE IN THE PRESENCE OF NITRITES USING THE AMES TEST

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The Ames test is an economic and effective bacterial mutagenicity assay, that uses *Salmonella typhimurium* as indicator bacteria. The *S. typhimurium* strains employed can not synthesize histidine, so they need this aminoacid in order to grow. A mutagenic substance can overflow this lack, making colonies grow (revertant colonies). According to Ames, a compound should be considered mutagenic if the reversion coefficient, R.C., exceeds 2.0. (R.C. = revertant number per tested plate/revertant number per control plate - spontaneous-). Besides of primary drugs, it is necessary to consider interactions that might occur into the organism, like the nitrosation of substances with an -NH₂ group by the nitrite added into food products (cured meats, fried bacon, etc). Mutagenicity of sodium sulfathiazole, NaST (antibiotic with an -NH₂ group in its molecule) in the presence and absence of sodium nitrite in acidic medium was evaluated in the Ames test, employing *S. typhimurium* TA 98 y TA 100 strains. Under these conditions, TA 98 strain showed a strong mutagenic activity (R.C. = 12.38) for the mixture of NaST (250 µg/plate) + NaNO₂, at pH ≈ 0, where it is formed a diazo compound. The diazo compound was also obtained at pH similar to the gastric juice one, so, it would be convenient, as a caution, to not eat food with nitrites added in concomitance with sulfadugs.

10.

PRELIMINARY STUDIES ABOUT THE INTERACTION OF MERCURY WITH PHTHALYLSULFATHIAZOLE

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Coordination chemistry of ligands with metal ions has a lot of applications. As part of a program devoted to the study of the interaction of Hg with sulfonamides, we began Hg-phthalylsulfathiazole studies. An aqueous solution of the complex was obtained, by mixing phthalylsulfathiazole, FST and Hg(II) in molar ratio = 1. No precipitate was obtained when NaOH was added to this solution, suggesting the absence of free Hg(II) in the solution. By evaporation of this solution, a yellow solid was obtained, which was insoluble in water. This solid was analyzed as follows: *Electronic spectra*: (Jasco V-530, 25 °C, in methanol and DMSO). Peaks observed for the ligand, FST, and the complex, Hg(II)-FST, are summarized in the following table. (s: shoulder)

solvent	FST	Hg(II)-FST	assignment
methanol	208	207	-COOH
	291 (s:263)	290	-CO-NH-
DMSO	292 (s:268)	296 (s:271)	-CO-NH-

The observation of the peak at 208 nm, due to a transition π/π^* of the carboxyl group suggests the non participation of this group in the coordination. The disappearance of the shoulder at 263 nm in methanol (and shift in DMSO) suggests the interaction with the N attached to the benzene ring. * ¹H RMN spectra (in DMSO, Bruker AC-200 E, at 22°C): all signals became broad and shifted to lower field, except: H_M (thiazole ring) and H_C (sulfonamide). The results suggest that the coordination of Hg(II) with FST might involve both N_{thiazolic} atom and the N atom from the -CO-NH- group.

11.

COMPLEXES OF Co(II) AND Co(III) WITH SULFANIL-AMIDE: THERMOGRAVIMETRIC AND SPECTROSCOPIC STUDIES

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The synthesis of metal sulfonamide complexes is of a great interest because some of the ligand properties are enhanced. As part of a program devoted to the study of the interaction of metal ions with sulfa drugs, we obtained complexes of sulfanilamide, SA, and cobalt, Co-SA, by adding an aqueous solution of CoCl₂ to the aqueous solution of SA, in mole ratio [SA]/[Co(II)] = 2. Co(III)-SA complex was obtained similarly, plus H₂O₂. Elemental analyses let us propose the following minimum formulas: [Co(SA)₂(H₂O)₄] for Co(II)-SA (green solid) and [Co(SA)(OH)₂(H₂O)₃] for Co(III)-SA (brown solid). *Vibrational spectra (IR)*: the bands due to $\nu_{\text{asym}} \text{NH}_2$, and $\nu\text{-SO}_2\text{NH}$, are shifted to lower frequencies in the IR spectrum of Co(III)-SA. $\nu \text{RSO}_2\text{S-N}$ are shifted the lower frequencies in the IR spectra of both complexes. *Electronic spectra*: (diffuse reflectance): Co(II)-SA *d-d* spectrum shows two transitions (669 and 622 (sh) nm). Co(III)-SA *d-d* spectrum shows superposition with bands of the ligand. In both cases the spectra suggests an octahedral geometry around the metal ion. The thermal decomposition of Co(II)-SA showed two strong DTA signals (334-361 and 530°C), giving volatile products with loss of 82.3% of sample weight, in agreement with the proposed formula. Actually we might conclude that SA gives complexes with Co(II) and Co(III) of different metal-ligand stoichiometry, but both with an octahedral geometry around the metal. The coordination of Co(II) with SA could include the N_{sulfonamidic} atom, while the coordination of Co(III) might include both N atoms of SA.

12.

ASSESSMENT OF NADP(H) DEPENDENT OXIDO-REDUCTASES OF SOXRS REGULON IN ESCHERICHIA COLI

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The *soxRS* regulon modulates a global response to oxidative stress imposed by superoxide (O_2^-) and nitric oxide. Among the identified members of the *soxRS* response are two NADP(H) dependent oxidoreductases, Glucose 6-phosphate dehydrogenase (G6PD) and Ferredoxin-NADP(H) reductase (FPR), encoded by the *zwf* and *fpr* genes, respectively. G6PD provides NADPH for reductive biosynthesis. FPR mediates the electron transfer from NADPH to ferredoxin or flavodoxin for several pathways. Mutant bacteria in *zwf* and *fpr* are susceptible to oxidative stress. In this context G6PD maintains a high ratio of NADPH/NADP⁺, under oxidative conditions that deplete NADPH. The antioxidant role played by FPR is less well understood. To investigate the relative contributions of G6PD and FPR as member of *soxRS* regulon we characterised the time course of their induction during oxidative stress and the overproduction of G6PD in *E. coli*. FPR increase is observed later and surpassed the induction relative to the one of G6PD. On the other hand, using a *soxS' lacZ* gene reporter we analysed the state of activation of the *soxS* gene in a G6PD overproducing strain. These observations indicate that lower activation of *soxS* exists, in agreement with a greater fraction of NADP⁺ in the reduced state. The results confirm the proposed function of G6PD, namely, contribution of the NADPH/NADP⁺ pairs to *soxRS* activation and support the FPR role in controlling NADPH accumulation.

13. EXPRESSION OF ALTERNATIVE OXIDASE IN HOST AND NON-HOST PLANT INTERACTION WITH *Xanthomonas axonopodis* pv. *citri*

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In plant mitochondria the electron transport resistant to cyanide or alternative pathway depends on a unique enzyme named alternative oxidase (AOX). It has been reported the induction of AOX expression in incompatible plant-pathogen interactions. In this work we study the participation of AOX from host and non-host plants in the interaction with *Xanthomonas axonopodis* pv. *citri* (*Xac*). We used as host plant orange and as non-host tobacco. Previously two type of AOX from orange were identified belonging to the families AOX 1 y AOX 2a. We analyzed the expression of AOX by RT-PCR. In the compatible orange-*Xac* interaction no induction in the expression levels of AOX were observed at the different times assayed, being approximately 3×10^7 RNA molecules per 100 mg of tissue. On the other hand, in the incompatible interaction tobacco-*Xac* we observed an increase of AOX expression 6 h post-infiltration with a maximum at 24 h post-infiltration of two times relative to the control. These results show that during the compatible interaction no changes in the levels of AOX expression are produced indicating that no increase of the alternative respiratory metabolism is taking place, while in the interaction with the non-host plant the alternative pathway is induced as it was observed in the response of resistant host plants. In this context we suggest that the incompatible host and non-host response may be sharing similar defense mechanisms.

14. ORAL DOSAGE FORMS FOR DICLOFENAC SODIUM SOLID DISPERSIONS. BIOPHARMACEUTICAL EVALUATION

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The solid dispersions were proposed as a novel method to enhance the dissolution and absorption of poorly water soluble drugs. These compounds were prepared by using an inert hydrophilic carrier at the solid-state by different technological methods, converting a fluid drug carrier combination to the solid state. Diclofenac sodium was selected as a model drug, a non-steroidal anti-inflammatory phenylacetic acid derivative. The purpose of this study was to develop oral formulations from solid dispersions, using high molecular weight polyethylene glycols, polyvinylpyrrolidone, semi-synthetic celluloses derivative and carbopol. The biopolymers were chosen because of their solubility and physiological tolerance. The study was based in the preparation of diclofenac sodium solid dispersions by kneading, solvent evaporation and physical mixture containing different drug:polymer ratios. The *in vitro* release profiles showed a noticeable enhancement dissolution rates, up to 30%, compared to the pure drug. The solid dispersions obtained demonstrated the advantages of the proposed methodology to increase the solubility of poorly water-soluble drugs with concomitant implications for improving the bio-availability. These delivery systems appear particularly advisable for oral administration of drugs that have marked gastrolesive effect and are used in treatment of chronic diseases.

15. TAXONOMY OF *Fusarium*-SOYBEAN SUDDEN-DEATH SYNDROME (SDS) CAUSING STRAINS IN ARGENTINA

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It was demonstrated that *Fusarium solani* f. sp. *glycines*, reported to be the causal agent of SDS, represented 4 different species in the *F. solani* species complex. The objectives of the paper were to characterize morphologically, molecularly and chemotaxonomically different *Fusarium*-SDS causing isolates. Eight *Fusarium* isolates that were obtained from taproots of soybean plants exhibiting typical SDS symptoms, two *Fusarium*-only root rot causing isolates (*F. solani* and *F. semitectum*), and reference strains were used. Monosporic cultures were subjected to macro and micromorphology studies. Molecular analyses were conducted by PCR utilizing as single primers (GACA)₄ and M13. The production of trichotecenes groups A and B and zearalenone was evaluated. Morphologically 3 SDS-causing isolates were identified as *F. virguliforme* and 5 isolates as *F. tucumaniae*. Both primers allowed to detect differences between *Fusarium*-SDS causing isolates and non-SDS causing isolates. M13 allowed to detect different profiles between *F. virguliforme* and *F. tucumaniae* isolates. PCR-fingerprinting has proven to be useful for identifying these strains. None of the *Fusarium* strains produced trichotecenes and zearalenone under these conditions.

16. EVALUATION OF BIOCONTROL AGENTS AGAINST SOYBEAN SUDDEN-DEATH SYNDROME *Fusarium* CAUSING ISOLATES

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Sudden-death syndrome (SDS) represents one of the main soybean diseases causing important economic loss. Two species included in the *Fusarium solani* complex, *Fusarium tucumaniae* and *Fusarium virguliforme*, are responsible for SDS in our country. Seed treatment with a biological agent would be a promissory alternative for the development of a competitive and sustainable agriculture. The objectives of the present work were a) to study the pathogenicity of five isolates of *F. tucumaniae* and one isolate of *F. virguliforme* and b) to evaluate the antagonistic activity "in vitro" of *Streptomyces* strains against these *Fusarium* isolates. The pathogenicity was evaluated under greenhouse conditions by artificial inoculations using the toothpick method, on four soybean cultivars: Ripley, Spencer, Pioneer 9492 RR and A 6445RG. Activity of nine *Streptomyces* strains was evaluated by a bioassay, registering inhibition zones. Results were statistically analyzed using an ANOVA (for a balanced factorial design) and the Tukey's multiple range test. All of the *Fusarium* isolates produced typical SDS foliar symptoms. Ripley showed the lowest foliar severity and there were not significant differences among the *Fusarium* isolates. The inhibitory effect produced by C202 against all *Fusarium* isolates "in vitro" was significantly higher than the other *Streptomyces* strains. Potential use of these microorganisms as biocontrol agents will be evaluated under greenhouse conditions.

17. ADHERENCE OF *Candida dubliniensis* AND OTHER *Candida* STRAINS TO BUCCAL EPITHELIAL CELLS

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Candida dubliniensis (Cd) is a new strain, phylogenetically related with *Candida albicans* (Ca), frequently isolated from HIV infected patients. Our aim was to compare the adherence capacity, as a potential virulence factor, among 14 *C. dubliniensis*, 3 *C. albicans*, 1 *Candida glabrata* (Cg) and 2 *Candida krusei* (Ck), isolated from the oral cavities of HIV infected patients. Each of them were grown in Sabouraud's broth during 24 hs at 37°C, washed twice with PBS and taken to a final concentration of 1×10^7 yeasts/ml. A pool of buccal epithelial cells was used. Adherence assays were performed according to Kimura *et al* technique and the number of yeasts attach to 100 BEC were counted. The highest adherence values were found for two Cd strains (Cd 89 = $510,7 \pm 126,4$ yeasts/ 100 BEC and Cd 109 = $488,8 \pm 6,4$ yeasts/ 100 BEC) and one Ca strain (Ca 73 = $504,8 \pm 12,0$ yeasts/ 100 BEC). The lowest adherence values were observed for the two Ck strains (Ck 106 = $24,7 \pm 17,5$ yeasts/ 100 BEC and Ck 89 = $28 \pm 19,0$ yeasts/ 100 BEC) and for the Cg strain (Cg 751 = $74 \pm 29,3$ yeasts/ 100 BEC). For other Cd and Ca strains, the adherence values varied between 144 y 350,3 yeasts/100 BEC. Differences in adherence between these groups were statistically significant. Nevertheless the *in vitro* adherence of Cd to BEC was similar to that of Ca, the frequency of oral carriage in HIV infected patients observed for Ca is greater than that observed for Cd. Low adherence values were found for Ck and Cg, in concordance with the low carriage rate of these strains in oral cavities of HIV (+) patients, according to that observed in previous studies.

18. ORAL COLONIZATION BY YEASTS IN HIV POSITIVE PATIENTS

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The objective of the present work was to determine the frequency of oral colonization by *Candida* species in HIV positive patients and to compare it with a population of HIV negative individuals. Samples were obtained by swabbing the oral mucosa of 108 HIV (-) individuals and 101 HIV (+) patients, without symptoms of oral candidiasis. Direct exams and mycological cultures were carried out and the yeasts obtained were identified by the following tests: colony color on CHRO Magar *Candida* media, morphology on corn meal agar with Tween 80, carbohydrates assimilation profiles using the API 20 C Aux kit, and morphology on sunflower seed agar. Seventy-three HIV (+) patients (72.3%) were colonized by yeasts on their oral mucosa (14 of them carrying more than one species) while only 43 (39.8%) of the HIV (-) individuals exhibited this trait (3 of them presented more than one species). The frequency of appearance of the different yeasts species was as follows: 60.7% *Candida albicans*, 20.2% *Candida dubliniensis*, 5.6% *Candida glabrata*, 5.6% *Candida krusei*, 4.5% *Candida tropicalis*, 3.3% other species for the HIV (+) group; and 73.9% *C. albicans*, 15.5% *C. tropicalis*, 2.1% *C. glabrata*, 2.1% *C. dubliniensis* and 4.2% other species for the HIV (-) population. *C. krusei*, which has intrinsic resistance to Fluconazol, was obtained only in HIV (+) patients and the prevalence of *C. dubliniensis* among this group was higher compared to that informed in other countries. The relationship of *C. dubliniensis* with HIV (+) patients and the mechanisms of transmission must be further studied.

19. ESSENTIAL OIL FROM *TARGETES MINUTA* FOR *VARROA JACOBSONI* CONTROL: LABORATORY TESTS

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Varroa Jacobsoni, an ectoparasitic acarid, is one of the serious pests of honeybees. The purpose of this study was to evaluate varroacidal potential of essential oils extracted from *Tagetes minuta* L, an aromatic vegetable species commonly used in apiculture, which is autochthonous from the Province of Chaco (Argentina). Oils were obtained from dried leaves and flowers (independently) by steam-distillation, then refrigerated and preserved anhydrous in brown flasks. Oil compositions were determined by gas chromatography, thus, the main compounds in oil from flowers were β -ocimene (35-55%) and tagetenone (30-40%), while in oil from non-blossomed plant leaves it was dihidrotagetenone (40-55%). Live acarid were obtained directly from chamber squares of breed and immediately taken to the lab. The bio-tests were carried out in conditioned rooms. Each test consisted in determining the survival rate of 10 mites after 3 and 6 hours. The mites were contained in a Petri capsule and 4 μ l of oil were placed on a 3mm diameter wooden cylinder located in the center of the capsule. Mite survival after 3 hours was 4% for those exposed to oil from flowers, 60% for the oil from blossomed plant leaves and 72% for the oil from non-blossomed plant leaves. After 6 hours, all exposed mites were dead, while 76% of not exposed ones were still alive.

20. GENOTYPIC IDENTIFICATION OF FUNGI ISOLATES OF ORAL MUCOSA

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The oral cavity is one of the anatomic parts which is subject to mycotic infections, especially, Candidiasis. It is necessary to make a correct clinical and mycological diagnosis in order to establish proper therapeutic patterns. The aim of work was to put into practice a reliable technique based on molecular biology for a proper taxonomic typification of different *Candida* species. Were studied 64 patients with clinical symptoms of oral Candidiasis. The yeasts were isolated and identified for traditional taxonomic procedures. The isolates were kept as stock cultures in Agar Sabouraud-Dextrose and in distillate water at room temperature. A PCR fingerprinting was carried out using the M13 core sequence, for the molecular identification: 44 positive isolates were obtained, 61% corresponded to *C. albicans*, 23% to *C. parapsilopsis*, 7% to *C. tropicalis*, 4,5% to *C. krusei* and 4,5% to *C. glabrata*. The identification by means of PCR fingerprinting was faster than any of the traditional methods which require at least 7 days apart from the time other possibly required tests would take. PCR fingerprinting requires 3 days, no additional tests are necessary and provides definite and faster identification results, which in many cases it is essential to start a proper antifungal therapy and avoid resistance to certain treatments.

21. EVALUATION OF FUNGUS FLORA IN GYNECO-LOGICAL PATIENTS

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Vulvovaginal candidiasis is the second most common type of vaginal infection, at least 75% of adult women are estimated to have an episode in their lives; 40%-50% of these women can even have episodes twice or more and a 5% present recurrent vulvovaginal candidiasis (VVC). This mycosis is associated with factors such as stress, diabetes, autoimmune diseases, antibiotics, corticosteroids, pregnancy, oral and intrauterine contraceptives, etc. The main etiologic agent is *C. albicans*. However, other *Candida* species can also cause the disease. The aims of this work were: to identify the yeast species isolated from the patients, to determine associated risk factors and to correlate the colonization with some signs and symptoms. Were studied 110 patients who came to the Gynecology Service at the Centenario Provincial Hospital. Vaginal specimen was collected and classified using fast methods such as corn meal agar, CHROMagar *Candida* media and the API 20 C Aux kit. Were positive for *Candida* 30 samples, the frequency of the isolation were: *C. albicans* (66,7%), *C. glabrata* (6,7%), *C. parapsilopsis* (16,7%) and 3,3% to each of the following species: *C. famata*, *C. lusitanae* and *C. tropicalis*. The presence of leucorrhea and pruritus showed a statistically significant association ($p=0,0000$) in those patients with *C. albicans*. The Odds Ratio for leucorrhea was [C.I. 95% (3,66-25,07)] and the Odds Ratio for pruritus was 10,8 [C.I. 95% (3,71-31,5)]. Besides, 44,4% of the pregnant women and 33,3% of the patients who used intrauterine device showed colonization.

22. IDENTIFICATION OF MALASSEZIA SPECIES ISOLATED OF PATIENTS WITH PITYRIASIS VERSICOLOR IN ROSARIO CITY

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Pityriasis versicolor (PV) is a fungal disease that causes alterations in skin pigmentation. This pathology has been extensively studied, its etiology and pathogenesis are not very well defined under the new system of classification of *Malassezia*'s genus. The aim of this work was to identify phenotypic and genotypically species of *Malassezia* isolated of patients with PV.

The study examined 129 samples of patients. Scales were taken by scraping, microscopically examined and were inoculated on modified Dixon agar (mDA) and Sabouraud-glucosa agar, with and without the addition of olive oil and incubated at 32°C for 10 days. The phenotypic study were realized by traditional probes.

A PCR fingerprinting was carried out using the primer M13, for the molecular identification.

With regard to direct microscopy, 91 were microscopically positive for *Malassezia* elements (yeasts or filaments alone or both structures together). Of all samples, 58 yielded positive cultures. Ten strains (17,2%) no survive to subculture, for this reason were classified as *Malassezia* spp in base of their macro and microscopic characteristics. Based on the identification of the 48 viable strains, we found that the species isolated were: *M. sympodialis* (56,9%), *M. furfur* (13,8%), *M. slooffiae* (10,4%) y *M. obtusa* (1,7%). Our results show that the most common species isolated was *M. sympodialis*. With respect to the methods of identification, there was a correlation of 100% between phenotypic and genotypic ones.

23. STUDY OF MEMBRANE PROTEINS IN SENESCENT RED BLOOD CELLS

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Because mature red blood cells (RBC) are unable to synthesize new proteins, the event that trigger removal of senescent cells from circulation might derive from alterations in pre-existing proteins that lead to recognisable changes at the membrane surface. The aim of this work was to study membrane proteins modifications in senescent (Se) RBC and young (Y) RBC.

Blood samples were drawn by venipuncture from 10 normal volunteer donors. SeRBC and YRBC were obtained by differential centrifugation. Ghost were prepared by osmotic lysis. Proteins obtained were dissolved in SDS and quantified by optical density at 230nm. ABH, MN and D antigens reactivity was assessed by hemagglutination. Erythrocytes membrane protein values in SeRBC (2.46 ± 0.53 mg/mL) were significantly lower ($p < 0.01$) than those obtained with YRBC (2.84 ± 0.50 mg/mL). A decreased ABH and MN antigenic expression ($p < 0.001$) and an increased D antigenic expression ($p < 0.001$) was observed in the population of SeRBC. The decreased values of membrane protein obtained with SeRBC might be attributed to the protein break down that occur during aging. Alterations in the Band 3 protein would be responsible for the altered expression of the RBC antigens study.

24. RHD HOMOZIGOSITY INDEX IN THE Rh PHENOTYPES

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The *RHD* and *RHCE* genes are tandemly arranged and their alleles segregate as haplotypes with frequencies varying in different ethnic groups. Determination of *RHD* zygosity is commonly inferred from serological testing. *RHD* is flanked by the homologous 5' and 3' Rhesus boxes. The *RHD* deletion, found in most RhD negative Caucasians, was due to recombination of the 5' and 3' Rhesus boxes resulting in the formation of a hybrid Rhesus box. The aim of this work was to determine the *RHD* homozygosity index in the different RhD phenotypes.

We studied 198 blood samples of unrelated individuals. The Rh phenotype was performed by hemagglutination. The *RHD* deletion was determined by PCR-ARMS with primers that selectively amplify a 1981 bp segment of the hybrid Rhesus box that is present in RhD negative and RhD positive heterozygous samples. We phenotyped 166 (83,8%) RhD positive samples, 68 (41,0%) were *RHD* homozygous and 98 (59,0%) were *RHD* heterozygous according to PCR. These results did not differ from those published in the literature based on the most probable genotype method. However, the homozygosity indexes were significantly higher in the DccEe (19,0% vs 6,6%) and Dccee (17,0% vs 3,3%) phenotypes. These results show an increase frequency of Dce haplotype in the DccEe and Dccee phenotypes. The homozygosity index of this genetic marker is similar to that found in Africans. These findings would contribute to estimate the genetic admixture of the population under study.

25. IMMUNOHISTOCHEMICAL STUDY OF DOPAMINE- β -HYDROXYLASE IN THE LOCUS COERULEUS OF NEONATE RATS EXPOSED TO 2,4-DICHLOROPHENOXACETIC ACID THROUGH MOTHER'S MILK

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Dopaminergic system alteration in rats exposed to 2,4-dichlorophenoxyacetic acid (2,4-D) through lactancy has been reported in previous histologic and biochemical studies. Changes in noradrenaline (NA), dopamine (DA), their metabolites and some metallic ions levels have also been observed in different brain areas (Cpf: NA +20%, DA -37%, Fe +50%, Zn +43%, Cu +25%). The aim of this work was to perform an immunohistochemical study of dopamine- β -hydroxylase (D β H), the enzyme that synthesizes NA from DA, in the locus coeruleus (LC) of neonate rats exposed to 2,4-D through lactancy. Rat litters from dams injected (i.p.) with 70 or 100 mg/kg/day of 2,4-D from post natal day 9th to 25th were used as treated groups 1 and 2 and litters from dams injected with saline solution were used as control group. 25-day-old pups were anesthetized and fixed by transcardiac perfusion. Control and treated serial sections from brain stem, that correspond to LC according to Paxinos and Watson atlas, were simultaneously processed with Sternberger (1970) PAP immunohistochemistry method for a-D β H. Using an image analysis system, the immunostaining optical density (OD) was measured as an estimation of the enzyme content. An OD significative decrease in the LC of 2,4-D exposed animals was observed.

26. BEHAVIORAL AND CENTRAL NERVOUS SYSTEM MYELIN STUDIES IN WISTAR RAT PUPS EXPOSED TO VANADIUM (V) THROUGH LACTANCY

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Neurobehavioral and myelin alterations on adult rats exposed to vanadium (V) were previously reported. In this work, Wistar rat pups were exposed to vanadium through lactancy. Dams from treated litters were intraperitoneally injected with 3mg NaVO₃/kg body weight/day during 11 days from the 10th.post natal day (10 PND). Dams from control litters were injected with same volume of saline solution. Each 48 hours, from 8th to 20th PND, the following behavioural tests were performed to neonates: surface righting reflex, negative geotaxy and hindlimb support. Open field test was performed the 21st PND. At 22nd PND, animals were perfusion-fixed through abdominal aorta, brains and cerebellums were removed and cut with a cryostat. Brain sections from control and treated animals were processed for the following histologic studies: Schmued myelin histochemistry and PAP immunohistochemistry for a-myelin basic protein. In the open field test, a significative decrease in the number of crosses (p<0.01) and rearings (p<0.04) in exposed pups were observed. Forelimb support latency was also decreased in exposed pups (p<0.008). A decreased myelin staining in corpus callosum and cerebellum in these pups was also observed. Results suggest that vanadium exposure through lactancy would induce behavioral and histological changes in rat developing CNS.

27. FACTORS AFFECTING THE CATALYTIC ACTIVITY OF Mn-CATALASASE MIMICS

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Manganese catalases (MnCAT) possess a diMn active site and disproportionate H₂O₂ by cycling between the Mn^{II}₂ and Mn^{III}₂ oxidation states. The crystal structure of MnCaT from *L. plantarum* and *T. thermophilus* reveal that the two Mn ions are triply bridged through a carboxylate from a glutamate residue and two solvent-derived single atom bridges. A wide variety of diMn complexes have been synthesized to mimic structural features of MnCAT, but they are 10³-10⁴ times slower than the enzyme. The obtention of more effective mimics of MnCAT requires a better understanding of the factors controlling their catalytic activity. With this in mind, we have evaluated the catalase-like activity of a family of Mn complexes with salpentOH (1,5-bis(salicylidenamino)pentan-3-ol) and its phenyl-ring substituted derivatives: [Mn₂(μ -OAc)(μ -OMe)X-salpentO (MeOH)₂]Br. These complexes are good structural mimics of the Mn^{III}₂ form of MnCAT (with Mn $\cdots$ Mn separation only 0.1-0.2 Å shorter than found for the enzyme), disproportionate more than 1000 eq. of H₂O₂ with saturation kinetics, and show substrate affinity comparable to that of the biosite. Unlike other complexes with similar geometry, atomic environment and redox potentials, but longer intermetallic separation, which cycle between the Mn^{II}₂/Mn^{III}₂ levels, complexes of the X-salpentOH family cycle between Mn^{III}₂/Mn^{IV}₂. Thus, the Mn $\cdots$ Mn distance is an important factor in these reactions: the short Mn $\cdots$ Mn separation (~2.93 Å) imposed by the ligand in the X-salpentOH models seems to be best suited for accommodating the higher oxidation states of Mn.

28. STUDY OF ERYTHROCYTE ADHESION MEDIATED BY AGGLUTININS USING THE MICROSCOPIC DIGITAL IMAGE PROCESSING

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Different substances as lectins and antibodies are called agglutinins because cause cells to clump (agglutinate) if they possess the proper antigen. They are a number of immuno-haematologic techniques that evaluate if the cells form visible clumps for to discern if the agglutinins are associated with the correspondent antigen on erythrocyte. In the present work, the agglutination reactions are registered in real time for different concentrations of monoclonal anti-A and autochthones vegetal lectins with a microscope (100x) connected to a CCD. The analysis of the digital images permits calculate the agglutination rate and measures the agglutinate dimensions in real time for to quantify the agglutination degree. The present technique is different from conventional blood tests because it is a live sample, where the agglutination is evaluated in real time, that permit obtain important information about the agglutinin, which would be very useful in the Immunohaematology Laboratory for the evaluation of the lectin qualities and the quality control of the antibodies. For other hands, this tests permits to obtain information about the antigen density on erythrocyte membrane and to study the kinetic of cell adhesion during the agglutination process.

29.

EFFECT OF THE CONSUMPTION OF TOBACCO ON THE SEMINAL QUALITY IN INFERTILE PATIENTS

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The adverse effect of consumption of tobacco over health is well known. Our objective was to know the effect that smoking produces on the seminal quality. A prospective study on a population of 172 men that consulted for infertility was carried out. Patients with some pathology able to alter the seminal quality were excluded. A sperm study and functional tests were carried out according to WHO. The patients were divided in the three groups: G1 (n=84): non smokers, G2 (n=48): smokers of 1 to 20 cigarettes/day, G3 (n=40): smokers more of 20 cigarettes/day. In the present study was researched: concentration of spermatozoa and progressive motility using an optical microscope, the sperm morphology with Papanicolaou, the functional state of the sperm membrane was evaluated with the Hypoosmotic Test, the DNA integrity was research using acridine orange as fluorocrom, condensation of nuclear chromatin with aniline blue and the oxidative stress with the Modified Sperm Test. The statistical analysis was carried out with the t-Student test and a significant difference between non smokers (G1) and smokers of more than 20 cigarettes/day (G3) was found for all the researched variables ($p < 0.01$). Significant difference in integrity DNA and sperm morphology were observed between the groups of non smokers (G1) and the smokers of under 20 cigarettes/day. Significant statistically difference was found only between the smokers (G2 vs G3) to evaluated the integrity DNA. Smoking cigarettes alters the seminal variables, being the most affected the sperm morphology and nuclear DNA. The smoker men present a marked decrease in the seminal quality that affects the reproductive capacity.

30.

BODY MASS INDEX AND ITS RELATIONSHIP WITH LIPIDIC PROFILE VALUES AND TENSIONAL FIGURES IN PEDIATRIC AGE

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Obesity in pediatric age leads to alterations of the lipidic profile together with other metabolic disturbances which condition high tensional figures. A body mass index above the 95 percentile ($BMI \geq P95$) is defined as obesity. Our study consisted in determining if there is a relationship between the lipidic profile, arterial pressure and BMI in our population with regards to sex and age group. A school population of 924 children and adolescents of both sexes, aged from 5 to 18 years was studied. Theirs systolic arterial pressure (SAP), diastolic arterial pressure (DAP), weight in kilograms and height in meters with standard techniques were measured, calculating the BMI (kg/m^2). The lipidic profile was carried out in blood samples obtained from venipuncture after a 12- hours fasting. Total Cholesterol (TC), Triglyceride (TG) and HDL-cholesterol (HDL-C) were measured by enzymatic methods. The LDL-cholesterol (LDL-C) was calculated with the Friedewald equation. The cut off values test were taken: values $\geq P95$ for TC, values $\geq P75$ for TG, LDL-C and values $\leq P25$ for the HDL-C. A SAP and/or DAP $\geq P95$ for age and sex was defined as hypertensive. The obtained results are:

Sex	GIRLS				BOYS			
Age	5-11		12-18		5-11		12-18	
	Nor	Obes	Nor	Obes	Nor	Obes	Nor	Obes
SAP	8 %	8 %	1 %	18 %	6 %	8,3 %	3,5 %	8,3 %
DAP	3,5 %	8 %	3 %	18 %	7 %	17 %	8,8 %	8,3 %
CT	11 %	23 %	8 %	52 %	3 %	17 %	7,2 %	8,3 %
TG	32 %	15 %	16 %	45 %	18 %	25 %	17 %	25 %
LDLC	24 %	46 %	24 %	42 %	25 %	50 %	16 %	8,3 %
HDLC	17 %	15 %	17 %	45 %	19 %	17 %	20 %	50 %

The statistical analysis wasn't deepened because of scarcenumber of obese people. According to our results in the obese population: * a modified lipidic profile is observed * significant differences between SAP and DAP with the normal population are not observed, except in women from 12 to 18 years old. The detection of these risk factors in the early age enhances the importance of promoting healthier habits to decrease the CVD incidence.

31.

RELATIONSHIP BETWEEN THE CONCENTRATION OF PLASMATIC CALCIUM AND ARTERIAL PRESSURE IN A PEDIATRIC POPULATION

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Epidemic and clinic studies suggest an association between the arterial pressure (AP) and alterations in the levels of calcium (Ca). Transverse evaluations provide evidence of association between hypertension and blood calcium (Ca_s) establishing that supplementation with Ca in normotensive and hypertensive patients would decrease the levels of (AP), with greater effect in patients with previous deficient ingestion, although other studies do not show association and are considered to be the effects of calcium very weak. The relationship between Ca and AP levels has been the focus of many studies in adults and in pediatric populations. The aim of this work was to study the relationship between the concentration of Ca_s and the levels of systolic arterial pressure (SAP) and diastolic arterial pressure (DAP) in a pediatric population. 240 healthy free-diet children (115 women and 125 men) aged from 5 to 14 years were studied. The SAP (1° RK) and DAP (4° or 5° RK) were measured with an anaeroide tensiometer (TYCOS) according to the WHO protocol and a blood sample was taken before breakfast for the determination of Ca_s by the colourimetric method (Wiener Lab). The values of SAP, DAP and Ca_s in the studied population were within the reference ranks for sex and age. The statistical study of the whole population and populations according to sex showed that there is no significant relationship between SAP and Ca_s nor between DAP and Ca_s . The correlation coefficients that were found were the following:

Women: $Ca_s / SAP = 0,078$ $Ca_s / DAP = 0,041$

Men: $Ca_s / SAP = -0,007$ $Ca_s / DAP = -0,042$

Although nutritional advice promotes the increase in calcium ingestion as a preventive measure to avoid AP in adults, there is no association observed between Ca_s and AP in the studied pediatric population, probably because: * the levels of Ca_s do not reflect considerably small variations in intracellular Ca * Ca_s comes from many nutritional, hormonal and environmental factors that are not analysed in this paper. No relationship was found between Ca_s and SAP, or DAP in the studied pediatric population.

32.

OVIDUCTAL MICROENVIRONMENT AND FERTILIZING ABILITY OF HUMAN SPERMATOZOA: A MODEL OF STUDY IN VITRO

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Within the female reproductive tract, the spermatozoon (sp) interacts with female tract fluids, which could maintain sperm viability. It is known that if sp prematurely loss their acrosoma they could not fertilize the oocyte. The objective of the study was to develop an *in vitro* system that mimic the oviductal microenvironment found by the sp and to evaluate the sperm fertilizing ability by mean of the acrosomal status. The oviductal tissue was obtained from pre-menopausal patients (n=4; $46 \pm 1,7$ years), scheduled for non-malignant hysterectomies at day $9 \pm 2,8$ of their cycles. Tissues were cultured in Ham-F12/DMEM medium for 48 h ($37^\circ C$, 5% CO_2). At the end of the incubation, the conditioned medium (CM) was collected. Protein concentration was determined by the Bradford method. Motile sp (selected by percoll gradient) from 5 normozoospermic donors were incubated in capacitating conditions for 22 h, and then, further incubated for 2 h in the presence of increasing CM protein concentrations (0,2-0,8 ug/ul). Sperm viability was assessed with Eosin Y. Sperm acrosome reaction (AR) was induced with follicular fluid (FF, 20 % v/v) and acrosomal patterns were evaluated by Pisum sativum technique. The difference between % induced-AR and % basal AR was defined as inducible population (IP). The % of viable sp after capacitation or following the distinct incubations was similar to that observed post percoll (> 75 % of viable sp). The IP was not affected by exposing of sp to different MC protein concentrations ($p > 0.05$, n=5). The results suggest that incubation of capacitated sp in the presence of the studied CM protein concentrations maintains sp viability and the sp ability to respond to physiologic-inducers of AR. These results support the hypothesis that the oviductal environment could contribute to maintain the sp fertilizing ability.

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33. DEVELOPMENT OF A SANDWICH EIA TO DETERMINE PLASMA LEVELS OF ANTI-OXIDIZED-LDL (a-ox-LDL) ANTIBODIES

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It has been found that the increased levels of oxidized low density lipoproteins (ox-LDL) and the presence of a-ox-LDL, are important markers of the atherogenic process. The aim of this work was to develop a sandwich EIA to determine plasma levels of a-ox-LDL. The LDL were purified from human plasma, by density gradient sequential ultracentrifugation in the range of 1.019–1.063. The purified LDL were analyzed in 5-15% gradient SDS-PAGE. A unique band of MW > 400 kDa was detected. The LDL were oxidized in the presence of Cu²⁺. The ox-LDL was used to immunize a rabbit to rise polyclonal antibodies (Ab). The reactivity of the immune serum was confirmed by Western blot from an 8% SDS-PAGE of ox-LDL and aged human serum. Non-oxidized LDL were used as negative control. The immune or pre-immune (control) rabbit serum were used as primary Ab and a peroxidase conjugated anti-rabbit IgG was used as secondary Ab. With the immune serum, a unique band of MW > 400 kDa was obtained for ox-LDL and aged human serum. None of the antigenic fractions showed reactivity with pre-immune serum. The ox-LDL were used to sensitize microwell strips in order to detect a-ox-LDL by means of EIA, which was developed with a peroxidase-conjugated anti-rabbit IgG, followed by tetramethylbenzidine-H₂O₂, stopped with H₂SO₄ and read at 450 nm. Preliminary results indicated an intra-assay variation coefficient of 8.32%. The early detection of a-ox-LDL plasmatic levels through the developed assay, would be a useful tool as an atherogenic marker in patients with immunological disorders, cardiovascular disease or diabetes.

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34. RHEOLOGICAL APPLICATIONS TO ALLOW INDUSTRIAL CONTROL OF PRODUCTS

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At the medicinal and cosmetic industry, is necessary to make control process, listed for The Good Manufacturing Practices, in order to decrease the number of avoid products listed from the control of Quality Laboratory. The goal of this work was the determination of the apparent viscosity at 10 rpm, at 25°C, and the values rate, in order to establish similar rheological characteristics. The apparatus used was a digital viscosimeter, like Searle. It was chosen two semisolid systems, both o/w. After the preparation, both systems were leave in rest, in order to obtain the definitive structure (t₀), and then it was determinate the apparent viscosity. Both systems showed that the media viscosity limits were low than 5%. It means that this rates were accepted to future productions.

35. USE OF THE PHYSIOLOGICAL SURFACTANTS FOR THE SOLUBILIZATION OF DRUGS NON SOLUBLE IN AQUEOUS MEDIUM

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The capacity of solubilization of the five physiological bile salts: cholate, deoxicholate, hiocholate, quenodeoxicholate and taurocholate were assayed on two non aqueous soluble antimicotic agents: griseofulvin and ketoconazole. The fluorescence emission of these antimicotic agents was used as tool to study their solubilization in bile salts micelle. Griseofulvin enhanced its fluorescence and shifted to the blue in the presence of bile salts micelles. The shift was dependent of the polarizability of the micelle zone where the antimicotic is located. Cholate and deoxicholate showed a good solubilization capacity for griseofulvin: 321 and 394 mol of surfactant per mol of antimicotic respectively. These values decreased in the presence of NaCl in agreement with a compactness of the bile salt micelle due to a decreasing in the repulsion between the bile salts momomers negatively charged. The imidazole and piperazine rings present in the ketoconazole molecule give to it the capacity of fluorescence emission with two vibronic bands at 364 nm and 382 nm respectively. The solubilization in cholate micelle induced an increase in the band at 382 nm, while deoxicholate induced the opposite effect, suggesting a strong interaction between the polar groups of ketoconazole molecule (imidazole and piperazine rings) and the -OH of these bile salts. The solubilization capacities were 47 mol and 117 mol surfactant per ketoconazole mol for cholate and deoxicholate respectively. The other bile salts assayed did not show any appreciable solubilization capacity.

36. STUDY OF THE INCORPORATION AND DELIVERY OF NON SOLUBLE DRUGS IN SEMI SOLID SYSTEMS OF TOPIC APPLICATION

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It has been demonstrated that the vehicle where a drug is placed in a semisolid system affects in a significant manner the drug delivery. The enhancers are substances that improve the permeation of the skin. The goal of this work was to study the incorporation of Ketoconazol and Griseofulvin in semisolid systems: hydro and lipo character. It was used semisolid system formed by: flexible chain polymer, fatty alcohol, White Petrolatum, non ionic ecologic surfactants proceeding from *Olea europaea*, such as Polyethylene Glycol G 7 Oleate, Oliven 700 and alkil polyglycoside. Plantaren 1200. The best incorporation was observed in the systems:

a- Cetyl Alcohol 8% + Oliven 700 (Polyethylene Glycol 4 olivate) 3% + White Petrolatum 9%

b- PEG 1500 5% + PEG 400 5% + Plantaren 1200 2%

Also, it was found that the drugs delivery follows a kinetics of first order which was independent of the pH medium and the initial drug concentration.

The results shows that the drugs placed in system "b" were better delivered.

37

JELLIFIED SYSTEMS I: DELIVERY OF α -TOCOPHEROL (VIT. E) IN SYSTEM OF DERMIC USE AND ITS REOLOGICAL PATTERN

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The massive use of dermatology preparations makes necessary the balance between physics chemical properties of stability y preservation, and the delivery of a drug into the extract cornea. The goal of this work was to study the delivery of alpha tocopherol (vitamin E) which it has been included in jellified system of carboxi polyethylene and its reologhical properties.

It was selected different systems formed by carboxypolymethylen 940 and several surfactants derived from the olive oil (*Olea europaea*) such as: Oliven 700 and 900, alkyl polyglycosides as: Plantaren 1200 from sead corn (*Zea maiz*). All these have a great ecological impact, and sorbitan monooleate polyoxyethylrne (Tween 80), polymers of flexible chain such as polyethylenglycoles. It was found that the drug delivery was carried out during the four first hours. The system Oliven 700 al 1% showed low dynamic viscosity and hysteresis at 25°C. The values of these parameters decreases at 37C improving the delivery of the Vitamin E, and in this way simulating the behaviour of the gel on the skin.

38.

JELLIFIED SYSTEMS II: DELIVERY OF HYDRO-CORTIZONE IN SYSTEMS OF DERMIC USE AND ITS REOLOGICAL BEHAVIOUR

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The knowledge in the chemistry of the siliconic derivatives have many advances This feature lets the utilization of many of them in dermathological systems due to some properties as chemical inertia, heat resistant and the condition of non irritation the skin. The goal of this work was to study the delivery of hydrocortizone included in gel system of carboxipolimethylen 940, its rheological properties were also studied. It has been selected gel systems formed by similar polymers. The systems selected were: Olivem derivatives (*Olea europaea*) such as Oliven 700 and 900, Plantaren 1200 from Zea mais, monooleato de sorbitan polioxietilénico Tween 80, and siliconics compounds ,such as (tetraméric ciclomethicon, DC 244), copoliol dimethycon (DC 190). It was studied the rheological patterns of the gels and the delivery of the hydrocortizone.

The siliconic compounds showed low delivery of the drug and the best dynamic viscosity. The optimal delivery rate was observed for the systems of Olivem 700. The hysteresis loop did not influence the drug delivery when a shearing stress of 100 rpm and a temperature of 37°C, in order to enhance the Hidrocortizone delivery, simulating the epidermal aplication.

39.

FREQUENCY AND PHISIOPATHOLOGY OF DOGS INFECTED WITH *Toxocara canis*

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Toxocara canis is a nematode that infects dogs naturally. The mature parasites are eliminated by puppies younger than 6 months old and females in the nursing stage. Dogs ingest the infecting eggs that hatch in the small intestine and the larvae penetrate the intestine wall, reach blood circulation, invade the liver, lungs and continue their maturation moving to the gastrointestinal tract where they mature. The aim of this work was to determine *T. canis* frequency by means of detecting eggs in faecal matter and antibodies in the serum using immunoblotting technique and to evaluate the phisiopathology in the studied dogs. 70 canines (puppies to adults) were studied. Faeces were analyzed both through direct observation and using concentration techniques. Antibody detection in sera was performed using western blotting techniques. Secretor - excretor antigen of *T. canis* larvae was obtained by the Savigny procedure. *Ancylostoma caninum* (22%) and *Trichuris vulpis* (12%) eggs were found in the adult faeces. *T. canis* eggs (10%) were found in puppies. The 200, 120, 70, 55 and 30 kDa bands detected with the western blotting technique were clinically compatible with Toxocariosis (30% of the whole group). The dogs showed symptoms as a swollen abdomen, dull rough skin, diarrhea and irritability. Eosinofilia, anemia and leukocytosis were observed in the blood count. The hepatic enzymes were: LDH over 300 UI/l, AST more than 30 UI/l, and ALT more than 40 UI/l. 30% of the dogs was observed to be infected with *T. canis*, being the liver the most affected organ.

40.

TAXONOMIC STUDY OF THE *SIMAROUBACEAE* OF THE SANTA FE PROVINCE

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The family *Simaroubaceae* comprises 25 genera and 200 species from tropical and subtropical regions of the world. In Argentina the family *Simaroubaceae* includes six genera, but only one of them is present in the Santa Fe province: *Castela* Turpin. The present work contributes to the knowledge of this family providing keys to the identification of its taxa based on morphological characters and a map of their geographical distribution. The methodology consisted of a deep bibliographical review, consultation of national herbaria with important collections of the province flora (SF: Esperanza; SI: Darwinion, San Isidro; UNR: Rosario), lab work to confirm their identity and the field experience of the authors. As a preliminary result, only two species of the genus *Castela* Turpin were assessed: *Castela coccinea* Griseb. with its geographical distribution concentrated mainly in the northern departments, and *Castela tweedii* Planchon, which reaches the south of the province. **NOTE:** samples of *Ailanthus altissima* (Mill.) Swingle belonging to this family were observed by one of the authors in the San Cristobal department, though it has been impossible to confirm if this species has spontaneously naturalized. It is here provided taxonomic information, illustrations and a distribution map.

41. EMULSION PROPERTIES OF PRE-HEATED BETA-LACTOGLOBULIN

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Because of its high nutritional value and functionality, whey protein concentrates (WPC) obtained from dairy processing has become an important source of functional ingredients used in many formulated foods. Beta-lactoglobulin (β -LG) is the major protein in whey and therefore dominates the behavior of the total whey protein system. Functional properties of β -LG molecules are primarily dependent on their degree of denaturation. When a heated sample of the protein is cooled, the species that can be present in solution are native and non-native monomers, and aggregates of denatured β -LG, associated through disulfide linkages. The relative amount of the different species depends on experimental conditions, such as concentration, temperature, pH and time of heating. In this work, 3 mM β -LG solutions (20 mM phosphate, pH 6.8) were heated at 65 and 85°C during 0, 1, 3, 5, 7, 10 and 15 minutes, and then cooled to room temperature. Polyacrilamide gel electrophoresis (PAGE) in the presence of sodium dodecyl sulfate (SDS), without 2-mercaptoethanol, was used to determine the extent of irreversible aggregation. Emulsifying properties of pre-heated samples of β -LG were determined by turbidity measurements of O/W emulsions (corn oil/ β -LG solutions). Heat treatment at 85°C decreases the emulsifying activity of the protein. This fact is due to the presence of oligomers of relative high molecular weight. These species are not fast enough to adsorb to the surface of a droplet before droplet-droplet collisions lead to coalescence, during emulsion formation.

42. A CONTINUOUS CENTRIFUGE BIOREACTOR FOR ALCOHOLIC FERMENTATION OF *Saccharomyces cerevisiae*

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The bioreactor consists of an anaerobic recipient with an external control of pH and temperature. An acrylic disc is driven by a motor, whose rpm can also be controlled. The gel medium (agar or alginate) which contains the yeast cells, is placed over the surface of the disc. The substrate (glucose) is supplied by dropping in the center of the disc using a peristaltic pump. The product of the reaction (ethanol) is eliminated through a lock placed at the bottom of the reactor. The substrate concentration (optically active) is determined by polarimetry. Ethanol concentration is determined by redox reactions in Conway's chambers and by density.

When mass balance is applied to this system:

$$\text{mass}_{\text{in}} - \text{mass}_{\text{out}} + \text{mass}_{\text{reaction}} = \text{mass}_{\text{accumulated}}$$

the performance of the reactor (expressed in percentage) can be obtained. In our case, the higher efficiency obtained was 58,93% when the control parameters were fixed in: days of work, 4; rotor speed, 25 rpm; substrate flux, 3 drops/minute; glucose concentration, 1M; temperature, 30° C; initial pH, 5,0.

This system has the following advantages: is a continuous reactor, the product is eliminated from the reaction plate avoiding the inhibition by final products, the system has the possibility of enclosing other microorganisms or enzymes and it can be easily scale-up.

43. EXTERNAL GLOMERULAR DIAMETER VARIATIONS IN REMAINING KIDNEYS OF "L" LINE RATS UNDERGOING UNILATERAL NEPHRECTOMY

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In a previous study demonstrated that the weight of the remaining kidney (RK) in "L" of line rats (LLR) undergoing unilateral nephrectomy is different depending on sex and age. The aim of this work was to assess external glomerular diameter (EGD) variations in the RK of LLR undergoing a nephrectomy depending on sex and age. We used 64 LLR, classified according to: 1) Sex: Females (F) (n=32) and Males (M) (n=32); 2) Age: Young (Y) 50 days (n=32) and Adults (A) 85 days (n=32) and 3) Treatment: Nephrectomized (N) and Control (C). The animals of group (N) underwent a left nephrectomy 45 days later the animals were euthanased, and the right remaining kidney was removed. The animals of group (C) were treated under the same conditions of group N. The kidneys were fixed in neutral 10% formaldehyde solution and embedded in parafin. 5 μ m sections were obtained and stained with hematoxylin-eosin. The observations were performed with a microscope equipped with a Shimadzu eyepiece, calibrated for μ m, and we measured the EGD at 100X going through the cortex counting 10 rounded-shaped glomeruli presenting vascular pole in the section. To study the effect of age, sex and treatment we used ANOVA. The results of the triple and double interactions were not significant. Concluded that there is a significative effect of sex and treatment, but not of age, on the EGD of the RK.

44. CONTROL LOCUS, HOPLESNESS AND FACING (OR SCOPING) IN ADULT PATIENTS WITH CONNECTIVE TISSUE DISEASES (SCLERODERMIA, SLE AND RA)

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The present work is focused on perception exploration of disease control (control locus), and corresponding symptoms in patients with diagnosed connective tissue diseases such as systemic lupus erythematosus, scleroderma and rheumatoid arthritis, assess their pólones level, and analyse the coping strategies utilized. The patients are adult women consulting in a Dermatology Service in the public hospital in Rosario, aged 21-71yr. Evolution time after diagnosis: 1 yr. Collecting data instruments: hopelessness index in arthritis, adapted for the present work, coping strategies listing, and questionnaire constructed for the present work to explore patient fear and concern. Conclusions of pilot sample show regarding control locus, there is no clear position in the interview (there is no internal or external blame). Nevertheless, the majority refer self responsibility regarding treatment and disease control, although they refer there is no effectiveness in pain management. Regarding coping, the majority of patients use strategies focussed on problem solution and search for social support. There are generally low and intermediate levels of hopelessness. Disease and complication fear, autonomy loss are frequently referred such as pain, autonomy loss, and secondly: family concern.

45. RHEOLOGICAL FACTORS AFFECTING BLOOD FLOW IN EQUINE LAMINITIS

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Acute laminitis is associated with a primary process involving other systems, mainly the digestive tract due to intake of rapid fermentable concentrate food (starch). The disease is characterized by involvement of the coronary fold and the over imposed and richly vascularized lamellae between third phalange and hoof. The solid structures activate venous backward circulation during the march providing reperfusion of capillary bed (layer). Although laminitis pathogenesis is not wholly characterized, the occurrence of microthrombosis and increase in plasma protein was demonstrated. Objective: to study white blood cells (WBC), total protein (TP) and fibrinogen (Fb) modifications, and their relation with plasma viscosity in horses with acute spontaneous laminitis. Methods. Whole blood samples of horses bearing laminitis (n=8) and 8 healthy controls were analyzed. Laboratory Assessments: WBC $\times 10^3/\mu\text{l}$ with automatic counter, plasma Fb, mg/dl by gravimetry; and TP (g/dl) by colorimetry (autoanalyzer, Hitachi 902). Plasma viscosity (η_{sp} , mPa.s) at 37°C with rotational viscometer at 115 s⁻¹ share rate. Wilcoxon test for paired data was applied. Results expressed as median and range show significant modifications ($p < 0.05$) in TPr [7.95, 7.50- 8.70], η_{sp} [1.8, 1.30-2.20] and Fb [1.05, 0.95-1.30]. WBCs increased significantly ($p < 0.01$) [11.5, 9-17]. We can conclude that in the acute stage, the phalanges pain conditioned the support and flow decrease, consequently, an increase in fibrinogen plasma viscosity and WBCs with a low deformability increase blood circulation impairment.

46. ERITHROCYTIC AGGREGATION AS A RISK FACTOR FOR DEVELOPING MICROANGIOPATHIC SKIN LESIONS IN DIABETIC PATIENTS

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Diabetic microcirculatory changes may produce clinical disorders in several organic territories as skin. This report studies erithrocytic aggregation in 65 diabetic patients (45 with microangiopathic skin lesions and 25 without it) and in 19 healthy controls for assessing this hemorheologic parameter as a risk factor in the development of skin lesions. Erithrocytic aggregation was analyzed through the Aggregate Shape Parameter (ASP). Blood was obtained from vein puncture, anticoagulated with EDTA and centrifuged for separating red blood cells from plasma. These cells were re-suspended in 2 % autologous plasma. Exceeding a higher ASP in diabetic patients in relation with controls (0.65 ± 0.15 vs. 0.25 ± 0.13 , $p < 0.001$), the homogeneity of the variance (Levene's test = 0.255) allowed to apply the one factor ANOVA showed highly significant differences among groups ($p < 10^{-5}$). The post test of minimal significant differences evidenced notorious disparities between patients with skin lesions and controls. Considering 0.65 as limit value for classifying diabetic patients with and without skin lesions, patients with $ASP \geq 0.65$ revealed a relative risk (RR) = 1.74 (95% confidence interval -1.23;2.45-). Diabetic patients with $ASP > 0.65$ have nearly a double probability for presenting microangiopathic skin lesions.

47. DIGESTIVE MORPHOMETRY IN THREE LINES OF DIABETIC RATS AT 12 MONTHS OF AGE

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Considering the link between diabetes and digestive system, the present report compares morphometrically distinct digestive organs in several spontaneous type 2 male diabetic rats: 5 β (obese and with late diabetes), 5 eSS (non obese and with early diabetes), 5 eSMT (early diabetic and with overweight) and 5 Wistar (controls) receiving a balanced commercial diet and water, both *ad libitum*. Animals were sacrificed with ether overdose at 12 months of age. Small and large intestine as well as liver were dissected out, weighed and/or measured. Data, expressed as $x \pm$ standard error, were statistically analyzed through the Kruskal- Wallis test. The small intestine weight did not differ between β (11.69 ± 0.88 g) and eSMT (12.08 ± 0.41 g) but significantly exceeded those of Wistar (7.39 ± 0.65 g) and eSS rats (8.95 ± 0.30 g) ($p < 0.05$). In congruence with these findings, the reticular structure of Auerbach's plexus disappeared at this age in β and eSMT rats but did not in Wistar and eSS animals. Finally, it is meaningful to add that all these spontaneously diabetic rats evidenced higher body weight and lower small intestinal weight and length than those reported in animals with chemically induced diabetes.

48. RENAL LECTINHISTOCHEMICAL STUDY IN 1 YEAR - OLD DIABETIC RATS USING TWO PROCEDURES

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This communication compares a previously used semi-quantitative procedure with a quantitative one: the optical density (OD) in kidneys of 1 year - old type 2 diabetic rats. 12 male rats: 6 β (obese and diabetic) and 6 α (controls) were sacrificed at 12 months of age. Kidneys were successively fixed in Bouin's fluid and 10% neutral formalin, embedded in paraffin and exposed to Arachis hypogaea (PNA), Canavalia ensiformis (Con-A), Dolichos biflorus (DBA), Glycine max (SBA), Ricinus communis (RCA), Triticum vulgaris (WGA) and Urex europeus (UEA-I). Complementarily, representative digital photos were processed through the program Image-pro Plus version 1.1 with a video camera Sony Exwavehad. Comparing the semi-quantitative procedure (0-1-2-3) with the quantitative one ($OD \geq 200 = 0$; $150 < OD < 200 = 1-2$ and $OD \leq 150 = 3$) some concordances and discordances for different lectins were registered in β and α superficial and juxtamedullary glomeruli. These finding would suggest that both procedures may be used in concordant data whilst OD could be the chosen procedure in case of discordant results for its higher validity. Furthermore, when OD is unavailable the diabetic kidney may be studied from a lectinhistochemical standpoint employing those lectins with matching results in both procedures.

49. DIGESTIVE SEGMENTS DERIVED FROM CAUDAL INTESTINE DURING EARLY FETAL DEVELOPMENT

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This communication evaluates morphologic aspects of transverse, descending and sigmoid colon and rectum during early fetal development. Serial 7 μ m thick sections from the abdominal-pelvic zone of 9th and 10th week-old six human fetuses, obtained from spontaneous abortions in public hospitals, were sequentially exposed to varied histological and histochemical procedures. They were planimetrically processed and superposed for spatial reconstruction. These celomic segments, excluding the rectum, were located from right to left side (transverse colon), from cranial to caudal zone at the left side (descending colon) and showed two loops (sigmoid colon) pushing medially the resting tube (rectum). The corresponding tubular diameters were constant but lower than that of the small intestine. From a histological standpoint, these segments did not differ from the remaining digestive ones. The colonic epithelium was columnar and evidenced higher portions where cells protrude towards the lumen providing a star-like aspect to the cavity. These elevations rendered more evident in the rectal zone where its apical portion acquired a fan-like aspect. Despite these accidents persisted in the anal conduct, they distally disappeared whilst the epithelium became progressively stratified. It may be supposed that the luminal reduction in these segments could be due to a volume diminution in the stream of amniotic liquid because of its possible reabsorption in the proximal digestive portions. The described structures in rectum and anal conducts preannounced the formation of columns, valves and rectal and anal sinuses.

50. MEASUREMENT OF RAT SERUM LEVELS OF α_2 -MACROGLOBULIN (α_2 M)

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In rats, α_2 M is an acute phase reaction glycoprotein. It inhibits proteinases and binds cytokines and growth factors. Sodium monofluorophosphate (MFP), a drug used in the treatment of osteoporosis and the prevention of caries, binds to α_2 M, forms a complex and modifies the antiprotease activity. There are several techniques for measuring α_2 M, but they permit to obtain relative values, and they are time-consuming. The aim of this work was to develop a method for the measurement of plasmatic levels of α_2 M in the rat. The proposed technique is based on the dot-blot technology for protein measurement. Basically, it consists of: 1- The binding of serum proteins to a nitrocellulose membrane and subsequent blocking of non-specific sites 2- The incubation with primary antibody to rat α_2 M. 3- The incubation with antiGuinea Pig Immunoglobulin G alkaline phosphatase conjugated secondary antibody. 4- The development of dots with a specific compound that produces a blue chromophore. 5- The digitalization of the blue area. 6- The measurement of intensity (I) and area (A) of the dot with a specific software. The technique described above was applied to solutions with known concentrations of rat α_2 M. A and I were measured and Axl was calculated. The linear regression between Axl and α_2 M concentrations was statistically significant. The coefficient of variation for a serum pool of normal rats was 11.5%. This technique allows to measure rat α_2 M concentration, the results can be obtained in approximately 6 hours and the complexity and the cost of the method are low.

51. DIFFERENT CHEMICAL FORMS OF RAT INTESTINAL ALKALINE PHOSPHATASE (AP)

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AP binds calcium (Ca), which modifies its activity in a biphasic pattern. Preliminary experiments suggested that Ca would alter the molecular weight of AP. The aim of this work was to investigate the change in molecular weight and activity of AP in the presence of Ca. AP was purified from the small intestine of rats. A primary anti-AP antibody raised in Guinea Pig and a goat anti-Guinea Pig IgG peroxidase conjugated secondary antibody were used to detect the enzyme. AEC (3-amin-9-ethylcarbazole) was used as peroxidase substrate and pNPP (p-nitrophenylphosphate) or BCIP (5-Br-4-Cl-3-indolyl phosphate) were used as substrates to detect AP activity. Two types of experiments were carried out with AP: 1) Chromatography on Sephadex G200: two fractions (f1 and f2), at different elution volumes (VE) were detected with AEC: $VE_{f1} = 13$ ml; $VE_{f2} = 21,7$ ml. Specific activity of f2 was superior than f1. Treatment of PA with Ca 50 mM reduced the amount of protein of f2 and proportionally increased that of f1, which in turn had negligible activity. 2) 7% polyacrylamide gel electrophoresis with and without Ca: after electrophoresis, proteins were transferred to a nitrocellulose membrane, the enzyme was detected with AEC and its activity with BCIP. Two bands of AP were detected, one of them without activity (f1) and the other with activity (f2). Relative mobility of bands were: $Rf_{f1} = 0.16$; $Rf_{f2} = 0.45$, respectively. Conclusions: 1- There are two forms of PA with different molecular weight and activity. 2- The fraction of greater molecular weight has negligible activity. 3- Ca^{++} would induce the conversion of the fraction of low molecular mass into that of high molecular weight.

52. CHANGES IN BONE PHOSPHOROUS CONTENT (BPC) IN RATS TREATED WITH SODIUM FLUORIDE (NaF)

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Administration of NaF increases bone mass in rats. Nevertheless, the composition and quality of bone formed under fluoride (F) stimulus is still matter of study. The aim of this work was to investigate the effect of treatment with NaF on BPC in rats and the factors which modify it. Experiments were carried out in 50-day old female rats IIM/FcM. Exp.1: 4 groups of 4 rats each were given the following doses (μ mol/day.100g bw) of NaF: Group A: 0, B: 20, C: 40 y D: 60. After 30 days rats were killed and femurs were excised. BPC (mg/g dry bone) decreased as a function of F dose. A: 93 ± 14 ; B: 79 ± 16 ; C: 61 ± 15^a ; D: 60 ± 9^a , $p < 0,01^a$ states differences with regards to group A. In the following experiments, the F dose was 40 μ mol/day.100g bw, by gastric tube. Exp.2: 4 rats received one dose of F. Phosphatemia (mg/dl) increased significantly (basal: 8.7 ± 0.6 , 60 min: 13.0 ± 0.3 $p < 0.001$) and phosphaturia (mg) decreased during the 1st hour. Exp.3: Renal and plasmatic flow, and glomerular filtration were not affected after F administration ($n=4$). Exp.4: 4 thyroparathyroidectomized (TX) rats received one dose of F. Phosphatemia increased in both groups but F did not modify renal handling of phosphate in TX. Exp.5: Phosphate concentrations in erythrocyte of rats after one dose of F did not change, although phosphatemia increased steadily. Conclusions: The treatment with F decreased BPC. One cause of this change would be the effect of F in renal phosphate handling. Neither renal hemodynamia nor the inhibitory effect on PTH actions would be the causes of disruption in phosphate renal handling. Efflux of phosphate from cells might not be the cause of the increase in phosphatemia.

53.

MODIFICATION OF RAT CYTOPLASMIC INTESTINAL ALKALINE PHOSPHATASE (AP) ACTIVITY BY CALCIUM

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In vitro, the activity of AP exhibits a biphasic response to calcium (Ca^{++}): it increases at Ca^{++} concentrations lower than 10 mM, and decreases if concentration of Ca^{++} is greater than 20 mM. *In vivo*, Ca^{++} produces an increase in brush border activity of enterocytes but decreases the activity in the cytosol. The aim of this work was to study changes in cytoplasmic activity of AP by modification of cytosolic concentrations of Ca^{++} . Female adult rats IIM/FcM were used in the experiments. 3 cm of duodenum were isolated *in situ* by mean of two ligatures. A catheter was inserted and it was used to flow 2 ml of the following solutions into the lumen: Group A and B: NaCl 150 mM; Group C and D: CaCl_2 50 mM + NaCl 150 mM; Group E: CaCl_2 50 mM + NaCl 150 mM + calcium ionophore A23187 0.4 mM. Within 20 min the duodenum was extracted, fixed, paraffin embedded and samples were sectioned at 10 μm . Sections of groups B y D were exposed to EDTA 500 mM and CaCl_2 50 mM for 30 and 20 min, respectively. AP activity was put in evidence by incubation with 5-Br-4-Cl-3-indolyl phosphate. The activity in cytosol was measured as absorbance units on digital images. Values were analyzed by one-way ANOVA. Mean $\pm$ SD are shown in the table. Differences were considered significant if $p < 0.05$. The same letter indicates differences between groups

Group	n	mUnits of absorbance	Comparison among groups
A	4	78 $\pm$ 32	a, b
B	4	5.2 $\pm$ 8.7	a
C	4	39 $\pm$ 26	b, c, d
D	4	103 $\pm$ 45	c
E	4	81 $\pm$ 45	d

AP activity in the cytosol of enterocyte decreases as a function of luminal Ca^{++} . This effect could be caused by a decrease in the concentration of the ion in the aforementioned compartment.

54.

PLASMATIC CLEARANCE OF α_2 -MACROGLOBULIN-MONOFUOROPHOSPHATE (α_2 M-MFP) COMPLEX IN THE RAT. COMPARISON OF TWO METHODS OF MEASUREMENT

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In the rat, $\alpha_2\text{M}$ is an acute phase reaction globulin, which protects tissues from the action of proteinases. $\alpha_2\text{M}$ plays an important role in MFP pharmacokinetic, forms a $\alpha_2\text{M}$ -MFP complex and inactivates the antiprotease activity. Based on these phenomena we developed two methods to calculate the plasmatic clearance of $\alpha_2\text{M}$ -MFP (k_c). **Method A:** with a mathematical model and the values of plasmatic $\alpha_2\text{M}$ -MFP concentrations after a single intravenous (iv.) dose of MFP. **Method B:** with plasmatic concentrations of $\alpha_2\text{M}$ after an oral dose of MFP. This work compares the values of k_c by both methods with that obtained by iv. injection of purified complex. Three types of experiments were carried out in adult IIM/Fcm rats. 1) iv. administration of $\alpha_2\text{M}$ -MFP and measurement of its concentration for 75 min. Data followed a mono-exponential function with $k_c = 0.1231 \pm 0.0227 \text{ min}^{-1}$. 2) iv. administration of a single dose of MFP, and measurement of $\alpha_2\text{M}$ -MFP concentrations. Data were fitted with a mono-exponential function with $k_c = 0.1819 \pm 0.153 \text{ min}^{-1}$. 3) iv. administration of an oral dose of MFP and determination of $\alpha_2\text{M}$ concentrations by dot-blot. Data were adjusted with a mono-exponential function with $k_c = 0.0082 \pm 0.0042 \text{ min}^{-1}$. k_c obtained in experiments 1 and 2 significantly differed from that obtained in experiment 3. ANOVA $P < 0.05$. Conclusions: 1- The value of k_c obtained with method A described the plasmatic clearance of $\alpha_2\text{M}$ -MFP. 2- Method B underestimates the value of k_c .

55.

FEMALE RAT PRODUCTION IN A NEW ENVIRONMENT LINE b

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At Cátedra de Biología, different line *b* rat parameters are studied nowadays in order to evaluate the possibility of replacing the extinct line α as control line β murine obesity model. Rats *b* come from Instituto de Investigaciones Médicas de Rosario (IIM). In this work variables connected with breeding production were analyzed: fertility coefficient (CF), superviving coefficient (CS) and combined coefficient (CC) in two circumstances: when the first generation is produced in the new environment (1^aG , $n=11$) and another, when the sixth generation is produced (6^aG , $n=14$). These values were compared through ANOVA and Tukey's test with those of the origin population (P.o) provided by the IIM (Media $\pm$ SEM; *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$, ns: $p > 0.05$)

	(P.o)		(1^aG)		(6^aG)
(CF)	1.150 $\pm$ 0.881	*	0.740 $\pm$ 0.045	*	1.069 $\pm$ 0.108
(CS)	0.830 $\pm$ 0.367	ns	0.660 $\pm$ 0.042	**	0.910 $\pm$ 0.053
(CC)	1.120 $\pm$ 0.880	***	0.480 $\pm$ 0.039	**	0.946 $\pm$ 0.305

CF, CS and CC differs ($p > 0.05$) between (P.o) and (6^aG). Breeding birth weight (g): differs ($p > 0.05$) (1^aG) = 6.90 ± 0.24 vs (6^aG) = 6.61 ± 0.27 ; and from joined for mating to birth time (days): differs ($p < 0.01$) (1^aG) = 39.30 ± 1.50 vs (6^aG) = 30.55 ± 2.30 . Pregnant female was: (1^aG) = 90% vs (6^aG) = 64%. In (1^aG) the new bioterio environmental conditions impacted in female production. In (6^aG) coefficients regained the origin population values. (6^aG) contrasting with (1^aG) shortened from joined for mating to birth time, but pregnant female decreased. Because of the importance that the registered values have in the breeding production economy, these should be evaluated in the following generations.

56.

EFFECT OF OLEOYL ESTRONE ON BODY WEIGHT, FOOD INTAKE AND SOME PLASMATIC VARIABLES IN OBESE β RATSPosadas M¹, Labourdette V¹, Gayol M del C¹, Olguin M², Zingale M², Elias H², Revelant G², Calderari S^{1,2}.¹Cát. de Biología. Fac. de Cs Médicas. UNR; ²Cát. de Bromatología. Fac. de Cs Bioquím. y Farm. UNR.

It has been proposed that oral administration of Oleoyl estrone, a synthetic acyl ester of estrone and oleic acid, promotes a decrease in food intake and an increase in global termogenic capacity originating a negative energetic balance this way. Energy deficit would be supplied mainly by fat storages allowing weight loss and the improvement of the problems associated with obesity. To investigate Oleoyl estrone effects in the obese hypertriacylglycerolemic diabetic β rats, 200 days old same weight male β rats were given a daily oral gavage of Oleoyl estrone (10 nmol/g) for 10 days. Animals were fed ad libitum and maintained in the same environmental conditions. Body weight and food intake values were registered daily. Glycemia, cholesterolemia and triacylglycerolemia were quantified on day 11.

Experimental group data (OE; $n=9$) were compared to those from control group (T; $n=9$) using Student t Test.

Body weight (g): OE: 365.6 ± 7.8 vs T: 426.4 ± 10.1 ($p < 0.001$)

Food intake (g/día): OE: 25.9 ± 2.1 vs T: 29.9 ± 3.1 ($p > 0.05$)

Glycemia (g/l): OE: 1.67 ± 0.08 vs T: 2.66 ± 0.34 ($p < 0.05$)

Cholesterolemia (g/l): OE: 0.49 ± 0.06 vs T: 1.21 ± 0.08 ($p < 0.001$)

Triacylglycerolemia (g/l): OE: 1.76 ± 0.15 vs T: 2.73 ± 0.13 ($p < 0.001$)

It can be observed that O-E group body weight resulted significantly lower than controls although there was no statistic difference in food intake between the groups. This is consistent with the hypothesis that postulates for oleoyl estrone a catabolic action in energy homeostasis circuit. Diminished glycemia, cholesterolemia and triacylglycerolemia in OE group might be due to the proved benefits that fat depots reduction exerts on such variables. Similar findings, although having different magnitude, have been verified in rodents with diverse types of obesity.

57. INFLUENCE OF PHYSICAL TRAINING IN THE MAINTENANCE OF THE BENEFITS OBTAINED BY PHYSICAL TRAINING-FOOD RESTRICTION IN OBESE β RATS

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Here it is evaluated the influence of physical training in the maintenance of the benefits obtained in male β rats by a treatment that combined physical training and food restriction, once this restriction was suppressed. Rats that had been deprived by prevented food source access each other day and physically trained from 60 to 120 days old. Training had taken place five times a week for 15 minutes a day at a speed of 30 m/min in an ECO 3/6 rodent threading machine. From 120 (Beginning) to 200 days old (End) all the animals were fed *ad libitum*. **ER**: went on training (n=6) and **R**: stopped training (n=5). Student t test for paired data. Mean $\pm$ SD, Beginning (B), End (E), difference (dif). **ER**: Biomass (g) (B): 269.0 $\pm$ 25.5, (E): 420.0 $\pm$ 29.6, dif: 151.0 $\pm$ 13.9 (p<0.001); Basal Glycemia (g/l) (B): 0.95 $\pm$ 0.13, (E): 1.33 $\pm$ 0.11, (dif): 0.38 $\pm$ 0.06 (p<0.001); Glycemia after overload (g/l) (B): 1.16 $\pm$ 0.22, (E): 1.27 $\pm$ 0.12, (dif): 0.11 $\pm$ 0.13 (p>0.05); Cholesterolemia (g/l) (B): 0.63 $\pm$ 0.14, (E): 0.80 $\pm$ 0.18, (dif): 0.20 $\pm$ 0.26 (p>0.05); Triacylglycerol (g/l) (B): 0.72 $\pm$ 0.34, (E): 2.24 $\pm$ 0.64, (dif): 1.53 $\pm$ 0.66 (p<0.01). **R**: Biomass (g) (B): 272.6 $\pm$ 34.6, (E): 410.0 $\pm$ 19.1 dif: 137.4 $\pm$ 27.1 (p<0.001) Basal Glycemia (g/l) (B): 1.18 $\pm$ 0.25, (E): 1.64 $\pm$ 0.26, (dif): 0.45 $\pm$ 0.24 (p<0.05); Glycemia after overload (g/l) (B): 1.23 $\pm$ 0.22, (E): 1.39 $\pm$ 0.10, (dif): 0.16 $\pm$ 0.20 (p>0.05); Cholesterolemia (g/l) (B): 0.84 $\pm$ 0.16, (E): 0.84 $\pm$ 0.19 (dif): 0.04 $\pm$ 0.03 (p>0.05); Triacylglycerol (g/l) (B): 0.76 $\pm$ 0.23, (E): 2.46 $\pm$ 0.60, (dif): 1.70 $\pm$ 0.54 (p<0.01). The fatty panicle weight (g/100g biomass) shows no difference (ER: 5.90 $\pm$ 0.60, R: 6.19 $\pm$ 0.43; p>0.05). Implemented physical training did not avoid either biomass regain or basal glycemia and triacylglycerolemia increase.

58. GROWTH PATTERN COMPARISON OF TWO INBRED LINES OF RATS

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Here it is a comparative study between adulthood diabetic obese β line rats, and non obese b line, looking for possible associations with the adult pathology expression. 20 male rats, 10 β and 10 b were used. After weaning, the animals were fed *ad libitum*, and food intake, biomass and tail length were registered each other day up to 70 days old. Growth curves were calculated, adjusting values according to von Bertalanffy model (characterized by an asymptotic value and a maturity rate, speed at which asymptotic value is reached). In biomass-age growth curve $P_t = A \cdot (1 - b \cdot e^{-k_1 \cdot t})$ and in tail length-age growth curve $L_t = L \cdot (1 - b \cdot e^{-k_2 \cdot t})$. (A and L: biomass/tail length asymptotic values; k_1 and k_2 : maturity rates) (Mean $\pm$ SE; Student t Test). **Biomass 21 days old** (g): β : 51.2 $\pm$ 1.1 vs b: 41.4 $\pm$ 2.6 (p<0.01). **Biomass 70 days old** (g): β : 315.6 $\pm$ 10.1 vs b: 282.1 $\pm$ 7.8 (p<0.05). **Tail length 21 days old** (cm): β : 7.2 $\pm$ 0.1 vs b: 5.7 $\pm$ 0.2 (p<0.001). **Tail length 70 days old** (cm): β : 16.6 $\pm$ 0.3 vs b: 14.4 $\pm$ 0.3 (p<0.001). **A** (g): β : 433.8 $\pm$ 29.0 vs b: 355.8 $\pm$ 13.2 (p<0.05). k_1 : β : 3.46 $\cdot 10^{-2} \pm 2.01 \cdot 10^{-3}$ vs b: 4.13 $\cdot 10^{-2} \pm 2.25 \cdot 10^{-3}$ (p<0.05). **L** (cm): β : 18.8 $\pm$ 0.6 vs b: 17.3 $\pm$ 0.4 (p<0.05). k_2 : β : 3.85 $\cdot 10^{-2} \pm 2.96 \cdot 10^{-3}$ vs b: 2.96 $\cdot 10^{-2} \pm 2.47 \cdot 10^{-3}$ (p<0.05). **Accumulated food intake** (g): β : 1276.0 $\pm$ 41.2 vs b: 1297.0 $\pm$ 37.8 (p>0.05). β line showed biomass and tail length values higher than b line at weaning, at 70 days old and in asymptotic values more speedily than b but showed a higher speed in reaching tail length asymptotic value, in accordance with the higher biomass it must support. Are different growth patterns for both lines. Must be studied thoroughly since the association with the adulthood diabetic syndrome can exist.

59. KERATITIS AND ENDOPHTHALMITIS INFECTIONS

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Keratitis and endophthalmitis are ocular infections that potentially threaten vision and may be caused by bacteria, viruses, parasites and fungi. Prior to the 1970s, keratitis was the consequence of traumatism or ocular surface disease; today the use of contact lenses is considered the most important risk factor in the development of keratitis. Endophthalmitis can be exogenous or endogenous. Those of exogenous origin can be caused by ocular surgery, traumatism, or noncontrolled keratitis. Those of endogenous origin can be caused by septic embolous or spread from periferic tissues. The primary purpose of this paper is to identify the etiologic agents prevalent in severe ocular infections in our setting. The ophthalmologic samples studied were taken either by scraping the cornea (keratitis) or puncturing (endophthalmitis) patients from Ophthalmologic Services at the Centenario and Provincial Hospitals in Rosario between July 2003 and July 2004. The microbiological studies included stains of Gram and Giemsa and cultures in blood agar, chocolate agar, tioglicolate broth, and Sabouraud agar. From a total of 92 samples evaluated, 34 (37%) were positive. The following microorganisms were obtained: *S. aureus* 17.6%, *S. epidermidis* 32.3%, *S. viridans* 8.8%, *S. pneumoniae* 8.8%, *S. pyogenes* 5.9%, *Propionibacterium spp.* 2.9%, *Bacillus spp* 2.9%, *P. mirabilis* 2.9%, *K. pneumoniae* 2.9%, *Enterobacter spp* 2.9%, *Haemophilus spp* 2.9%, *P. aeruginosa* 8.8%, *S. maltophilia* 2.9% and *A. fumigatus* 5.9%. The etiologic agents found in our setting correspond to those presented in similar papers in our country and abroad.

60. CHANGES IN ADIPOSE TISSUE IN β OBESE DIABETIC RATS TREATED WITH OLEOYL-ESTRONE

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Two hundred day-old male rats of inbred IIM β line with obesity, hypertriacylglycerolemia and spontaneous diabetes were treated with 10nmol/g of oleoyl-estrone in 0.2 ml of sunflower oil during ten days (OE). In OE (n= 9) as well as in controls (CO) (n= 9), offered 0.2ml of sunflower oil, biomass and feed intake were assessed daily. On day 11 animals were sacrificed and mesenteric, subcutaneous and retroperitoneal fat depots were dissected. Weight and proximal composition of depots were determined. Fatty acid profile of triacylglycerols in mesenteric depots of both groups was studied with gas chromatography of methyl esters.

OE showed a significant diminution in biomass (g) compared to CO: OE 365 $\pm$ 7.8; CO 426.4 $\pm$ 10.1 (p<0.001). Oleoyl-estrone promoted a significant reduction in the weight of the three fat depots as well as in their lipid content.

No significant differences were found in the reduction rate of lipid content neither in the different fat depots studied (OE) nor in the fatty acid composition in mesenteric depots of OE and CO suggesting that oleoyl-estrone develops an unspecific lipolitic action.

61. PALATABILITY OF DIETS WITH DIFFERENT PHYSICAL FORM AND THEIR EFFECT IN RATS FROM THE DIABETIC LINES eSS AND eSMT

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Certain aspects of the nutritional behaviour can help to identify measures for the prevention or treatment of diseases like type 2 diabetes, strongly influenced by nutrition. Likewise, sensorial characteristics can constitute determining aspects of the appetite as well as of the election of nutrients. We studied in the murine models of genetic diabetes eSMT and eSS the answer to a commercial diet with different physical form (D) and the same declared composition as the previous one (P), in males and females of both lines. At 4 months, 10 animals of each genotype and sex were weighed during 15 days, as so was D intake. Average weight (AW), average daily intake (ADI) and average daily intake related to weight (RDI); fasting glycemia (G0) and glycemia after glucose overload (G120) were measured. Data were compared with those obtained from animals fed with diet P. Line effect ($p < 0.05$) was verified in AW; diet effect in AW, ADI, RDI, G0 and G120; sex effect in AW, RDI, G0 and G120; line by sex effect in AW, ADI, G0 and G120; diet by sex in all the variables and line by diet by sex in AW, G0 and G120. The results might be attributed to the different palatability of diets and/or to the heterogeneity between sexes in the regulation of food intake.

62. *IN VITRO* EFFECT OF Al^{+3} UPON HUMAN RED BLOOD CELLS AGGREGABILITY AND DEFORMABILITY

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Aluminium (Al^{+3}) is a metal with no biological function. Its organic accumulation can lead to toxic effects. We studied the *in vitro* effect of aluminium upon human red blood cells aggregability and deformability. Washed red blood cells from donors without (control) and with Al^{+3} (1 and 10 μM) were used. a) suspension viscosity with cone-plate viscometer; b) rigidity index using nucleopore membranes and c) erythrocyte aggregability by optical method ($2k_2n_0$ and s_0/n_0) were determined. Statistical analysis: Wilcoxon's test for paired groups, data are presented as median and confidence interval (95 %).

(n:10)	η	IR (%)	$2k_2n_0$	s_0/n_0
Control	2,64 (2,48-2,89)	8,7 (6,16-9,27)	1,00 (0,78-1,13)	1,86 (1,84-1,87)
Al^{+3} (1μM)	2,87 ** (2,64-3,36)	13,8 ** (12,2-16,8)	0,65 *** (0,61-0,82)	1,83 *** (1,81-1,84)
Al^{+3} (10μM)	2,81 ** (2,61-2,89)	13,2 ** (12,3-17,5)	0,91 ** (0,79-0,98)	1,81 *** (1,80-1,83)

** $p < 0.01$; *** $p < 0.001$

Our results suggest that at both concentrations Al^{+3} has an effect on plasmatic membrane, disorganises the lipid bilayer thus changing its mechanical properties: increased erythrocyte rigidity and viscosity at high shear rate and decreased red blood cells aggregability.

63. *IN VITRO* EFFECT OF INSULIN UPON HUMAN ERYTHROCYTES AGGREGABILITY

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Previous studies in our laboratory with β strain rat (hypertriglyceridemic obesity and diabetes model) have shown increased erythrocyte aggregability (EA) by *in vitro* effect of insulin. In the present work, the *in vitro* effect of two physiological insulin's concentrations (120 and 480 pmol/l) upon human erythrocytes aggregability were studied. Washed RBCs were divided in three aliquots and incubated during 15 minutes: I0: PBS containing 10 mg/dl glucose without insulin (control); I120: PBS plus 120 pmol/l insulin; I480: PBS plus 480 pmol/l insulin. The supernatant was removed and RBCs were resuspended in Dextran 500 at 40% hematocrit for EA assays (s_0/n_0 and $2k_2n_0$). Statistical analysis: Student's *t* test for unpaired data.

	I ₀	I ₁₂₀	I ₄₈₀
$2k_2n_0$	0.60 ± 0.14	1.02 ± 0.17 ***	0.94 ± 0.28 ***
s_0/n_0	1.79 ± 0.07	1.85 ± 0.03 ***	1.85 ± 0.04 ***

***: $p < 0.001$

Our results showed a significant increased in two EA parameters at both insulin concentrations. Dextran 500 was used as aggregant agent, therefore the observed results might be due to changing erythrocytes intrinsic factors. Probably, the insulin's interaction with the membrane components increased erythrocyte aggregability owing to an decreased red blood cells deformability.

64. *IN VITRO* EFFECT OF INSULIN ON RED BLOOD CELL RHEOLOGICAL BEHAVIOUR IN b RAT STRAIN

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Previous studies in our laboratory in β (hypertriglyceridemic obesity and diabetes model) and b (eumetabolic) rats strains have shown increase in erythrocyte aggregability and decrease in erythrocyte deformability by *in vitro* effect of insulin. In the present work, the *in vitro* effect of two physiological insulin concentrations (120 and 480 pmol/l) upon erythrocyte deformability and osmotic fragility of b rats strains aged 200 days were studied ($N=18$). Washed RBCs were divided in three aliquots and incubated during 15 minutes: I0: PBS containing 10 mg/dl glucose without insulin (control); I120: PBS plus 120 pmol/l insulin; I480: PBS plus 480 pmol/l insulin. The supernatant removed and RBCs resuspended in: a) saline for erythrocyte deformability estimation (T_k), and b) 0 to 0,145 M NaCl for the study of osmotic fragility. Statistical analysis: Student's *t* test for unpaired data. We were not observed a significant variation in all parameters studied from I120 and I480 aliquot (regarding control). These results suggest that insulin did not produce changes neither in erythrocyte deformability nor in the erythrocyte membrane associated with osmotic fragility. Since the osmotic behaviour of RBCs is determined by haemoglobin concentration, the area and volume, the findings indicate that the insulin-red blood cell interaction does not alter any of these factors. Perhaps, increase in erythrocyte aggregability that we observed in studies previous with rats RBCs might be related with intrinsic factors from erythrocyte different of the erythrocyte deformability.

65. OSMOTIC FRAGILITY AND HUMAN RED CELLS SHAPE IN PRESENCE OF ALUMINIUM (Al³⁺): *IN VITRO* STUDY

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The object was to study the *in vitro* effect of aluminium upon the osmotic response and shape of human red cells, taking into account its possible introduction in the erythrocyte membrane. Washed red blood cells from donors without (control) and with Al³⁺ (1 y 10 µM) were used. Red blood cell resistance to osmotic haemolysis was determined photometrically and one parameter was obtained (x₅₀: NaCl concentration for 50% haemolysis). The shape of red cells was quantified through a morphological index (MI) (according to Bessis). Statistical analysis: Wilcoxon test for paired groups, data are presented as median and range. Osmotic fragility: x₅₀ (mM): control: 68,47 (67,76-72,67); 1 µM: 78,37* (66,57-79,20); 10 µM: 70,61* (67,00-72,41) MI: control: -0,65; 1 µM: -1,7(-2,24/-0,60); 10 µM: -2,43* (-3,06/-1,88). * p<0,01. The presence of stomatocytes can be attributed to a shrinkage of the membrane external monolayer due the interaction between the Al³⁺ and polar heads phospholipid of the external hemilayer. The bifasic response to osmotic stress guarantees the theory about that little ions disorganise the lipidic bilayer but to have higher number the present ions, these are joined new polars groups favouring a cooperative rearrangement of lipidic molecules.

66. SMOKING HABIT IN FIRST YEAR STUDENTS OF THE SCHOOL OF MEDICINE

Palmisano E, Macagno M, Barchetta Y, González J, Luraschi D, Rossi E, Cena P, Acosta A, Giorgetti M, Vargas D, Forla M, Rabasa F, Almiron M, Varela E, Bellocchio V, Gumpel R, Spina G, Padros M, Cucurullo M.
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Smoking is a premature and avoidable cause of death. Objective. To assess smoking prevalence and smoker profile in students (both sexes) (School of Medicine-UNR 2004). Transversal descriptive study. Participants were invited to fill, voluntarily and anonymously, a questionnaire. Definitions: Smoker (S): Daily consumption of at least one cigarette. Ex-smoker (ExS): complete habit cessation on the 6 last months, and non-smoker (N-S). Results (%). Participants: n=533, 61(323) women (W) and 39 (210) men (M). Smoking prevalence. W: S 24; ExS 14, Non-S 62; M: S 28, ExS 12, Non-S 60. Habit Profile. W I. Age of onset. W 16±3yr; M 16±2; II. Cigarettes/day cut-offs: 1. <5, 2. 6-10, 3. 11-20, 4. >20. W. 43; 37; 13; 7. M. 42; 40; 9; 9. III. Tobacco type: 1. blonde, 2. black, 3. both, 4. others. W. 90; 3; 1; 6. M. 92; 3; 5; 0. IV. Early motivation: 1. family/friends influence, 2. commercials, 3. others (o), 4. several (s). W. 5; 4; 76; 15. M. 12; 3; 66; 19. V. Current motivation: 1. entertainment/relax, 2. pleasure, 3. both. W. 29; 53; 18. M. 37; 44; 19. No sex difference (χ²). ExS profile. I. Motivation. 1. family influence (FI), 2. Medical council (MC), 3. o, 4. s. W 20; 17; 41; 22. M 24; 8; 52; 16. No sex difference. Non-S profile. Motivation. 1. FI, 2. MC, 3. o, 4. s. W 20; 11; 49; 20. M. 17; 10; 50; 23. No sex difference. Conclusions. 1. High prevalence of smoking habit. 2. Age of onset: adolescence. 3. Similar S, ExS and Non-S profiles between sexes. 4. It is important to develop learning-teaching strategies in the current curricula to fight this extended habit.

67. PHYSICAL ACTIVITY IN STUDENTS OF FIRST YEAR OF THE SCHOOL OF MEDICINE

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Physical activity (PA) is of the utmost importance in cardiovascular disease prevention. To attain this goal 30 min/daily four times weekly is recommended. Objective: to assess PA in 1st year students of the School of Medicine, UNR, 2004 (both sexes). Methods: Transversal descriptive study. The participants were invited to fill, voluntarily and anonymously, a questionnaire. The following items were registered: 1. PA; 2. PA category: aerobic (a), anaerobic (an), both (a-an); 3. Daily schedule (min/day); 4. Weekly schedule (4 times/wk). Data were stored in Microsoft Excel spreadsheet and processed with a statistical commercial package (SPSS 10). Results. Participants: n=533, 61% (323) women (W) and 39% (210) men (M). 1. PA (%): W 51; M 71 (χ²: 13.81, df: 1, p=0.0000). 2. PA category (%). W: a 65, an 7, a-an 29. M: a 53, an 10% and a-an 37. 3. Daily schedule (%). W 81 more than 30 min (+30): 6, less than 30 min (-30) 19. M: 82 (+30) and 18 (-30) (χ²: n.s.). Weekly schedule (%). W 54 more than 4 times/wk (+4) and 46 less than 4 times/wk (-4). M 45 (+4) and 55(-4), (χ²: n.s.). Conclusions. According to our results: 1) it is noticeable the high percentage of students engaged in aerobic physical activity for more than 30 min/day. 2) Women fulfil the week requirement while men do not. Nevertheless, physical activity is significantly higher in the latter group.

68. CANDESARTAN AS GASTRIC CYTOPROTECTOR AS EXPOSED TO ETHANOL AND STRESS. COMPARISONS WITH LOSARTAN

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It has been shown that Angiotensin II AT1 receptor and ACE antagonists drugs, like Telmisartan, attack the gastric mucosa when exposed to 20% and 96% ethanol and gastric stress. Candesartan (Ca) is an Angiotensin II receptor (type AT1) antagonist, which increases gastric blood flow, reduces adrenalin and TNFα and significantly reduces gastric erosions in hypertense rats and in stress. The purpose of this paper was to study the gastric cytoprotective role of Ca versus Losartan (Lo), when exposed to ethanol and stress. The following experiments have been carried out in randomized groups of Wistar female rats (n=7 per group) of 200 g, with 24-hour fasting, except from ad-libitum water and avoiding coprophagy: I.a) 1ml of 96% ethanol (Et) by orogastric bolus, with 20 min wait (witness). b) Ca 1-10 mg/kg Sc 2 h, then Et and c) Lo. 25-50 mg/kg Sc 2 h, then Et. II a) Immobilization (I) and water immersion stress at 18° C, 6 h (witness). b) Ca 1-10 mg/kg Sc 2 h, then I. c) Lo 25-50 mg/kg Sc, then I. Later, the rats were killed by ether overdose after laparotomy, total gastrectomy, gastric opening through greater curvature, quantification of necrotic area % or gastric macroscopic erosion by computerized image analysis, followed by histology (HE and MPO). Results: Percentaje of macroscopic gastric damaged area: I, a. Et 35 ± 5, Ca - Et. 1.0 ± 0.1 (< 0.001); Lo. Et. 70 ± 7 (< 0.05). E. a) witness: 75 ± 5%, b) Ca- I. 30 ± 4 (< 0.01) and Lo-I 90 ± 4 (< 0.05). MPO in I: a) witness: 350 ± 30 mg/protein Ca. I. 75 ± 7 (< 0.001) and Lo-I 750 ± 50 (< 0.01). Ca caused significant gastric cytoprotection compared with Losartan, which worsened ethanol and stress-caused damage.

69. INHIBITION SIMULTANEOUS OF COX - 2 AND COX - 3 IN THE SATOH MODEL, WITHOUT ANTRAL GASTRIC ULCER AND MULTIPLE INTESTINAL EROSIONS

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The objective was to study if the simultaneous inhibition of ciclooxigenasa 2 (COX-2) and ciclooxigenasa 3 (COX-3) in the model with solid food (Sato) cause ulcers in the gastric antrum and erosive injuries in thin intestine. To random groups of Wistar rats (n = 7 c/group) avoiding coprografia, the following experiment was made: Group 1- fast of 24 hs, solid food Pellets de Cargill during 2 hs, subcutaneous injection (SC) 2ml of physiological solution and solid food during 24 hs, without water. 2- fast 24 hs and solid food 2hs. Parecoxib (inhibiting selective COX-2) 50 mg/Kg/weight SC, solid food 24hs. 3- fast 24hs, solid food 2 hs. Paracetamol (inhibiting selective COX-3) 500mg/Kg/weight intraperitoneal, solid food 24 hs. 4- fast 24 hs, solid food 2 hs. Simultaneous Parecoxib with Paracetamol, solid food 24 hs. The rats were sacrificed by ether overdose. Laparotomy, gastrectomy, enterectomy, were carried out tabulating the percentage (%) of the gastric macroscopic lesional area and intestinal erosive area (mm²), and cuts were obtained for histological studies (H-E). The results were evaluated statistically with the "t" of student and ANOVA. Results: The lesional area in the gastric antrum gave: 1- 0% 2- 0% (n.s.) 3- 0% (n.s.) 4- 0% (n.s.). The erosive area in thin intestine gave: 1- 0 mm² 2- 0 mm² (n.s.) 3- 0 mm² (n.s.) 4- 230 ± 20 mm² (P<0.001). It was concluded that inhibition COX-2 and COX-3 in the model of Sato did not cause antral gastric ulcers and yes marked intestinal erosive injuries.

70. RESPIRATORY BURST (R.B) FROM PMN AND MN CELLS RELATIONSHIP WITH TNFALFA (TNF α) LEVELS IN TUBERCULOSIS (TB) PATIENTS TREATED WITH THREE DOSES OF *M. vaccae*

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In previous studies *M. vaccae* (1 dose) immunostimulatory effect was demonstrated. The effect of 3 doses of *M. vaccae* on RB of *M. tuberculosis* stimulated CMN and PMN from TB patients and its relationship with TNF α levels was evaluated. 12 patients TB (HIV-) of both sexes and medium age of 36±13.5 years, DS \were studied. On day 0 patients received the 1st dose of *M. vaccae* and DOTS (conventional therapy) and a sample of blood was extracted i.v, for the immunological studies. In the two subsequent months they received the same treatment. During the 3 following months, clinical, x-ray, bacteriological and immunological studies were done A control group received DOTS plus Placebo (n:12). CMN and PMN were separated by Ficoll- Triyoson and cultured in RPMI 1640 with and without heat inactivated H37Rv. R.B data were obtained by Flow Citometry, and expressed as R= Basal Fluorescence / Stimulated Fluorescence. TNF α was evaluated in s.c (R&D).

M. vaccae therapy increased significantly basal and stimulated RB on CMN and PMN (p<0.01). Therapy also increased TNF α levels which were correlated with RB. These data suggest an increasing immunostimulatory effect of three doses of *M. vaccae*.

71. STUDY OF THE ERYTHROCYTE MEMBRANE FLUIDITY IN THE SCLERODEMA

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The hypothesis that the free radicals generated in the systemic scleroderma, with the subsequent lipid peroxidation and the resultant damage in the tissue, have given place to the idea that the injury mediated by free radicals could play an important role in the patogenia of the disease. The free radicals can alter the physics and chemical properties of the cellular membranes modifying the distribution and packing of their lipids. The objective of this work was the membrane deformability and fluidity (MF) determination in 15 patients with scleroderma and in 15 controls by the study of the correlation between these values. The deformability is measured through its inverse, the rigidity index (RI), which relates the filter time of the red blood cells with the filter time of the suspension media. The MF is measured by the spectroscopy method with the fluorescence polarization, using TMA-DPH (Trimethylammonium-Diphenylhexatriene) as marker and calculating the anisotropy coefficient (r), which relates inversely with the MF. The main value of r obtained from the patients was significantly higher than the normal ones (0.2078±0.0146) vs 0.1940±0.0127; p<0.05); the same as the RI (15.1±4.36 vs 7.35±1.57; p<<0.01). Besides it was found that RI correlates positively and significantly with r (correlation coefficient: 0.76; p<0.01), which demonstrate that the diminishing in the lipid fluidity of the membrane can be one of the causes of the lose in erythrocyte deformability and contribute in this way to the damned fluid in the microcirculation.

72. STUDY OF RED CELL DEFORMABILITY AND OSMOTIC FRAGILITY IN SCLERODERMA

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Systemic scleroderma is a disease characterized by vascular damage in the microcirculation, where the deformability of red blood cells (RBC) is an essential condition for the blood flow in operation properly. The aim of this work was to determine the RBC deformability and their osmotic fragility in 15 patients with scleroderma. The values obtained were then compared to those of 17 healthy donors. Deformability is measured through its inverse, the rigidity index (RI), which relates the RBC filtration times to those of the suspension medium. The hemolysis curve is drawn according to the hemolysis data obtained by the equation: $H = 1 / e^{\beta(x - x_{50})} + 1$. Where H is the fraction of hemolysed cells, x is the ClNa molar concentration, x_{50} the ClNa concentration (in moles/l) wich causes a 50% hemolysis and represents the fragility of the largest number of cells. β is the measure of the breadth of the erythrocyte fragility distribution which shows the population's degree of homogeneity. The results showed that the patients' RI (15,10±3,45 vs 7,35±1,58; p<0,01) as well as the x_{50} (0,414±0,020 vs 0,393±0,004 g/dl de NaCl; p<0,05), were significantly greater than those of the control groups. However, no significant difference in β values was found. Moreover, a remarkably positive correlation (r= 0,49; p<0,05) was found between RI and x_{50} values, which shows that the RBC of patients are less resistant to the osmotic lysis since they are less flexible.

73.

ERYTHROCYTE AGGREGATION IN METABOLIC SYNDROME

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Metabolic syndrome (MS) comprises a group of risk factors that predisposes the person who suffers it to develop diabetes and diverse cardiovascular problems. Five variables are taken into account in its definition: abdominal obesity, high triglyceridemia, low HDL-cholesterol levels, high glycemia and high blood pressure. It is commonly considered that a person suffers from SM when three or more values surpass the established thresholds. The aim of the present paper was to determine, in blood samples collected from 13 MS patients, erythrocyte aggregability (EA), plasma viscosity (η_p) and plasma fibrinogen (F). Determinations were repeated in 17 normal controls of similar age. EA was estimated by measuring the variation, through time, of light transmitted through a whole blood sample; thus determining $2k_2n_0$ which estimates the initial velocity of the process. η_p was measured with a cone-plate rotational viscometer and F, through gravimetric method. Results showed that MS patients presented $2k_2n_0$ values significantly higher than controls ($1,7 \pm 0,37$ vs $0,58 \pm 0,19$; $p < 0,01$). η_p also resulted higher ($1,59 \pm 0,05$ vs $1,50 \pm 0,04$; $p < 0,05$), as well as F ($427,36 \pm 62,93$ vs $299 \pm 35,58$; $p < 0,001$). Besides, a significantly positive correlation has been found ($r = 0,55$; $p < 0,05$) between η_p and F, between F and $2k_2n_0$ ($r = 0,56$; $p < 0,05$) and between η_p and $2k_2n_0$ ($r = 0,79$; $p < 0,01$). These results are evidence for the fact that, in MS patients, the plasma fibrinogen contributes to increase plasma viscosity, and they both have influence on the increased aggregation; this could partly explain the high cardiovascular risk present in metabolic syndrome.

74.

ANGIOTENSIN II RECEPTOR ANTAGONISTS IN THE GASTROINTESTINAL PROTECTION IN THE PRESENCE OF INJURIES OF NON STEROIDAL ANTI-INFLAMMATORIES

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It is tested that the Angiotensin II AT1 receptor antagonists, like Candesartan (CA) provoke an increase of gastrointestinal blood flow, as well as a decrease of neutrophils, from TNF α , from P-Selectin and from ICAM-1. The objective was to study if different Angiotensin II AT1 receptor antagonists, like CA, Losartán (LO) and Valsartán (VA), would or would not have anti-inflammatory and anti ulceration effects against the gastrointestinal mucous aggression from non steroidal antiinflammatories. At different groups of Wistar rats randomly chosen ($n = 7$ each group), 200g, 24 hours (h) fasting, ad-libitum water, avoiding coprophagy: I a: Satoh's Method. 24 h fasting, solid food (Pellets Cargill) during 2 h, then 1ml physiologic subcutaneous (SC), with 24 h wait; b: 24 h fasting, solid food, Indometacina (Indo), 50 mg/kg SC, 24 h Witness (W); c: 24 h fasting, solid food, Indo as wells as CA 10mg/Kg SC; d: 24 hs fasting, solid food, Indo with LO 50mg/Kg SC 24 hs; e: 24 h fasting, solid food, Indo with VA 40mg/Kg SC 24 hs. II a: 24 h fasting, physiologic 1ml SC, 24 h; b: Indo 50mg/Kg orogastric (OG), 24 h (W); c: Indo as well as CA, 24 h; d: Indo with LO, 24 h; e: Indo with VA, 24 h. Later, the rats were killed by ether overdose, after laparotomy and total enterectomy. It was tabulated the percentage (%) of the ulcerous antral gastric area as well as the wounded gastric area and also the eroded intestinal area. It was calculated the "T" student and ANOVA. Results: I: The percentage of the ulcerous antral gastric area was: a: 0%; b: 30%; c: 0% ($< 0,001$); d: 0% ($< 0,001$); e: 0% ($< 0,001$). II: The percentage of the wounded gastric area was: a: 0; b: 65; c: 5 ($< 0,001$); d: 15 ($< 0,001$); e: 35 ($< 0,05$); the intestinal area in mm^2 was measured as follows: a: 0%; b: 185%; c: 35% ($< 0,01$); d: 55% ($< 0,01$); e: 101% ($< 0,05$). Microscopic studies confirmed macroscopic findings. Conclusions: Angiotensin II AT1 antagonists receptors, CA, LO and VA impeded the formation of antral gastric ulcers induced by SC Indo and also reduced the extension of gastric and intestinal lesions caused by Oral Indo.

75.

DENTAL HEALTH IN THREE ROSARIO CITY AREAS

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The actual economical situation produced the moving of patients looking for odontological attention to public system where preventive programs are only available for youngsters than 14 years. Description of Rosario City buccal health situation in 15-45 years old subjects, was the objective of the study. Studied variables were teeth health, first extraction age, elapsed time from last dental consult (ETDC). Private and public dental health services, one from the down-town area and one from a poor peripheral area, were incorporated. 517 persons, 65% women and 35% men, with a mean age of 27 years, were studied; 81% from the public and 19% from the private services.

Mean $\pm$ SD	Public down-town	Public Peripheral area	Private
Carious teeth	4,1 $\pm$ 2,9	4,1 $\pm$ 2,8	1,7 $\pm$ 2,0
Lasted teeth	7,0 $\pm$ 5,4	6,6 $\pm$ 5,5	5,4 $\pm$ 5,2
Obtured teeth	1,2 $\pm$ 2,0	1,5 $\pm$ 2,4	4,4 $\pm$ 4,2
First extracted age	17,6 $\pm$ 6,3	18,7 $\pm$ 6,2	19,7 $\pm$ 7,7
ETDC	30,0 $\pm$ 37,8	35,2 $\pm$ 45,0	22,3 $\pm$ 30,1

Major dental health problems in the last consult were obturations in the private (47%) and extractions in the public service (52%). While in the public service a higher number of lost teeth was found, in the private service prevailed the obtured pieces. First extraction age was higher in the private than in the public service. Elapsed time from last dental consult was lower in the private service, where the most frequent were obturations while in the public were extractions.

76.

ASYMPTOTIC BODY WEIGHT AND MATURING RATE IN EXPERIMENTAL HYBRIDS OF RANGE BROILERS

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To characterize the body weight growth pattern of three hybrids of range broilers with Cornish Red paternal genotype and different maternal genotype [Barred Plymouth Rock (Casilda I), Rhode Island Red (Casilda II), Estirpe E (Nuevo Campero)] longitudinal body weight-chronological age data, recorded at weekly intervals from hatching to slaughter, were adjusted by a nonlinear regression procedure using the Gompertz growth equation. The genotype effect on asymptotic body weight and maturing rate was assessed by means of a one way analysis of variance. Hybrids did not differ ($P > 0.05$) in their mean asymptotic weight (Mean $\pm$ SEM - Casilda I: 4960 ± 147 ; Casilda II: 4876 ± 181 ; Nuevo Campero: 5123 ± 158). Casilda I and II, obtained from non conventional maternal genotypes, showed a lower maturing rate than Nuevo Campero ($p < 0.05$) without significant differences between them (0.1828 ± 0.0034 , 0.1911 ± 0.0043 , 0.2162 ± 0.0055 , respectively). It is concluded that, in relation to body growth pattern, the alternative hybrids proposed in this study (Casilda I and Casilda II) represent viable options as range broiler genotypes because, even though they reached the proposed objective slaughter weight (males: 2500g, females: 2200g) at a later age than the control hybrid Nuevo Campero (84 versus 70 days), it was achieved between the limits established by the official protocol for Campero broilers.

77.

PRE AND POST-INFLECTION GROWTH IN MICE BELONGING TO A SEGREGATING F₂ POPULATIONZucchetti A¹, Silva PS¹, Font MT^{1,2}, Di Masso RJ^{1,2}.¹Inst. de Genética Experimental, Facultad de Ciencias Médicas.²CIC-UNR.

Although mice growth is a continuous process, the sigmoid pattern displayed when body weight is plotted against time can be divided into a self-accelerating phase with an increasing slope and a self-inhibiting phase with a decreasing slope. With the aim of studying the relationship between early and late growth the post-inflection asymptotic exponential growth rate was estimated by a non linear regression technique for 46 mice (23 per sex) belonging to each tail of the frequency distribution of the pre-inflection exponential growth rate also estimated by regression for 400 males and 400 females belonging to a F₂ segregating population obtained by crossing two lines of mice with different mature body weight. Animals of the same sex with low (males: 0.0664 ± 0.00072 ; females: 0.0624 ± 0.00037) and high (males: 0.0836 ± 0.00031 ; females: 0.0768 ± 0.00025) pre-inflection exponential growth rate (mean $\pm$ SEM) did not differ significantly ($P > 0.05$) in their post-inflection mean growth rate (low males: 0.0372 ± 0.00297 ; low females: 0.0267 ± 0.001590 and (high males: 0.0356 ± 0.00213 ; high females: 0.0246 ± 0.00248). These results are in accordance with the detection of different quantitative trait loci (QTLs) for early and late murine growth as well as with the evidence available that, in rodents, early and late growth, both of them characterized in terms of body weight behavior, have a different cellular basis and are regulated, in part, by different genetic and physiological systems.

78.

TIBIAL MORPHOMETRY AND ALKALINE PHOSPHATASE (ALPo) ACTIVITY IN MICE DIFFERING IN THE PREINFLECTION EXPONENTIAL GROWTH RATE (PiEGR)Gialdini C¹, Silva P¹, Hinrichsen L^{1,2}, Font MT^{1,2}, Di Masso RJ^{1,2}¹Instituto de Genética Experimental, Fac. de Ciencias Médicas, U.N.R.; ²Consejo de Investigaciones, CIC-UNR.

The association between tibial morphometric traits and osteogenic potential estimated by ALPo, were studied in 150 days old mice differing in PiEGR. Mice belonged to a segregating F₂ population obtained by crossing two lines divergently selected for body conformation (CBI/C, high weight - short skeleton, and CBI/L, low weight - long skeleton). Twenty-three animals of each sex, of each tail of the frequency distribution of PiEGR (lowest rate – highest rate) were used. PiEGR was estimated by fitting the data body weight – age with an exponential equation. Body weight (g), tibial dry weight (mg) and length (mm), and ALPo activity (mU) from tibial extracts were determined. Significance of the differences between tails – within sex or between sexes – within tail were analyzed with Student's "t" test. There were no differences between tails - within sex in the variables studied. As expected, 150 days old females showed lower body weights than males ($p < 0.01$); interestingly, males and females did not differ ($p > 0.05$) in tibial weight and length, indicating that acquisition of adult bone mass is a process independent of PiEGR and influenced by sex. A significant effect ($p < 0.01$) of sex was observed in ALPo, females showing a higher mean value than males. Since a certain level of ALPo activity seems to be necessary for the initiation of bone mineralization, this result suggests that females at this age, as opposed to males, are actively engaged in bone remodeling.

79.

ADIPOSIITY TRAITS AT MATURITY IN MICE WITH DIFFERENT PRE-INFLECTION EXPONENTIAL GROWTH RATEFidalgo P¹, Silva PS¹, Font MT^{1,2}, Di Masso RJ^{1,2}.¹Instituto de Genética Experimental, Facultad de Ciencias Médicas.²CIC-UNR.

Mice models have been instrumental in the understanding of the genetic basis of many diseases and many of them have been identified and are being used as animal models of obesity. With the aim of studying the relationship between early growth and adiposity levels in the adulthood, body weight (BW), body length (BL), absolute and relative perigonadal fat pad weight and body mass index ($BMI = BW/BL^2$) were recorded in 46 mice (23 per sex) belonging to each tail of the frequency distribution of the exponential growth rate estimated for 400 males and 400 females belonging to a F₂ segregating population obtained by crossing two lines of mice with a mean difference of 120% in adult percent body fat. Animals of the same sex with low (males: 0.0664 ± 0.00072 ; females: 0.0624 ± 0.00037) and high (males: 0.0836 ± 0.00031 ; females: 0.0768 ± 0.00025) preinflection exponential growth rate (mean $\pm$ SEM) did not differ significantly ($P > 0.05$) in body weight and adiposity traits measured at 150 days of age. These results are in accordance with the complexity of body weight definition as it includes lean and fat tissue, as well as with the detection of independent quantitative trait loci (QTLs) for fat deposition and body weight. They also indicate that fat deposition, a process that mainly occurs during the post-inflection phase of the growth curve, shows a genetic determination independent of early growth when protein accretion predominates.

80.

BODY WEIGHT AND BODY CONFORMATION IN EXPERIMENTAL HYBRIDS OF RANGE BROILERSLibrera JE¹, Canet ZE¹, Antruejo AE², Di Masso RJ^{1,3,4}, Font MT^{3,4}, Dottavio AM^{1,4}.Cátedras de ¹Genética y ²Producción Animal I, Fac. Cs. Veterinarias. ³Instituto de Genética Experimental, Fac. Cs. Médicas, ⁴CIC-UNR.

Genetic selection based on economic traits such as growth rate has resulted in gross changes in commercial meat poultry. Along with these improvements in growth and feed efficiency a concomitant increase in skeletal abnormalities was seen. With the aim of studying the interrelationship between body weight and meat type conformation three hybrids of range broilers with Cornish Red paternal genotype and different maternal genotype [Barred Plymouth Rock (Casilda I), Rhode Island Red (Casilda II), Estirpe E (Nuevo Campero)] were compared in terms of: (1) body weight at three ages (42, 56, 70 days) and (2) a conformational index calculated from six linear measures by means of a principal component analysis. In both sexes the index (first principal component) explained an 80% of the phenotypic variance and it was negatively correlated with each linear trait ($>$ index value $<$ skeletal size) and with body weight. Casilda I and II were lighter and showed a higher index mean value than Nuevo Campero without significant differences between them. Comparisons at the same chronological age corroborated the direct relationship between body weight and body conformation predicted from the positive genetic correlation values reported in the literature. The results did not confirm previous experimental evidence concerning to the at least partially independent genetic determination of both traits.

81.

TIBIAL AND GASTROCNEMIUS MUSCLE WEIGHTS IN MICE SELECTED FOR BODY CONFORMATION

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According to Hoopers's hypothesis the greater force exerted by large muscles increases the mass of the bones to which they are attached. To characterize the genetic quantitative control of this muscle-bone relationship, tibial and gastrocnemius weight were recorded in adult male mice (150 days old) belonging to two lines with agonistic selection for body conformation (CBI-: low body weight-short tail; CBI+: high body weight-long tail), two other lines with antagonistic selection for the same trait (CBI/L: low body weight-long tail; CBI/C: high body weight-short tail), and the reciprocal hybrids within selective procedure. Additivity for body weight was evident for both, agonistic and antagonistic selected mice. Hooper's proposal was corroborated when selected lines were compared within selective procedure (a higher muscle weight was accompanied by a higher tibial weight). Hybrids showed a dominant deviation towards high muscle weight values and additivity for tibial weight in agonistic selected mice while antagonistic selected animals shared the muscle weight behaviour (dominant deviation) but showed an heterotic effect (overdominance) on tibial weight. These results suggest the presence of at least partially independent genetic control mechanisms acting on muscle mass (dominant deviation) and bone mass (additivity-overdominance) regulation in each set of lines. These differences could be ascribed to the exploitation of different sources of genetic variance for each trait in each selective procedure.

82.

SIEVE TEST OF BPA IN MILCH BOVINE FROM TWO DEPARTMENTS OF SANTA FE PROVINCE

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The objective of this work was to know the number of reactionary agents to the BPA technique (Buffered Plate Antigen) in premises of small producers in the Departments of Rosario and San Lorenzo in Santa Fe.

The small producers (1-20 animals per premise) are not included in the National Plan for Control and Eradication against that illness. Blood tests were taken from 261 animals belonging to 61 premises. The serums were analysed in order to determine the presence of antibodies against *Brucella abortus*, for the diagnosis of brucellosis and this was compared with the data obtained –through the same technique – from premises which are free and under guarantee from the official records of SENASA. No significant differences were found in the positive reactionary agents between the groups under guarantee and those not included in the plan (parametric test χ^2). Although the sanitary condition of the animals from the small premises is not different from that observed in the premises under guarantee, the support that can be offered to that sort of producers will contribute to improve their life quality and to the Public Health of society in general.

83.

ARTERIAL IRRIGATION OF THE STOMACH OF THE DOMESTIC CAT *Felis domesticus*

Smacchia CA, Montero LE.

Cátedra de Anatomía Descriptiva y Comparada I° Parte. Fac. Cs. Veterinarias -Universidad Nacional de Rosario.

A greater part of the classic anatomy bibliography describes arteries of the cat in an uncompleted way. Feline species is not specifically mentioned. The increasing interest that this species has in our society has prompted us to develop a working plan aimed to know in detail the feline angiology. Thirteen male and female adult felines were used. Firstly a thoracotomy was performed, then the descending aorta artery was localized, it was channelized and saline flushed and colored latex was injected. The specimen was refrigerated at 2° C during 72 hours, then the abdominal cavity was opened and arteries arriving at the stomach were dissected. It was observed that the stomach receives the following irrigation: From linealis artery: 1) a gastric branch to cardias (inconstant); 2) first gastric-pancreatic branch; 3) first spleno-gastric branch (branch of the dorsal pole), this branch gives off 2-4 special branches to the left third of the visceral surface of the stomach; 4) second pancreatic-gastric branch; 5) second spleno-gastric branch (branch of the ventral lobe); 6) gastric branches of the left – gastroepiploic artery. From left gastric artery: 1) cardias branch, 2) short gastric branches. From hepatic artery: 1) cardias branches (inconstant); 2) right gastric artery, this branch gives off short gastric branches and then anastomoses with left gastric artery. It was concluded that the irrigation of the domestic cat in general coincides with the dog arterial pattern with the exception of the presence of special branches from the dorsal pole branch of linealis artery whose presence must be taken into account by the surgeon working in the area to not compromise the irrigation.

84.

ANATOMY OF THE ARTERIAL IRRIGATION OF THE LIVER OF THE DOMESTIC CAT *Felis domesticus*

Smacchia CA, Montero LE.

Cátedra de Anatomía Descriptiva y Comparada I° Parte. Fac. Cs. Veterinarias, Universidad Nacional de Rosario.

This study is a part of a working plan carried out by this Chair aimed to know in detail the feline angiology. Thirteen male and female adult felines were used. Firstly, a thoracotomy was performed, then the descending aorta artery was localized, it was channelized and saline flushed, then colored latex was injected adding a radio-opaque substance, then the abdominal cavity was opened, the celiac trunk was dissected and the hepatic artery was followed up to its entrance in the porta hepatis., then the liver was separated and photographed to determine the divisions of the branches in the parenchyma. Two variants were found in the hepatic artery origin according to two division patterns of the celiac trunk, 1) the celiac trunk was trifurcated in splenic, hepatic and left gastric arteries in a mean distance of 6 mm; 2) the celiac trunk gave off a hepatic artery and continued with a short common trunk to the linealis and left gastric arteries, being the mean distance to the first division of 6 mm and the second of 2,5 mm. On the visceral surface of the liver the artery gives off three branches: 1) a lateral right branch to the lateral right lobe and portion of cauded lobe; 2) medial right branch (double 23% of the cases) irrigating medial right lobe, portion of cauded lobe and portion of medial left lobe; 3) the left branch divides into medial left branch irrigating the medial left lobe and square lobe, giving off the cystic artery and lateral left branch to the lateral left lobe. After giving off the three branches the hepatic artery divides into right gastric artery and gastroduodenal artery. It was concluded that the general scheme of mammals is maintained.

85.

FARM SIZE AND BREEDING INDEXES IN CHINCHILLA MICRO FUR INDUSTRYNistal A¹, Marconi A¹, Di Masso R.J.^{2,3}, Catalani G¹.¹Cát. Fisiología, ²Cát. Genética, Fac. Ciencias Veterinarias, ³CIC-UNR, Universidad Nacional de Rosario.

High productive efficiency in complete cycle production systems requires maximization of offspring per litters. In *Chinchilla lanigera* production efficiency is reflected on quality and number of furs. Consequently, maximization of litters per female and kits per litter is required, as well as mortality minimization and growth optimization to attain a proper size at euthanasia (approx. 11 mos). Objective: to correlate breeding indexes with farm size (18), close to Fac. Cs. Veterinarias, were assessed and classified as follows: Familiar ranch (F): up to 100 breeding females, 2 family members with employees, less than 3 hr/day, and Intermediate ranch (I: 100-250 breeding females, whole family, plus temporary sign up employees, over 4hr/day. Information about number of births/mother/year (NB: 1; >1, to categorize each establishment, number offspring/mother/yr (Off/mother/yr: 1.5; >1.5), and number of weaning kits (WP)/mother/yr. Association was estimated performing the exact Fisher test. Values (median and range) were: NB [F 1.1 (0.8-2.1); I 1.0 (0.8-1.4)]; OffN [F 2.0 (0.6-2.4); I 1.8 (1.4-2.0)] and WK [F 1.8 (0.7-1.9); I 1.9 (0)]. There were no significant differences in breeding indexes in function of ranch size, which is low. Lack of association between ranch size and breeding could be due to the fact that each rancher set up an strategy in accordance with the provider, no matter if there is an increase in mothers number. These are preliminary results, therefore, the problem considering breeding aspects could be associated with the lack of scientific information about species and lack of specialized technical support.

86.

HEMORHEOLOGICAL MODIFICATIONS IN HORSES TREATED WITH A VEGETABLE EXTRACTCatalani G², Dottavio ME¹, Miglietta MC³.¹Cát. Física Biológica, Fac. Cs. Médicas; ²Cát. Fisiología, Fac. Cs. Veterinarias-Universidad Nacional de Rosario; ³Anatomía Veterinaria, Fac. Cs. Veterinarias- Univ. Nacional del Litoral.

Variable nasal haemorrhage induced by exercise (EIPH) occurred in training thoroughbred horses (TBH). The animals were treated with 30% hydroalcoholic solution of crude vegetable extract with 5% dry residue. *Portulaca sp.* foliar parenchyma with high Omega 3 (30,000 ppm.), and linolenic acid (4,000-80,000 ppm) content, and *Liliaceas* with high glucose and manose hydrocarbon jell were used. At treatment completion bleeding decrease and more rapid respiratory recover after exercise were observed. Objective: to study the rheological modifications produced by treatment in non-training healthy adult TBH with same management and feeding conditions for the whole group were studied. Fifty ml extract were administered orally in the morning each seven days. TBH pre- and post treatment whole blood samples (n=6) obtained by jugular venipuncture, with EDTA as anticoagulant, were analyzed. Erythrocyte deformability by means of erythrocyte rigidity index (RI), relating filtration time of red blood cells (RBC) suspension through a polycarbonate membrane (5µm diameter) to filtration time of medium suspension was assessed. Erythrocyte aggregation: registering light density variation in time in a whole blood sample. Curve mathematical analysis yielded two parameters: 1. size average aggregate estimation (Sz), and initial aggregation rate (R). Assays were performed at 40% haematocrit in autologous plasma. Statistics: Wilcoxon test for paired data. Results (median and range), show a significant post treatment R (p<0.05), whereas, Sz is not modified (n.s.). RI was decreased (p<0.01) at day 7 post-treatment. Conclusion: results on both rheological parameters studied in EIPH, point out a capillary improvement.

87.

NUMBER OF EMBRYOS PRODUCED BY HORMONAL INDUCTION IN WEIGHT-SELECTED MICEBernardi S¹, Brogliatti G², Oyarzabal M³.¹Histología II y Embriología Especial, Fac. Cs. Veterinarias. UNR.²Centro Genético Bovino EOLIA S.A. ³Producción Animal II, Fac. Cs. Veterinarias y CIUNR.

The weight selection at 49 days of age (P) and endobreeding performed in two pairs of lines of diverging selection (negative s and h; positive s' and h'), originated from a t population of CF1 mice caused differences in fertility-related characters. With the aim of testing if these differences were kept when ovulation was induced, the response of each line was compared with over-ovulation through the number of embryos (E), and was related to weight, selected character. Over-ovulation was achieved administering 5UI of CGe to each female, 48 hours after the administration of 5UI of CGh and were bred in a 1 ♂:1♀ proportion. Positive lines had a higher E number than negative ones in the treated group (E_{CT}) and in the non-treated group (E_{ST}). The same relation was seen in the non-treated group when E was related to P (E_{ST}/P). Induction suppressed the differences between positive and negative lines for E_{CT}/P, due to a greater relative increase in the number of embryos in negative lines. These results are in agreement with those obtained after analyzing the number of corpora lutea in control females whose over-ovulation was achieved with the use of gonadotrophins.

88.

ROLE OF HEPARIN IN THE CAPACITATION OF PORCINE SPERMDapino D¹, Marini P², Cabada M².¹Cát. Fisiología. Facultad de Cs. Veterinarias; ²División Biología del Desarrollo (IBR-CONICET) y Á. Biología. Facultad de Cs. Bioquímicas y Farmacéuticas. U.N.R.

Heparin has been extensively used *in vitro* to study the endogenous action of sulfated glycosaminoglycans (sGaG) in female oviduct. Its capacitating effect has been demonstrated in bovine, equine and human sperm. However, this action has not been reported in pigs. Objective: to determine the effect of heparin on pig sperm physiology. Assays: semen was collected from 8 fertile boars, centrifuged and resuspended in TALP (10⁷sperm/ml). The sample was divided in three aliquot with 0, 10 and 100 µg/ml of heparin (C0, C10 and C100) and incubated at 39°C, taking aliquots at 0, 90 and 120 min (T0, T90 and T120). Maturation state was assessed by chlortetracycline assay obtaining three fluorescence patterns: non-capacitated(A), capacitated(B) and reacted sperm(C). Viability was evaluated by eosin/nigrosin staining. Statistics: ANOVA. The rate of capacitated sperm was T0: 5,8 ± 1,8; 4,6 ± 1,8; 5,3 ± 2,5; T90: 27,7 ± 4,1; 40,6 ± 4,7; 44,0 ± 7,4 y T120: 33,5 ± 5,8; 43,5 ± 3,2; 44,3 ± 6,8 at C0, C10 y C100, respectively. There were no significant differences between capacitation values in the presence of heparin at 90 and 120 min of incubation with both concentrations used (P>0.01) and there were significant differences with respect to the control (P<0.01). Sperm vitality at the end of the experiment was ~70% in all samples, except at 100 µg/ml of heparin (~40%). In conclusion, heparin augments the *in vitro* capacitation of pig spermatozoa, with optimal value at 90 min of incubation and 10 µg/ml of heparin. The evidences of a rise in sGaG in the oviduct during estrous period in sows, and these *in vitro* results, suggest that heparin-like compounds play an important role in fertilization in porcine species.

89.

ANALYSIS OF THE SEROLOGICAL STATUS AGAINST *Neospora caninum* IN A DAIRY HERD

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Neosporosis is a parasitic disease caused by the intracellular protozoan *Neospora caninum*, it infects a great number of animals such as dogs (definitive hosts) and bovine, goats, sheep, equine and deer (intermediate hosts). Clinical signs in cattle include reproductive failure, infertility, embryonic resorption, abortion and vertical transmission of the infection. Antibodies titres against *N. caninum* fluctuate during the lactation and they are associated with the congenital transmission and the abortion. The aim of this study was to evaluate the serological status in a dairy herd and to analyze the results obtained with the relationship among the animals. Samples of serum from 82 adult milking cows Holando Argentino breed were obtained. The samples were tested for antibodies to *N. caninum* by indirect immunofluorescent in 1:200 dilution. Antibodies against *N. caninum* were detected in 28 serums (34%). In 15 positive cows, 5 (33%) were born from positive cows and 10 (66%) from negative cows. In 21 negative cows, 17 (80%) were born from negative cows and 4 (20%) were born of positive cows. In two positive cases, the mother and the grandmother were negative. The serologic prevalence of the herd is in the range according with Argentinean and foreigners authors. It is interesting the high percentage of positive cows (66%) that are daughters from negative mothers, this could indicate the posnatal infection posnatal or the possibility of false positive. In conclusion, the analysis of the seroestatus with the relationship and the age of the animals gives an important information on the prevalence of infection in the herd and the routes of transmission, congenital and posnatal infection.

90.

SURVIVAL OF EGGS OF *TOXOCARA CANIS* IN CONTROLLED ENVIRONMENT

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Toxocara canis (Werner, 1782 y Stiles 1905) is a nematode that in their adult stage inhabits the thin intestine of the dog. Each female of this parasite can eliminate 200.000 eggs daily. It is estimated that a dog with a parasitosis mild would eliminate daily with its dregs 1,4 million of eggs. With the objective of establishing the survival of eggs that cause infestation of *T. canis* over time, its was sown in sand faecal matter dog's of two years old, with a load of 4556 eggs by gram, adding fields of poliestireno expanded of 5 mm of diameter to favor the oxygenation. It was maintained at ambient laboratory temperature (9°C a 19°C) and weekly it was added water. It was used the flotation technique in order to recover the eggs, the technique of rapid artificial digestion in order to obtain larvae from mice. From the moment of the sow (February of 2003) was observed daily the evolution of the eggs until the larvae stage and from day 30 of the sowing and monthly were recovered eggs to evaluate their survival, using the optical microscope, the morfologic characteristic and movement of the larva within each egg. Throughout 18 months (February 2003 to August 2004) were recovered in average 5,2 eggs by sand gram and the percentage of viability oscillated between 91,20 and 100%. It was proven the larvae eggs recovered to 20 days and 8 months administering them by oral way to the mice have capacity to infest. There were found larvae in liver, lungs and muscles. Its was not observed a meaningful trend of the variable percentage of viability during a long period of the time ($p > 0,05$).

91.

MOLECULAR EPIDEMIOLOGY OF BOVINE MASTITIS CAUSED BY *Streptococcus spp*François S¹, Limansky A², Sutich E², Ebner G², Toresani P, Viale A²¹Cát. de Microbiología, Fac. de Cs. Veterinarias, Ruta 33 y Bv. Spangenberg, Casilda. ²Dpto. de Microbiología, Fac. de Cs. Bioq. y Farm. UNR. Suipacha 531, Rosario, Sta. Fe, Argentina.

Streptococci bovine mastitis is an udder infection mainly caused by *S. agalactiae* (EGB), *S. dysgalactiae* and *S. uberis*. The aims of the study were: i) to analyze the genetic variability between isolates of *Streptococcus spp.* responsible for bovine mastitis in different herds; and ii) to analyze specific strain dissemination and persistence. Four hundred and sixty-six samples of milk from cows with mastitis of herds from the South of Santa Fe were studied. They were cultured in agar blood and Todd-Hewitt culture. The catalase negative populations were identified with specific agglutination of the group and biochemical tests. The genotypic methodology employed was PCR with degenerated oligonucleotides. Seventy-eight isolates of *Streptococcus spp.* were recovered, of which 46 were *S. agalactiae*, 14 *S. dysgalactiae* and 18 *S. uberis*. The genomic analysis of the EGB population allowed for the observation of 17 genotypes. The dissemination of a particular genotype within each herd was detected. Moreover, the herd that was followed for a year, showed the persistence of a specific clone as well as resistance in one cow; denoting a high capacity of dissemination of the clone. Twelve strain were found in 14 isolates of *S. dysgalactiae* and sixteen in the 18 isolates of *S. uberis*. For these species the coexistence of different clones in the same herd was detected. The results suggest the radication of virulent strains in each herd in the case of the intra-mammary pathogen EGB, while *S. dysgalactiae* and *S. uberis* show a higher genetic variability, probably for being environmental agents.

92.

KINETICS OF ALFALFA HAY DEGRADATION IN RUMEN ENVIRONMENTS OF ALFALFA HAY AND PASTUREFigallo R¹, Smacchia A¹, Faienza H, Pidello A¹.Lab. Química Biológica, Fac. Cs. Veterinarias. ¹Consejo de Investigaciones- CIC -UNR. E-mail: rfigallo@fveter.unr.edu.ar

The aim of this work was to study the microbial activity in the rumen environments of ovine fed alfalfa hay (AH) and cattle pasturing alfalfa (PA). The microbial activity was studied through the kinetics of rumen dry matter degradation (RDMD) of AH. An AH sample was used. It was dried at 60°C, ground and sieved through a 2mm mesh. Three ovine (O) fed AH twice a day and three bovine (B) PA *ad libitum* and with 2.5kg of concentrate at 4 h and 16 h had rumen cannulae. Small ASTM 230 (pore size: 62 microns) nylon bags were incubated with 3g of DM. (17mgDM/cm²) during 0, 2, 4, 8, 12, 18, 24 and 48 h, in rumen for two periods (Meherz and Orskov, 1977). The data observed were adjusted to: % RDMD = $a + b(1 - e^{-ct})$; the fractions were: **a**: fast, **b**: slowly degradable, and **c**: degradation rate (%/h); and studied under ANDEVA and Tukey's Test ($P > 0.05$).

Rumen Environment	Incubation Time								Fraction		
	0	2	4	8	12	18	24	48	a	b	c
Media											
OAH	27	34	39	56	66	73a	74	76	23.6	53.3	0.1215
BPA	27	34	38	51	58	72b	71	75	24.7	51.6	0.0959
CV %											
OAH	3.4	3.8	3.8	6.6	4.3	1.2	2.0	1.0	2.2	2.58	10.2
BPA	3.4	4.7	11.3	7.7	11.8	0.3	1.8	0.6	3.4	4.5	22.9

Note: a, b, different letters in a column show differences ($P < 0.05$).

RDMD was similar in most of the incubation times. It was higher in HAO only 18 h after degradation. The degradable fractions and degradation rates showed the same pattern. The PAB rumen environment showed higher CV % between 2 and 12 h incubation and in the degradable fractions and degradation rate. These results suggest that the studied rumen environments through microbial degrading activity act in a similar way and, moreover, ovine rumen environment is less variable.

93. METHANE PRODUCTION FROM PRESERVED FORAGES AND PASTURES INCUBATED IN RUMEN *IN VITRO*

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The aims of this work were to compare methane production from preserved forages and pastures; to determine if gas production, CO₂ and CH₄ are related with C-oxidizable concentration; and to identify variables associated with methane concentration in gas produced by forages incubated in rumen. The forages were: Preserved: alfalfa and moha hays and corn silage (n:6) and Pastures: oats, bromus, and three alfalfa cuts (n:9). C-oxidizable was determined by Walkey and Black (WB) and Harris (HA) methods. Rumen degradation (DMRD) in sacco, fast (a) and slow (b) degradable fractions as well as degradation speed (c) were determined. *In vitro* incubations were performed in batch with 30 ml of ovine rumen fluid and 1% of sample (24 h, 39°C); gas production, CO₂ and CH₄ were measured by chromatography

Forages	Gas ⁽¹⁾		CO ₂ ⁽²⁾		CH ₄ ⁽²⁾		CH ₄ ⁽³⁾		CH ₄ ⁽⁴⁾	
	x	±	x	±	x	±	x	±	x	±
Preserved	52.2	10	17.6	1	7.2 a	0.5	18.3 a	2.8	111 a	25
Pastures	77.2	7	25.9	3	13.8 b	0.4	41.3 b	7.7	297 b	20

Note: ⁽¹⁾ (ml/gMS) ⁽²⁾ (mmol/l) ⁽³⁾ (l/kgDRMS_{24h}) ⁽⁴⁾ (l/cow/day). a,b; different letters in the same column show differences (p≤ 0.05).

Multiple regression analysis showed that CH₄(mmol/l) was related with CO₂(mmol/l) (r: 0.925; p< 0.00) and a fraction (r:0.597; p<0.018) with DMRD_{24h} (r: 0.79, p< 0.0004); C-oxidizable with WB only with DMRD_{24h} (r: -0.648; p< 0.008) and CO₂ (mmol/l) with gas produced (ml/24h) (r: 0.526; p< 0.04). The 90 % of CH₄ concentration (mmol/l) was accounted for by CO₂ (mmol/l) and by the a fraction; thus, CH₄ (mmol/l) can be estimated with the following equation: y: -4.57 (±1.54) + β₁*0.531(±0.060) + β₂* 0.127(±0.047); where y: CH₄ (mmol/l); β₁: CO₂ (mmol/l); β₂: a fraction. Results show that pastures are the most methanogenic, that C-oxidizable was not related with methane production but with DMRD. CH₄ concentration is mainly determined by CO₂ concentration and by fast-degrading DM fraction in rumen.

94. METHANE PRODUCTION IN RUMEN: COMPARISON BETWEEN FORAGES THROUGH THE COMPLEMENTARY USE OF *IN VITRO* AND *IN SACCO* METHODS

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The aim of this work was to compare methane produced by different forages by means of the complementary use of *in vitro* e *in sacco* methods. Oats (*Avena sativa*) (A), barley (*Bromus catharticus*) (B), alfalfa hay (*Medicago sativa*) (AH), moha hay (*Setaria italica*) (MH) and corn silage (*Zea mays*) (CS) were incubated in *in vitro* systems for 24 h; after incubation CO₂ and CH₄ production was measured with gas chromatography, and pH and redox potential were determined in the fluid fraction. Samples of those forages were incubated *in sacco* and degradable dry matter (DDM) was determined. CH₄ rumen production, estimated (l/day) for a cow fed with 10kg DM, was: 222.7b; 219.1b; 208.5ab; 195.5a and 194.3a for A, B, AH, MH and CS, respectively (p≤ .01). CH₄ *in vitro* production was related with DDM (r=.97; p≤ .01). CH₄ production per 10kg of DDM was: 286 b; 252 a; 286b; 337c and 336c (p≤ .01) for A, B, AH, MH and CS, respectively. Results suggest that methane production is about 54.5 kg /animal/yr (range 50-59 Kg/ animal/yr), this value coincides with 54 kg/yr estimated for Argentina by Crutzen (1986) and with the estimated value for latinamerica (58/kg/animal/yr) by the Intergovernmental Panel for Weather Change. Results show that, as with DDM, A and B were less methanogenic than MH and CS and that the methodology employed, available for developing countries, is viable of being used for the study of the methanogenesis of pasturing animals.

95. CHANGES IN THE ELECTROACTIVE CHARACTERISTICS OF OVINE RUMEN FLUID SUPPLEMENTED WITH GLUCOSE

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The attempt to improve the efficiency of the rumen system at a scale of microorganisms is related with a better understanding of the bidirectional interaction between functional populations and forage-added substrate availability. In the anaerobic environment of the rumen, forage constituents contribute to the formation of a large number of intermediary. Many of them which, in the function of their electroactive character, are effectors of the redox state of the rumen system and can affect the structure of the functional microbial population of the rumen. On the other hand, the microbial compound can modify the concentration and/or speciation of the chemical elements. The study of the fast biochemical changes that take place at a microbial scale require techniques that could be used in a complex and heterogeneous system such as the rumen fluid. In this work glucose was added to the filtered ovine rumen fluid (1%, p:v) and the electrochemical characteristics of the suspension were explored for 3h after the addition employing cyclic voltammetry (initial potential: - 1000 mV; final potential: + 949 mV) (Radiometer Analytical, France). A glassy carbon electrode was employed as the work electrode, KCl 0.4 mM as electrolyte, and water as solvent. The cyclic voltammetry graphs were well defined and those resulting from derivation of voltangrams clearly showed the glucose effect (which showed a characteristic peak of about - 500 mV) and that the glucose supplementation also generated changes in the potentials of - 700 to - 1000 mV.

96. BIOSOLID-TREATED SOIL: IMPACT OF THE AVAILABILITY OF TERMINAL ELECTRON ACCEPTORS UPON THE CO₂ AND N₂O PRODUCTION

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The addition of biosolids to soil introduce organic compounds that favour, by one way, the fertility through the biological oxidations, and by other way, the gas-production that increase the air pollution. In this work, the soil used was an Argiudol vertic (from Zavalla - Santa Fe province-Argentina) supplemented with: 0, 3, 6, 12, 24 % of biosolid (%OC: 12,2; %tN: 0,9 and pH: 6,8) incubated in microcosms condition during 40 days.

The CO₂ and N₂O production were measured after 30 hours of samples incubation, using laboratory incubation procedure. The production of the CO₂ in aerobic and anaerobic conditions was relationsheeped with the quantity of biosolid added. The polinomic regression lines obtained indicated that the quantity of CO₂ production in aerobic and anaerobic conditions was limited with the highest biosolid-dose. When the NO₃⁻ was the terminal electron acceptors the CO₂ production was limited at 12% of biosolid. Our study also suggest that the amount of endogenous NO₃⁻ limited the expression of the functional microflora that produce the N₂O.

97. ESTIMATION OF VARIANCE COMPONENTS FOR FRUIT TRAITS IN TOMATO

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Genetic variation is due to both additive genetic variance (V_A) and non-additive genetic variance (V_D) and together with the environmental variance (V_e) they are present in F_2 generations. The aim of this work was to estimate the broad sense heritability (h^2_b) and the narrow sense heritability (h^2_n) for some fruit traits in the following genotypes and generations ($n=160$): a genotype carrying the nor mutant of *L. esculentum* var. *esculentum* (P_1), the accession LA1385 of *L. esculentum* var. *cerasiforme* (P_2), the F_1 ($P_1 \times P_2$), the resulting F_2 generation and the backcross BC_1 ($F_1 \times P_1$) and BC_2 ($F_1 \times P_2$). The fruit traits evaluated were: diameter (D), height (H), weight (W), shelf life (S), color (C) and firmness (F). The h^2_b and h^2_n were estimated as: $(V_A + V_D)/(V_A + V_D + V_e)$ and $(V_A)/(V_A + V_D + V_e)$ respectively. Results are detailed in the following table:

	D	H	W	S	C	F
h^2_b	0,69	0,81	0,77	0,45	0,65	0,67
h^2_n	0,69	0,81	0,77	0,00	0,10	0,02

Genetic variation for the D, H and W traits were mainly due to additive variance but the genetic variance estimated for the S, C and F traits was mainly caused by non additive genetic component. The improvement of D, H, and W traits could be achieved by directional selection while to improve the S, C and F traits in this genetic context it would be better to produce hybrid genotypes.

98. ANALYSIS OF THE VEGETATION OF THE ECOLOGICAL RESERVE 'EL BAGUAL' (FORMOSA-ARGENTINA)

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The Ecological Reserve 'El Bagual' (REB) is a protected area in southeastern Formosa province, Argentina. The present knowledge of the local and regional vegetation is too scarce for the establishment of a management and conservation plan. The objectives are: to obtain an inventory of the flora, to determine the existing environmental units and to classify them according to their floristic composition, and to establish their phytogeographical position according to the lineage of their floristic elements. The methodology consisted in the analysis of satellite imagery to determine the main vegetation units, later land survey of their floristic composition and finally to determine their physiognomy. The results are: a complete floristic list of the REB, classification and mapping of the environmental units and their phytogeographic position. The vegetation of the REB shows a marked heterogeneity both floristical and physiognomical. It is remarkable that in this relatively small area, 558 species and 102 vascular plants family are found; these represent 71% of the total of families of the province and almost 40% of the total of species of Formosa. These results confirm the importance of the creation of protected areas as tools of conservation of the natural patrimony.

99. THE WOODY SPECIES OF THE ORDER RANALES IN THE SANTA FE PROVINCE (ARGENTINA)

Lusardi MB, García RC, Prado DE.

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The Order *Ranales* comprises 11 families of aquatic, herbaceous and woody plant species with hypogynous flowers, free carpels or instead a single carpel, perianth not very well differentiated in calyx and corolla. In the Santa Fe province there exist only three families with native woody species. They are: in the *Lauraceae*, *Nectandra angustifolia* (Schrader) Nees, 'laurel amarillo', and *Laurus nobilis* L., the common laurel, an exotic species growing subspontaneous in several localities in the province; in the *Annonaceae*, only *Rollinia emarginata* Schltdl., the 'arachichú' of edible fruits; and in the *Berberidaceae*, only *Berberis ruscifolia* Lam., the spiny 'uvilla' or 'espinilla amarilla'. The present work contributes to the knowledge of this family providing keys to the identification of the different taxa based on morphological characters. The methodology consisted of a bibliographical review, consultation of national herbaria with important collections of the province (SF: Esperanza; SI: Darwinion, San Isidro; UNR: Rosario), and lab work to confirm their identity. It is here provided taxonomic information, illustrations and a distribution map.

100. STUDY OF THE ANACARDIACEAE FAMILY IN THE SANTA FE PROVINCE, ARGENTINA

Oakley L, Maturo H, García R, Prado DE.

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The family *Anacardiaceae*, in the Order *Sapindales*, comprises 74 genera and 600 species of pantropical and temperate distribution in the world. In Argentina the family *Anacardiaceae* includes six genera, four of which are present in the Santa Fe province. The present work contributes to the knowledge of this family providing keys to the identification of its taxa based on morphological characters and an analysis of their geographical distribution. The methodology consisted of bibliographic revision, consultation of national herbaria with important collections of the province flora (SF, SI, UNR), lab work to confirm their identity together with the field experience of the authors.

In the genus *Lithrea* Hook. & Arn., the 'molle de beber' *L. molleoides* (Vell.) Engl. has been cited but could not be confirmed so far. In *Myracrodruon* Fr. Allemão (= *Astronium* Jacq. subgen. *Myracrodruon* (Fr. Allemão) Barkley), there occur *M. balansae* (Engl.) D.A. Santin (= *Astronium balansae* Engl.); in *Schinopsis* Engl. two species: *S. balansae* Engl., *S. quebracho-colorado* (Schltdl.) F.A. Barkley & T. Mey. (= *S. lorentzii* (Griseb.) Engl.), and one alleged interspecific hybrid: *S. heterophylla* Ragon. & J. Castigl.; in *Schinus* L. five species and four varieties, all known as 'molles': *S. bumelioides* I.M. Johnst., *S. engleri* F.A. Barkley var. *uruguayensis* F.A. Barkley, *S. fasciculata* (Griseb.) I.M. Johnst. var. *fasciculata* y var. *arenicola* (Hauman) F.A. Barkley, *S. longifolia* (Lindl.) Speg. var. *longifolia* y *S. molle* L.

101.

THE FAMILY ZYGOPHYLLACEAE IN THE PROVINCE OF SANTA FE (ARGENTINA)

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The *Zygophyllaceae*, in the Order *Geraniales*, is a pantropical family of over 25 genera and 250 species. It comprises herbaceous, shrubby or arboreal plants with opposite stipulate leaves, perfect tetra- or pentamerous flowers, solitary or in pauci-flowered cymes. The object of the present contribution is to set off the taxonomical study of the family for Santa Fe, through observation in the field, analysis of plant collections (Herbaria SF, SI and UNR) and revision of the literature. Seven genera and 16 species are known to occur in Argentina, of which only three genera are present in Santa Fe. These are the genera *Kallstroemia* Scop., with the small herbaceous *K. tucumanensis* Descole, O'Donnell & Lourteig found only in the Dpt. 9 de Julio; *Porlieria* Ruiz & Pav. known only through the presence in the Dpts. 9 de Julio and Vera of the shrub *P. microphylla* (Baill.) Descole, O'Donnell & Lourteig; and in the genus *Tribulus* L. the 'roseta' *T. terrestris* L. has been collected, appearing in Santa Fe in areas under high anthropic activity and intensive cultivation.

102.

QUALITY OF SUNFLOWER SHOOTS REGENERATED *IN VITRO* UNDER DIFFERENT CULTURE CONDITONS

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In vitro manipulation of sunflower has some difficulties such as early flowering and hyperhydric shoots, both of which produce losses in the micropropagation process. The aim of this work was to analyse the culture conditions that influence the quality of the sunflower shoots regenerated *in vitro*. The genotype tested was the inbred line HA300B, using as explants cotyledons from mature seeds. The basal medium was a Murashige and Skoog (MS) (1962) supplemented with growth regulators: M1=MS + 5,71 µM IAA (indol acetic acid) + 9,29 µM KIN (kinetin); M1Ag= M1 + 5,0 µM AgNO₃. Two kinds of culture handling were tested: with and without plating the explants to a medium free of hormones after a seven-day exposure to the first medium. In the two cases cultures were grown for thirty days under three different photoperiods: 16/8, 12/12 and 8/16 hours. The traits evaluated were *percentage of shoot* (V), *callus* (C), *hyperhydricity* (H) and *early flowering* (F). Data were analysed through the non-parametric *Chi-square* test (χ^2). Significant differences were observed among the combination of treatments for V, C and H under the three photoperiods. Both the addition of AgNO₃ and the fact of plating the explants reduced the hyperhydricity of shoots. Nevertheless, this quality improvement was associated to a reduction in regeneration ability. Losses due to early flowering were observed however no significant differences were observed among media, culture handling and photoperiods. To conclude, both the media and the culture handling seem to have some influence on hyperhydricity of shoots regenerated *in vitro*.

103.

EVALUATION OF CALLUS DEVELOPMENT IN SUNFLOWER INBRED LINES

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The aim of this study was to establish culture conditions for the development of callus in three sunflower inbred lines with different levels for resistance to imidazolinones. The genotypes evaluated were HA425B (resistant); 1058B (intermediate) and M1711B (susceptible). Culture media (M) were formulated on Murashige and Skoog (1962) (MS) saline base supplemented with growth regulators: Mc₁= MS + 0.1 mg/l GA + 0.1 mg/l 2,4-D; Mc₂= MS + 0.5 mg/l KIN; Mc₃= MS + 1 mg/l 2,4-D; Mc₄= MS + 0.1 mg/l BAP + 2 mg/l 2,4-D. Cotyledons from mature seeds and hypocotyls from seedlings were used as explants. Cultures were grown at 25±2°C with a 12-hr photoperiod in a randomized design with two replications. Percentage of callus (C) and fresh callus weight (FCW) were evaluated. On Mc₂ hypertrophy of the explants was observed for all treatments. Under Mc₁ conditions all lines developed callus from hypocotyls but the percentage varied with the genotype. Percentage of callus was 100% for all treatments on Mc₃ and Mc₄. Effect of genotype (F=10,02; p<0,0011) and explant (F=45,94; p<0,0001) was observed for FCW. Callus from hypocotyls presented lower mean values (404,53 mg) than those obtained from cotyledons (1120,63 mg). On Mc₄ callus from cotyledonary explants presented higher mean values than those obtained on Mc₃ conditions. For these genotypes Mc₄ culture conditions would be useful for the *in vitro* callus proliferation from cotyledonary explants. These results would provide the basis for the study of *in vitro* response to imidazolinones.

104.

VEGETATIVE AND REPRODUCTIVE CHARACTERISTICS OF DIFFERENT SPECIE AND CULTIVARS OF WALNUT IN ZAVALLA (STA FE). PRELIMINARY STUDY

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Juglans genus has aproximality 23 specie, being *J. regia* and *J. nigra* of great economical significance. *J. hindsii* and paradox are also stocks of *J. regia*. *J. australis* is a native specie in the NW of our country, and also used as stock of *J. regia*. They are monoic plants, with male flowers in long and alone aments in one year branches. Female flowers are in terminal or lateral bunches in the present vegetative growing. The purpose of this work is to study the phenology of the *Juglans* genus and the evolution of buds. The study took place in Zavalla (33°1' LS-60°53' LO-30 msn) in the period April 2003-March 2004. 3 specie of the *Juglans* genus: *J. nigra*, *J. australis* and *J. regia* and 4 cultivars of *J. regia*: Davis, Chandler, Tulare y Franquette were evaluated. *J. nigra* and *J. australis* are not submitted to cultural work, while cultivars of *J. regia* are in the experimental orchard of Fac. de Cs. Ag. UNR. 3 plants of each specie or cultivar and 2 branches of each one were studied. Foliation to fructification dates were observed and diameter and long of male buds were registrated. Measurement were taken periodically and monthly expressed. In April 2003 Davis, Tulare, Chandler and *J. nigra* firstly showed signs of leaves falling, while the lastones were Franquette and *J. australis* in June-July 2003. Male aments differentiated in Summer and the maximum development was in Spring. Foliation took place in September for *J. australis*, and in October for *J. nigra*, and Davis, Tulare and Chandler cultivars. Franquette budded in November. *J. australis* and *J. nigra* firstly enlarged fruits in October, Davis, Tulare and Chandler in November. Tulare and Davis showed pericarp dehiscence in February. Fruit falling was 100% in March, only in *J. australis*.

105.

SOYBEAN: THE RELATION BETWEEN THE VEGETATIVE MASS AND THE YIELD

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To obtain high yield it is required to have high solar interception and an efficient partition of the biomass towards the seeds, measured through the harvest index. In order to evaluate this hypothesis, a trial was carried out at Zavalla with two sowing dates: 10/11(SD1) and 19/12/2003 (SD2). The cultivars (Cv) were: AW2886 RR (GM II); A4201 RG; A 4303 RG and Aw RR (GM IV); RA 514 and RA 516 (GM V) and A 7322 RG (GM VII). The experimental design was a complete blocks with three replications. To the harvest, it was determined the height of the plants (alt), the number of knots in the main stem (nu), total air biomass (g/m², BAT), yield (kg/ha, REND), weight of seeds (PUS) and the number of seeds/m² (NSEM). The variance analysis was performed and for all the studied variables, the statistical analysis detected significant differences ($P < 0.05$) between dates, cultivars and interaction date*cultivars. The greatest vegetative mass was done in the SD2; while the greatest yield occurred in SD1. This results did not establish a positive association between vegetative mass and yield. The great production of biomass in SD2 was due to high temperature in the vegetative phase, average of the 7 CV, 1,8°C higher. Besides, the Cv of GM V and VII had a great duration in the vegetative period, so they produced more BAT, falling and more hydric consumption in this period, and due to low rain during grain filling, PUS and REN were reduced. Besides the low radiation interception and the short period of filling explained the low average values of IC. The Cv A7322 RG manifested a high susceptibility to *Diaporthe phaseolorum*, so it showed a reduced value of IC and REND too.

106.

THE HEALTHY QUALITY SEED OF MAIZE

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The objective of this study was to evaluate the different performance the *Fusarium* (FUS), *Penicillium* (PEN), *Diplodia* (DIP), *Aspergillus* (ASP), *Alternaria* (ALT) and *Nigrospora* (NIG) under field conditions (chisel plow, no till management) interacting with three nitrogen (N) level and two sulfur (S) level). The research was conducted from 1999 to 2003 at Zavalla (33° 01' S. L.; 60° 53' W. L.) Environmental variables: mean daily maximum temperature (Tmax), mean daily minimum temperature (Tmin), mean daily temperature (Tm), relative maximum humidity (Hrmax), relative minimum humidity (Hrmin), mean relative humidity (Hrm) and precipitation (Ppt) from stage R₁ (50% plant with visible silk) to 30 days later were registered. The percentage of seed infected by each pathogen and the germination (PG) of each samples were established. Data were analyzed using ANOVA method and Pearson correlation analysis between percent of seed with pathogens and environmental variables. The tillage systems (TS), environmental (E) * TS interaction, N*S, N*S*E interaction were no significant. The E was significant for FUS, PEN and PG ($p < 0,01$) and for DIP ($p < 0,05$). FUS was negatively correlated with Hrmax_{1,2 and 3}, Hrmin₃, Hrm₁ and Tmin₁ ($r > 0,70$) and PEN was positively correlated Tmax₂, Ppt₃ ($r > 0,60$). DIP was negatively correlated with Tmax₁ and Tm₁ ($r > 0,76$). PG was positively correlated with Tmin₂ and Tm₁ ($r > 0,77$) and was negatively correlated with Hrmin_{1,2 and 3} and Ppt₂ ($r > -0,72$). The data shown FUS, PEN and DIP were more influenced by the E than the TS and the N* S combinations.

107.

LATENCY GROUP AND NUTRITIONAL CHARACTERS OF ALFALFA CULTIVARS IN FOUR MATURITY STAGES

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Alfalfa (*Medicago sativa* L.) is the most indicated legume forage for meat and milk production systems because of its animal acceptability and its high quantitative and qualitative productive potential. The objective of this work was to evaluate the effect of latency group (LG) on Crude Protein (CP%) and Neutral Detergent Fiber (NDF%) content and *in vitro* Organic Matter Degradability (IVOMD%) of alfalfa cultivars during spring-summer growth period. Six alfalfa cultivars of different LG were sown in autumn of 2003: Tahoe and Victoria (LG 6); Mayaco (LG 7); Monarca (LG 8-9); Araucana and Reina (LG 9). It was applied a completely randomized design with 3 replications. During spring and summer of the first year cuts were made in four maturity stages: Vegetative (V), Bud (B), Early Bloom (EB) and Full Bloom (FB). In the samples of each cultivar obtained by cut with mechanical scissors (6 cm on the surface of the ground) and dried in stove (60°C) were determined CP% (Kjeldahl), NDF% (Van Soest) and IVOMD% (Menke). Results obtained for LG 6-7 and 8-9 were analyzed by oneway ANOVA. Only in EB content of CP in LG 8-9 was greater than in LG 6-7 (20,7±1,45 vs. 18,2±0,54; $p = 0,07$). This greater protein availability in rumen could partially explain the greater IVOMD found in the group of smaller latency (64,43±0,60 vs. 62,19±0,47; $p = 0,01$) for the same maturity stage. No differences were found in studied variables ($p > 0,10$) for the other maturity stages studied. In the experimental conditions of this work results suggest that in spring-summer growing period, nutritional characters are similar for alfalfa cultivars of different winter rest.

108.

PREVALENCE OF *Diaporthe phaseolorum* var. *caulivora* AS CAUSAL AGENT OF SOYBEAN STEM CANCER IN ARGENTINA

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Soybean stem canker (SSC) is a disease of economic relevance in Argentina since 1996/97. In previous studies, we reported: i) the co-existence of both causal agents of SSC (i.e. *D. phaseolorum* var. *meridionalis* (DPM), and *D. phaseolorum* var. *caulivora* (DPC), in the core soybean growing area of Argentina, and ii) that genes conferring resistance to DPM do not protect against DPC. During the growing season 2003-2004, the occurrence of infected plants showing SSC symptoms typical of DPC increased dramatically in the field. The objective of this work was to identify, morphological and molecularly, the causal agent of the SSC prevalent in the core soybean producing area of Argentina. Plants showing typical SSC symptoms were collected from soybean fields randomly distributed in the Santa Fe and Buenos Aires Provinces. From these samples, nineteen DPC isolates were obtained and characterized by colony morphology and color, perithecia grouping and shape, and asci and ascospores size. The fungal genomic DNA was analyzed by the RAPD method, using proper DPM and DPC controls. Others isolates of the *D. phaseolorum* complex, as well as isolates of *Fusarium* and *Colletotrichum* were also included as additional controls. All isolates collected were characterized, morphological and molecularly, as DPC, showing the prevalence of this variety in the core soybean area of Argentina. We suggest this happened because the breeding for SSC resistance was directed to the incorporation of DPM resistance genes. As stated above, those genes do not confer resistance to DPC. Thus, the spread of DPC would have been indirectly favored.

109.

HISTOLOGICAL MODIFICATIONS ON SOYBEAN STEM TISSUES INDUCED BY *Diaporthe phaseolorum* var. *meridionalis*

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The objectives of this work were i) to quantify the disease infection progress, and ii) to characterize some histological modifications in the stem tissues of soybean infected by *D. phaseolorum* var. *meridionalis* (DPM), one of the causal agents of soybean stem canker. Seedlings of soybean cv. Tracy M (*Rdm1* and *Rdm2* resistance genes) and cv. RA 702 (susceptible, without *Rdm* genes) were inoculated with DPM mycelia. The inoculation point (ip) was 0,6 cm below of the cotyledonal node. Five inoculated and control seedlings were harvested at 5, 11 and 21 days after inoculation (dai). Three internode sections: proximal, media and distal (~1,2 cm each) were obtained from the first internode, starting at the cotyledonal node. These sections were included in paraffin, and stained with safranin-fast green to evaluate the temporal and spatial progress of infective process, and to determine the infection rate. Calosa deposit on the cribose plate of phloem tissue was observed by staining with resorcin blue. Additionally, histo-chemical tests (starch, lignin, lipids, tannins) were done on transversal sections of fresh tissues (~0,4 cm from the ip). DPM induced histological alterations in both, resistant and susceptible cvs. However, these changes were not effective to stop DPM colonization in the susceptible cv. RA 702. It is concluded that the resistance to DPM, present in the cv. Tracy M, involve other/s defense mechanism/s not evaluated in this instance.

110.

EFFECT OF ALUMINIUM ON CALCIUM ABSORPTION IN SMALL INTESTINE OF PREGNANT RATS

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During rat pregnancy occurs an early adaptive increase of intestinal calcium (Ca) absorption to allow maternal skeleton accumulates Ca to meet the peak of fetal demand by mid-pregnancy, before the onset of rapid skeletal mineralization of the fetus. Prolactin (PRL) and estradiol (E2) can partially mediate normal increase of Ca absorption in the intestine during this stage. Previously we have found that inhibition produced by aluminium (Al) of intestinal Ca transport in cycling female rats was dependent of 17 β -estradiol serum levels. The aim of present work was to study the effect of oral Al overload on intestinal Ca absorption in pregnant rats. Adult female Wistar rats were treated from day 1 of pregnancy with 0 (C), 60 or 120 mg AlCl₃/kg b.w per day o.g. Experimental groups treated with larger dose of Al were injected from day 20 with 0.5 mg/kg b.w per day i.p. of bromocriptine (BrC, PRL secretion inhibitor) or 200 μ g/kg b.w per day s.c. of E2. Controls without Al that received the same treatment, were included. Before delivery, *in vitro* Ca transport of everted duodenum-jejunum sacs (JCams) by using ⁴⁵Ca as flux marker, were determined. Results were expressed as μ mol Ca/h/ g w.w. Means of at least n=4 measurements per group were compared by ANOVA. Al produced a significant reduction of JCams in a dose-related fashion, as compared to control (C=4.23 $\pm$ 1.65; Al 60=2.61 $\pm$ 0.10*; Al 120=1.78 $\pm$ 0.36* μ mol Ca/h/g w.w., *P < 0.05). BrC treatment diminished the percentage of inhibition of JCams produced by Al. On the contrary, the same parameter was significantly increased by E2 injection. We can conclude Al diminishes transcellular Ca absorption in the small intestine of pregnant rats. Al inhibition could occur by interference with physiological mechanisms of Ca transport mediated by serum level increments of both PRL and E2, observed during late pregnancy.

111.

UTILIZATION OF N-ALKANES AND PETROLEUM DERIVATES BY BACTERIA PRESENT IN A CONSORTIUM

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Microbial bioremediation has been studied in the last decades. Isolated microorganisms can use limited number of compounds, but in the environment they form a mixed population that increases the biodegradation rate. Little is known about the interactions among them. A microbial consortium able to grow with hexadecane (Hxd) as carbon source has been isolated from a gas oil-contaminated soil. The microorganisms were identified as *Corynebacterium* sp and *Agrobacterium* sp. The aim of this work is to study the utilization of n-alkanes and petroleum mixtures as carbon source by these bacteria. The utilization of the different compounds was analyzed by increases in optical density after 14 days of incubation at 25°C. The following n-alkanes were added at 0.2% to a mineral salt medium: hexane, heptane, octane, iso-octane, undecane, dodecane, pentadecane, Hxd; gas-oil and 3 different petroleum cuts were used as complex mixtures. Then, the optimum concentration for growth was determined by supplementing the medium with 0.025 to 10% of each. The results show that only *Corynebacterium* is able to use n-alkanes longer than 8 C to grow. Although both bacteria were able to grow in gas-oil, only *Corynebacterium* grew in cut n° 2, which is rich in n-decane. Both types of microorganisms were found after co-cultures incubation adding undecane, dodecane, pentadecane and Hxd. These indicate that both bacteria use different compounds for growth and it could produce an increase in the degradation rate of gas-oil components. Moreover, the existence of a metabolic interrelation between them due to the fact that *Agrobacterium* is able to growth at the expense of a subproduct generated by *Corynebacterium* sp.

112.

APOPTOSIS IN THE RECTAL CRYPTS OF TAXOL® TREATED MICEGonzález NV¹, Barbeito CG^{2,3}.¹Cátedra de Histología y Embriología Animal, FCN y M. 122 y 60;²Cátedra de Histología y Embriología, ³Instituto de Patología, Fac. de Ciencias Veterinarias, Universidad Nacional de La Plata, 60 y 118. La Plata. E-mail: nvgonzal@infovia.com.ar

Paclitaxel (Taxol®) and colchicine are two microtubule poisons that induce apoptosis *in vivo* and *in vitro*. In previous work we demonstrated that colchicine induces cell death in the rectal crypts while this agent's effect displays daily variations. Taxol is a drug employed for the treatment of a variety of tumors. In this communication we present the analysis of the results of paclitaxel's effect on the rectal crypts along a complete circadian period. 53 male CH3/S adult mice under standardized conditions were used. Every 4 h, since midnight until the end of a 24h period, lots (n=4-5) were sacrificed, having been injected IP paclitaxel (20 μ g/g body weight) or saline four hours before. Rectum samples were excised and processed to assess apoptotic index. Values of each lot were expressed as mean $\pm$ SEM. The statistical analysis was performed by ANOVA and Bonferroni test. Under the present conditions, taxol® did not induce apoptosis in the rectal crypts. No statistical differences between apoptotic indices of time points were found. These results differ to the findings when colchicine was used in an identical experimental design. The colchicine treatment enhances the number of apoptotic cells and the drug's effects shows significative circadian variations.

113.

HISTOLOGICAL AND MORPHOMETRIC ASPECTS OF THE OVARY OF THE PLAIN VISCACHA (*Lagostomus maximus maximus*)

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The plain viscacha (*Lagostomus maximus maximus*) is a *Histricognathi* rodent that presents some reproductive characteristics such as having natural polyovulation (200-800 ovocytes), implantation of 10-12 embryos and birth of 1-3 breedings. The goal of the present work was to microscopically and morphometrically describe the ovary of the viscacha and to establish its variations in different reproductive conditions. Thirty six mature females, weighting 4,5 to 5 Kg were used. They were captured in different month of the year. The ovaries were processed for paraffin inclusion. Slices were stained with Hematoxylin-Eosin, Trichromic of Masson, Gomori, PAS and Alcian Blue, pH 2.5. The diameter of ovocytes and follicles and the area and roundness of follicles and luteal bodies were taken. All the numeric data were statistically analyzed. The ovary is formed by cords of tortuous and irregular aspect. Each cord is constituted by cortex and medulla. The primordial follicles are the most abundant and they form clusters. Morphometric data showed that the follicles have a similar size of other *Histricognathi*, except for the tertiary follicles that are smaller in the viscacha. These follicles do not protrude on the ovarian surface. The presence of numerous accessory luteal bodies and of abundant interstitial tissue is another characteristic that the viscacha shares with other members of the suborder.

114.

EVALUATION OF MORPHOLOGICAL AND FUNCTIONAL ASPECTS OF CELLULAR POPULATIONS OF THE IMMUNE SYSTEM AND OF ASSOCIATE CELLS, IN BOVINES INTOXICATED WITH *Solanum glaucophyllum*

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Solanum glaucophyllum (Sg) is one of the calcinogenic plants causing Enzootic calcinosis. It possesses glycosides of the 1.25 dihydroxvitamin D3. Vitamin D has multiple effects, highlighting its immunomodulating activity. High amount of vitamin D are incorporated into the organism during intoxication with Sg. To experimentally determine the effects of Sg on the immune system, the plant was orally administered at a dose of 50g of leaves per week to 2 year-old female bovines. Animals were sacrificed at 15, 30 or 60 days pi. Two animals were used as controls. Peritoneal macrophages were obtained for *in vitro* phagocytic test. At necropsy, all the intoxicated animals showed some type of compatible lesion with calcifications. Fresh lymphnodes isolated dendritic cells were used for detecting S-100 protein. The amount of immunostained cells was digitally determined. Cervical lymphnodes were processed for histopathological analysis. The cortical area width of the lymphnodes of intoxicated animals decreased as well as the relative number of cellular populations of the immune system and related cells, per unit area. Structural changes of the lymphnodes were observed with intoxication time. The analysis of isolated cells showed a progressive reduction of dendritic cells as well as a significant decrease of their size in intoxicated animals. The results demonstrate that the intoxication with Sg alters the immune system and related cells.

115.

EVALUATION OF Na⁺ EFFECT IN THE CONTRACTION OF THE MARGINAL SPHINCTER OF THE SEA ANEMONE *Phymactis clematis* (ACTINIARIA)

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The aim of this work is to study the role of Na⁺ in the release of Ca²⁺ from intracellular stores (IS) to activate smooth muscle contraction of this sphincter. Upon KCl stimulus the isolated sphincter is contracted and the contraction force increases with [K⁺]. We studied voltage sensitive Ca²⁺ channels with blockers such as Felodipine. In absence of Ca²⁺ the muscle is stimulated to low [K⁺]. Contractile responses are recovered when Ca²⁺ is added to the medium, demonstrating that Ca²⁺ is released from IS by depolarization of the membrane. The low response when the medium is replaced by sucrose suggests Na⁺ participation in Ca²⁺ release from these stores. The sphincter was placed in a chamber for isolated organ at controlled T°, pH and O₂ and connected to a polygraph. Dose-response curves in sea water (SW), in artificial SW without Na⁺ and in SW with a channel Na⁺ blocker Saxitoxina (STX), were estimated. In this last experiment the media used were: 1° SW, 2° SW + 5nM STX, 3° SW. In all cases KCl was used as depolarizing solution. Student's t test was used (n=10). P values < 0.05 were considered statistically different. The effect of time was statistically discarded. In all cases, the contractile force augmented at increasing [KCl] in the sigmoid form, without significant differences with each other (p>0.05). These results suggest that in these experimental conditions Na⁺ did not have an active participation in the contractility. KCl depolarization of the membrane might open Ca²⁺ channels promoting the influx of this ion or releasing it from the IS in order to initiate contraction.

116.

BIOLOGICAL EFFECTS OF PROLONGUED TREATMENT WITH ALUMINUM LACTATE (Lac₃Al) IN ADULT MALE RATS

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The goals set in this work were the analysis of sequential variations in hematological and renal parameters that could be affected by Lac₃Al, and to evaluate possible oxidative damage in kidney and liver. Male Wistar rats were distributed in 2 groups: C (control, n=6) and LacAl (aluminized, n=7). For a 3 month period, LacAl group was injected with 0,5ml of Lac₃Al (0,8mg Al/ 100g body weight, 3 times per week), and C group 0,5ml of sodium (Na) lactate, in the same frequency. At the end of the 1st, 2nd and 3rd month, water and Na balances were measured, as well as osmolarity, urine concentration tests, hematological parameters and seric urea. At the end of the 3rd month, we measured proteins in urine, seric aldosterone (Ald), hepatic and renal glutation-SH (GSH, Elman) and lipoperoxidation (LPO, Ohkawa). (*P<0,05). Results: a) 3rd month, LacAl group increased red blood count* (7,94vsC:6,75M/L), and decreased hemoglobin (Hb) concentration* (10,87vs C:13,80g/dL), hematocrit* (31,3vsC:37,14%), medium corpuscular volume* (39vsC:53fL) and medium corpuscular Hb* (14,30vsC:19,58Pg). b) glomerular filtration, indicated by seric urea, was similar in both groups; c) Na balances were altered in LacAl due to a minor food intake, in spite of the increased Ald I(C:69,8,LacAl:161,5 pg/ml); d) Water balance decreased in the first 2 months, and was recuperated in the third due to a diuresis reduction* (C:12,21,LacAl:8,55ml/day). In a water depletion situation, a diminished urine concentration capacity was observed, with an increased volume minute* (C:3,08e-3, LacAl:7,10e-3ml/min) and low osmolarity (C:2484,LacAl: 1862mOsm/l). Conclusions: Lac₃Al administered to adult rats for 3 months induces hematological changes, and alterations in the renal function, associated with tubular processes. In a metabolic level, it increases the oxidative stress in liver and kidney according to elevated LPO and low GSH.

117.

PREVENTIVE EFFECT OF VITAMIN E ON THE OXIDATIVE STRESS CAUSED BY ALUMINUM (AL) IN RATS

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In previous works we have demonstrated that Al produces an increase of the lipoperoxidación (LPO) accompanied by a decrease in the antioxidative enzyme activity catalase (CAT) and glutathione peroxidase (GSH-Px) likewise also in the glutathione (GSH) hepatic content. The objective of the following work was to analyze the antioxidative effect of vitamin E (vit E) on the oxidative stress caused by Al in adult male Wistar rats livers (n=5 for each group), weight average 300 g, divided in the following experimental groups: A: controls; B: treated with Al (elementary one, 27 mg/kg corporal weight ip, 3 times per week during 3 months); C: vit E, 600 mg/kg corporal weight, subcutaneous, during 3 months; D: simultaneously treated with vit E and Al. We studied the activity of catalase by the Beers method, glutathione peroxidase by the Paglia and Valentine method, lipoperoxidation through the measurement of reactive substances with tiobarbituric acid by the Ohkawa method, and the content of glutathione by the Elman method. The protein concentration was determined by the Lowry method. The results were expressed like the average $\pm$ SEM (* significant difference $p < 0.05$) LPO (nmolMDA/gtissue) A: 275.51 $\pm$ 38.5 B: 573.2 $\pm$ 75.5*; C: 289 $\pm$ 21.3; D: 166 $\pm$ 17.3. GSH (nmol/g liver.) A: 3.47 $\pm$ 0.14; B: 2.01 $\pm$ 0.12*; C: 4.56 $\pm$ 0.2; D: 4.01 $\pm$ 0.2; GSH-Px (U/mgprot) A: 82.84 $\pm$ 7.05; B: 45.2 $\pm$ 3.31*; C: 95.2 $\pm$ 2.98; D: 89.23 $\pm$ 2.92. CAT (U/mgprot) A: 0.49 $\pm$ 0.031; B: 0.04 $\pm$ 0.003*; C: 0.39 $\pm$ 0.029; D: 0.28 $\pm$ 0.01. We observed that the Vit effects are in resisting the oxidative stress caused by chronic administration of Al. These findings could have clinical relevance, since they suggest the hepatotoxic effect caused by Al, through the induction of oxidative stress and the decrease of antioxidative defenses associated to the same one could efficiently be resisted by the antioxidative treatment with Vit E.

118.

INFLUENCES OF SEX IN KIDNEY FUNCTIONS OF RATS SUBMISSIVE PARTIAL HEPATECTOMY

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Our objective was to evaluate the influence of the sort on the kidney functions in a model of partial hepatectomy (65% HP) to the 4 days post surgery. Male adult Wistar rats and females were distributed in four experimental groups: control males (MC n=11), males + HP (MHP n=8), control females (HC n=9), females + HP (HHp n=7). Groups HP showed a reduction of the water balance, associated in MHP to a diminution in the ingestion, whereas in HHp was due to an increase of the diuresis. In both groups a significant reduction of the urinary osmolarity was observed. On the other hand, single MHP simultaneously presented a decreased Na balance. HHp did not modify glomerular filtration rate (GFR), but presented an increase of p-aminohippuric clearance (CPAH ml/min.100g): HC 1.9 $\pm$ 0.12; HHp 3.22 $\pm$ 0.12 * ($p < 0.05$). In MHP a reduction in GFR was observed (ml/min 100 g): MC 0.81 $\pm$ 0.2, MHP 0.67 $\pm$ 0.05 * ($p < 0.05$) without changes in the CPAH. Both MHP and HHp increased water's fractional excretion (EF%), in spite a single significant increase in the EF%Na and in the reduction of the osmolarity in HHp. Both groups showed a similar increase in the seric concentration of aldosterone (pg/ml): MC 330 $\pm$ 40, MHP 660 $\pm$ 96 *, 314 HC $\pm$ 36, HHp 503 $\pm$ 44* ($p < 0.05$) without changes in the seric concentrations of Na, potassium, and creatinine. The hepatic function agreed with an increase in LDH. There are gender differences in renal hemodynamic parameters. During the process of hepatic regeneration, renal hemodynamic and tubular changes can be observed, which in addition would be influenced by sex. In females, the kidney could self-regulate the GFR, in spite of the increase of the plasmatic flow. On the other hand this increase could be the one in charge of the accentuated natriuretic diuresis observed in the same ones. The increase of seric aldosterone could maintain the sodium balance in HHp, not so in MHP in spite of the reduction in the sodium elimination.

119.

INITIATION OF ROOT PRIMORDIA IN CUPHEA GLUTINOSA CHAM ET SCHLECHT (LYTHRACEAE) PLANTS ROOTS

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The objective of the present work was the initiation of root primordia in *Cuphea glutinosa* plants grown in nutritive solutions (Hoagland 50%) and light (220 mE) and temperature (15 $\pm$ 2°C) controlled conditions, with a primary root average length of 37 $\pm$ 2 mm. The observations were made on cuts obtained with ultramicrotome.

The initiation zone was showed optically dense as a result of the pericycle initial division. It corresponds to stage 1 of the radical primordia development over a total of 7 stages (0-6) according to the scale described by Blakely, L. M. *et al.* (1982). In this phase the length of pericyclic perturbation area placed in the primordia beginning is 75 $\pm$ 5 μ m, involving 6 hardly regular cells, each one with an average diameter of 12.5 $\pm$ 1 μ m. The initiation places were showed confronted to protoxylematic poles and it was not observed primordia initiation in adjacent and opposite poles. The presence of mechanic impediments in the growth substratum can produce curvatures and in these cases the differentiation of radical primordia was originate in the convex zone of them. The primordia initiation was determined by inner morphogenic patterns without the presence of external factors capable of modify the initiation place of lateral primordia.

120.

ROOT CAP AND MUCIGEL CHARACTERIZATION IN PLANTS ROOTS OF CUPHEA GLUTINOSA CHAM ET SCHLECHT (LYTHRACEAE)

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The objective of the present work was to characterize the root cap and mucigel present in *Cuphea glutinosa* plants grown in nutritive solutions (Hoagland 50%) and light (220 mE) and temperature (15 $\pm$ 2°C) controlled conditions, with a primary root average length of 37 $\pm$ 2 mm.

The radical apex of *Cuphea glutinosa* showed a prominent root cap with an average length of 192.5 $\pm$ 15 μ m and a diameter in the superior part of 140 $\pm$ 5 μ m. Assuming that the root cap has a cone regular shape is possible to determine its volume by the equation $\frac{1}{3} \pi r^2 h$ and replacing the *Cuphea glutinosa* value the total volume outcome is 988,000 μ m³.

From the root cap extreme to nearly over the radical hair zone is observed a remarkable hyaline, viscous layer with hardly uniform thickness between 1 and 2 mm. The greatest thickness is in the radical extremes and is getting smaller in basipetal direction up to vanish at an average distance of 1,400 $\pm$ 30 μ m from the root cap extreme, maturation zone with presence of radical hair.

Similar outcomes were observed in barley roots which growing in nutritive solutions presented a 0.9 mm thickness mucigel. However, works made with various species farmed in nutritive solutions showed that the mucigel thickness ranged from 10 to 200 μ m, both values were very inferior to the ones presented by *Cuphea glutinosa* in similar farm conditions.

121. IDENTIFICATION OF SPOILAGE MICROORGANISMS AND THEIR INHIBITION IN LEMONCELLO

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The process of lemoncello consists on the maceration in alcohol of washed lemon skin. It is filtered and mixed with syrup. The mixture is packed in glass bottles and sealed. During storage it was observed an important muddiness and discoloration in the neck of the bottles that demerit their aspects. The aim of this work is to determine the origin of the muddiness and the possibility of its prevention. Sample from the neck of the bottle was inoculated in surface by triplicate in dishes with agar HyL (molds and yeasts). It was incubated at 25°C by four days. Blue and orange growth of colonies was observed and identified as *Penicillium* spp. and *Crhisonilia* sp. respectively. *Penicillium* spp. is pollutants of several substrate and also well known as potentials producing of micotoxinas. *Crhisonilia* sp. are in the floor and in the skin of fruits and they generate allergic problems. To prevent these developments, the added of an appropriate concentration of Potassium Sorbate (PS) to the lemoncello were investigated. The concentrations of PS added to dishes with agar HyL were: 0, 0,05, 0,1 and 0,2. It was carried out inoculation in surface, of 0,1 lemoncello ml, from sample of the neck (concentrated) and of the homogenized content. In the inoculation from the neck had growth starting from the third day to 0 and 0,05% of PS. In the homogenized sample this it was presented to the third day only in the dish control. It was incorporated to the lemoncello 0,05%. The product it is not formed the muddiness of growth in the neck of the bottle and it doesn't present development in their entirety.

122. TRANSVERSAL CHARACTERIZATION OF ROOT ELONGATION AND MATURATION ZONES OF *CUPHEA GLUTINOSA* CHAM ET SCHLECHT (LYTHRACEAE) PLANTS

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The objective of this work was to characterize transversally the radical tissues present in elongation and maturation zones. The samples were made on growth plants in nutritive solutions (Hoagland 50%) and light (220 mE) and temperature (15 +/- 2°C) controlled conditions, with a primary root average length of 37 +/- 2 mm. The observations were made on cuts obtained with ultramicrotome.

The primary root presented an extensive elongation zone with an average length of 1007 +/- 100µm, which begins where the cortical cells double their length. From there and in basipetal direction the size ratios were 1.5x, 1.35x, 1.25x until keep constant its length reaching 60 and 75µ being the more probable size 62,5µ.

In the limit of the elongation and maturation zone it was possible to observe an extensive cortical parenchyma represented by cells of great size, thin walls, notable intercellular spaces and with great amount of cytoplasmatic inclusions. It was observed clearly the ratio 3:1 in favor of the cortical tissues in relation with the conduction tissues, inverse relation to the one presented in the base of the elongation zone. The maturation zone was characterized by the presence of root hair in its whole extension of 6.058µ with variable length and thicknesses.

123. VARIATIONS IN CELLULAR CONTENTS OF *CUPHEA GLUTINOSA* (LYTHRACEAE) PLANT STEMS THAT GROW IN DIFFERENT PLACES IN SIERRA DE LOS DIFUNTOS

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In previous studies made in Sierra de los Padres were found variations in the cellular contents (amyloplasts and TLS: tannin like substances) of different stem tissues in relation with ambiental growing conditions.

The objective of this work was to determine the existence of variations in the cellular contents of the different tissues in young and mature stems of plants growing at different conditions of water and light. We worked in Sierra de los Difuntos with plants that grow in dry and wet sectors with and without direct sunlight. Mature and young stems were collected in land and fixed in FAA, then free hand sectioned and later observed with optical microscopy.

Young stems from wet sectors with sunlight presented a greater amount of cells with TLS in perimedullary zones and epidermis than ones in the dry sectors with sunlight. The stems from wet sectors with shadow presented a greater amount of cells with TLS in pith than ones in the wet sectors with sunlight and ones in the dry sectors. In mature stems from wet sectors we observed a greater number of cells with TLS in pith and inner phloem compared with the dry sectors. Young stems from wet sectors presented more cells with TLS respect to the dry sectors. In every analyzed tissue the amyloplasts content in pith and cells with TLS increases with the stem maturity.

124. ERITHROCYTE DEFORMABILITY ANALYSE AND NA⁺/LI⁺ COUNTERTRANSPORT IN HIPERTENSION

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The purpose of the present work was to study the rate of Na⁺/Li⁺ countertransport and erythrocyte deformability in patients with essential hypertension (HTA) (n=12), and normal subjects (NS) (n=11). The Km (mM) and Vmax (mmoles l⁻¹ cell h⁻¹) values were determinated and then the efflux (30min) for the maximum counter lateral concentration of Na⁺ (CCT) was identified. Red blood cells morphology was evaluated by the EMI (Erythrocyte Morphology Index) defined as the relationship between bowl and discocyte forms.

SUBJECT	NORMAL	HTA
Km (mM)	4.959	8.891
Vmax mmoles l ⁻¹ cell. h ⁻¹	0.3230	0.1014
CCT mmoles l ⁻¹ cell.	0.2985	0.0868
EMI	2.13±0.15	0.91±0.05

The Vmax and CCT values were significantly different (p<0.002, U Mann-Whitney). The EMI values were significantly smaller in hypertensive patients than normal subject (p<0.001). The cell morphology is influenced both by extracellular and cellular factors which might connected with the structural conformation of the cell membrane. The decreased EMI reveal alterations in the dephormability which could be associated to the countertransport Na⁺/Li⁺.

125.

THE INFLUENCE OF ENVIRONMENTAL AGENTS AS MODERATORS ON BUCAL HEALTH. ANALYSIS OF A SAMPLE OF PREHISTORIC ADULTS OF “LAGO SALITROSO”

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The aim of this paper is to verify the pressure put on the masticatory apparatus by certain environmental agents such as consuming water and solid diets with fluoride. Ten dentitions were analyzed on human remains of prehistoric adults, original inhabitants of the watershed of Lago Salitroso in Sierra Colorada, Provincia de Santa Cruz. The features which supplied more information about the influence of the above mentioned environmental agents, were the low prevalence of cavities, the remineralization of the enamel and the abundance of dental hypoplasias (white spots, pits, lines, brownish teeth). The fluoride in some components of the solid diet, as well as in consuming water influences the crystal structure of the enamel, stopping the increase of the carious lesions or remineralizing grooves and fissures. The excessive consumption of fluoride during the preeruptive stages alters the structure of the enamel inhibiting the action of the ameloblasts, favouring the production of faulty enamel. The study of the traces was carried out by direct observation with natural light. For the detection of remineralized cavities, grooves and fissures, and for the specific diagnose of the hypoplasias were used calibrated probes and binoculars magnifiers. Cavities without remineralized or partially remineralized (active cavities), cavities totally remineralized and remineralization without cavities were considered. Endemic hipoplasias reveal that fluorine was present in high concentrations and that it was consumed during the first five years of life, when enamel is formed. Detections of dental remineralizations would show fluoride consumption in diets for long steady periods after five years of age. The bucal health of this population would therefore be due to the compensatory balance between the action of the fluoride against the aggression of the cariogenic components; and not only due to a diet with limited elements productive of caries.

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