BIOCELL 2008, 32(3): A1- A28 ISSN 0327 - 9545 PRINTED IN ARGENTINA



ROSARIO BIOLOGY SOCIETY

(SOCIEDAD DE BIOLOGÍA DE ROSARIO)

Abstracts from the

IX CONGRESS XXVII ANNUAL MEETING

November 29 - 30, 2007

Facultad de Ciencias Bioquímicas y Farmacéuticas Universidad Nacional de Rosario Rosario, Santa Fe, Argentina

Abstracts were revised by a Scientific Committee

MECANOREGULATION CELLULAIRE : IMPORTANCE EN INGENIERIE TISSULAIRE MECHANOREGULATION OF CELLS: IMPORTANCE IN

TISSUE ENGINEERING

Stoltz $JF^{1,2}$, Decot $SV^{1,2}$, Kadi A^1 , Muller¹, Menu P^1 , Traore M^1 , Bensoussan D^2 .

¹Ingénierie Cellulaire et Tissulaire UMR CNRS 7563 - Faculté de Médecine - 54500 Vandoeuvre lès Nancy, France. ²UTCT-CHU-54 500 Vandoeuvre Lès Nancy, France.

Almost all of the cells of the human body are subjected to mechanical stresses. In endothelial cells, mechanical stresses can vary from some Pascal (shear stress) to some-hecto Pascals. Now it is know that mechanical stresses have a decisive action in cellular physiology. However, if the main biological effects of mechanical force are well know, the mechanisms allowed the relation between mechanical stress to physiological phenomenon remain nearly unknown (mechanotransduction phenomenon). In this work, through personal results and published works, the authors considers some effects of mechanical stresses on vascular endothelial cells and chondrocyte and also as example the applications in cartilage engineering.

2. EFFECTS OF KINESIOTHERAPY ON AVERAGE LENGTH OF HOSPITAL STAY FOR PEDIATRIC PATIENTS WITH RESPIRATORY PATHOLOGIES IN A PUBLIC HOSPITAL Guadagnoli E¹, Vila H².

¹Kinesiology and Physiatry. UAI. ²School of Medicine. UNR. Email: hvila@fmedic.unr.edu.ar

Respiratory kinesiotherapy (RKT) comprises a series of procedures that complement and support the treatment of respiratory diseases. Diverse authors coincide in the positive results that RKT offers as far as health conditions of pediatric patients with respiratory pathologies are concerned. The aim of this study is to analyze how kinesiotherapy affects the average length of hospital stay for pediatric patients with respiratory pathologies compared with those who do not receive kinesiological treatment. The method used to evaluate this impact takes into consideration data obtained from clinical histories provided by Centenario Provincial Hospital. The information obtained is from the year 2000, when patients were not treated with RKT, and from the year 2004 when they were treated with RKT. During these years, therapeutic management did not change considerably. The population studied included 139 hospitalized children (72 in 2000 and 67 in 2004) with the following diseases: pneumonia, acute bronchitis and acute bronchiolitis. The average length of hospital stay for the 72 patients of the year 2000 (without RKT) was 9.4 days and for the 67 children of the year 2004 (with RKT) 5.4 days. It can be observed that the average length of stay was reduced 4 days with KTR Taking this variation into consideration and although these data is not statistically significant, it is concluded that RKT is a practice that helps reduce the length of stay for pediatric patients and therefore it provides relevant data when making decisions in clinical practices.

3.

SIALOPROTEINS AS MARKERS OF BONE FORMATION IN OVARIECTOMIZED RATS

Chapo G, Fernández M del C, Morosano M, Tarrés MC, Montenegro S, Caferra D.

Cát. Química Biológica. Fac. Cs. Médicas. UNR. E-mail: gfchapo@hotmail.com

Sialoproteins (Sip) play an important role in ossification, so it should be possible to consider them as a biochemical marker of bone formation. A decrease in bone density during menopause results from an imbalance between resorption and bone tissue formation, the former prevailing. Ovariectomy in rats leads to a similar effect. Objective: to determine whether resorption increase is accompanied by a decrease in bone formation, using Sip as marker and ovariectomized (Ox) rats as model. Gonadectomy was performed to Sprague Dawly rats 90 days old (Ox, n=18). Others were kept intact (control, n=18). Animals were killed 2, 30, 60, 90 and 150 days after castration, also controls of the same age were killed. An exhaustive extraction of bone dust Sip was performed (tibiae and femora); associated sialic acid was determined. Results: values in mg. Sialic/g bone (average \pm DS): <u>Controls</u>: 2.03 \pm 0.3 (n=3); 1.66 \pm 0.09 (n=3); 1.68 \pm 0.05 (n=3); 1.25 ±0.03 (n=4); 0.79 ±0.12(n=5) and Ox.:1.84 $\pm 0.38(n=3); 1.33\pm 0.06(n=3); 1.14\pm 0.07(n=3); 1.02\pm 0.09(n=6); 0.67$ ± 0.04 (n=3). In both groups there was a inverse relation between age and sialic acid content. When comparing animals 120 (p=0.05) and 150 (p=0.046) days old (30 and 60 Ox days, respectively), using the Mann Whitney non parametric test, values were lower in Ox than in control while no significant differences were found in the other ages. The analysis of Sip vs. time showed gradual decrease in controls, while in Ox an abrupt decrease was noticed 30 days after surgery and later, a sloping fall which is similar to the controls. Conclusion: Sip decrease could be associated to a fall in bone formation in osteoporosis induced by a lack of ovarian hormones.

4.

ASYMPTOMATIC VERTEBRAL FRACTURES (VFx) AND KYPHOSIS IN OSTEOPOROTIC AND NON-OS-TEOPOROTIC POSTMENOPAUSAL WOMEN

Morosano M, Tomat MF, Bocanera R, Tozzini R, Masoni A. Centro de Estudios del Climaterio (CEC), Hospital Provincial del Centenario y Cátedra de Química Biológica, (FCM), UNR.

Objective: to determine asymptomatic VFx prevalence and clinical and radiological kyphosis in menopausal women; to assess the association between wedge vertebrae and clinical kyphosis. Anamnesis, physical exams, thoracic and lumbar X Rays (antero-posterior / lateral views), and DEXA hip bone densitometry were performed in 88 postmenopausal outpatients. Thoracic Spine radiological morphometry was performed to check wedge vertebral compression fractures (20% or + reduction in the anterior related to the posterior height in each vertebral body). Cobb's Angle and wedge vertebral angle addition (thoracic 4 - 12) were calculated. Variables: age, body mass index (BMI), Cobb's Angle, Wedge Angle Addition, VFx, Bone Mineral Density (BMD) and clinical kyphosis. Statistics: prevalence, average and standard deviation, chi square (χ^2), t-Student, and lineal regression. VFx prevalence was 34%. 23% of VFx patients had normal BMD or mild/moderate osteopenia according to WHO. Association between clinical Kyphosis and vertebral deformities was confirmed ($\gamma^2=8.03$, p<0.005). Cobb's Angle was 65.6°±13.8 in VFx patients (n=30), and 45.4°±14.3 in non-VFx patients (n:58); (t=6.43; p<0.001). Wedge angle addition is inversely associated to Hip BMD (r: - 0.36; p < 0.005). Age or BMI significant differences were not found. Wedge Vertebrae are associated with severe Osteopenia and Osteoporosis. 1 in 4 asymptomatic women with BMD values not usually considered for treatment, showed VFx. X-rays are as important as densitometry when prescribing studies.

5.

ASYMPTOTIC BODY WEIGHT AND MATURING RATE IN MICE FROM A F2 POPULATION DISCRIMINATED BY THEIR OF EARLY AND LATE GROWTH RATES

Silva PS¹, Zucchetti AE¹, Font MT^{1,2}, Di Masso RJ^{1,2}.

¹Instituto de Genética Experimental, Facultad de Ciencias Médicas; ²CIC-UNR.

The postnatal growth of the mouse can be partitioned in two phases: early pre-inflection and late post-inflection with apparently independent genetic controls. For characterizing both phases the rates of exponential increasing growth (exponential function, k1) and exponential asymptotic growth (Brody's function, k2) were estimated in 400 males and 400 females belonging to a F2 population derived from crossing two lines of mice with different mature body weight. The dispersion diagrams resulting from combining the individual values of both of growth rates for males and females separately were divided in four quadrants by means of two perpendicular lines that intersect both axes of coordinates at their mean values. Mice belonging to each quadrant (Q1: > k1 > k2; Q2: < k1 > k2; Q3: < k1 < k2; Q4: > k1 < k2) were identified and their body weight - chronological age longitudinal data were fitted by the Gompertz equation. The effect of quadrant localization on the parameters of asymptotic (A) body weight and maturing rate (k) for body weight was assessed by means of a one way (quadrant) analysis of variance. Differences among quadrants were more evident in the behavior of k that in A. The results reveal a greater effect of the different combinations of the partial exponential rates k1 and k2 on the overall maturing rate k than on asymptotic body weight A and an independent effect of pre and post-inflection growth rate on the maturing rate for the whole growth period.

6.

MEAN DOUBLING TIME OF A MAMMARY ADENOCAR-CINOMA IN TWO LINES OF INBRED MICE

Rico MJ^{1,3}, *Zacarías Fluck MF*¹, *Scharovsky OG*^{1,2}, *Font MT*^{1,2}, *Di Masso RJ*^{1,2}, *Rozados VR*^{1,2}.

¹Instituto de Genética Experimental, Facultad de Ciencias Médicas, ²CIC-UNR, ³CONICET.

Breast cancer is a complex disease that affects nearly 10% of women in industrialized countries. M-406 mammary adenocarcinoma appeared spontaneously in a female mouse of the CBi line of the Instituto de Genética Experimental. The aim of this work was to analyze the growth rate of M-406 in the line of origin and in an artificially selected derived line (CBi/L). Six males and six females of each genotype were s.c. challenged with M-406. Tumor volume was determined $[V = (minor \ diameter)^2 \ x \ major \ diameter \ x \ 0.4].$ Tumor growth rate was characterized in terms of tumor volume doubling time (0.69/K) estimated from fitting an exponential model Vt=Vi*exp (k*t), where Vt: tumor volume (mm3) at t days, Vi: initial tumor volume, k exponential growing rate, t: time (days). The effect of genotype (G), sex (S) and GxS interaction over mean doubling time was evaluated with a two-way ANOVA. A significant effect of G (p = 0.004) attributed to the smaller tumor growth rate in CBi/L was observed. Sex (p = 0.09) and GxS (p = 0.45) effects were not significant. However mean doubling time tended to be higher in males than in females (both lines), being this difference more evident in CBi/L (p = 0.08) than in CBi (p = 0.42). It is concluded that both lines bear genes involved in the susceptibility to the challenge with a mammary tumor and that CBi/L, derived from CBi, has fixed, by selection or genetic drift, genes that would explain the differential response to the tumor challenge that is evidenced by a slower growth rate and by differences between sexes.

EVALUATION OF PRECLINICAL RESULTS AND LIKELI-HOOD OF TRANSLATION TO THE CLINIC OF CANCER METRONOMIC CHEMOTHERAPY (MCT)

Rozados VR^{1*}, Mainetti LE^{1*}, Rossa A¹, Sánchez AM¹, Matar P¹, Berra HH², Gervasoni SI¹, Bonfil RD³, Scharovsky OG^{1,4}.

¹Inst. de Genética Experimental, ²Fisiología Humana, Facultad Cs. Médicas, ⁴Consejo de Investigaciones, U.N.R., Rosario. ³ Wayne State University School of Medicine and Karmanos Cancer Institute, Detroit, MI, USA. * Contributed equally.

MCT consists of a chronic, equally spaced, administration of low doses of chemotherapeutic drugs without extended rest periods. Our goal was to synthesize our experience with different kinds of rats and mice tumors treated with MCT to evaluate the likelihood of its clinical application. Cyclophosphamide (Cy) in low dose, three times/week i.p. (10 and 5 mg/kg for L-TACB rat lymphoma and S-E100 rat sarcoma, respectively) showed antitumoral effect (p<0.01). Also, tumor growth delay was achieved with low dose Doxorubicin i.p. or Cy (i.p. or p.o.) in mammary adenocarcinomas M-406 and M-234p bearing mice (p<0.05). Metronomic administration of Cy (25-30 mg/kg/day in the drinking water) plus Celecoxib (30 mg/kg p.o. 5 times/week) inhibited tumor and metastasis growth and increased survival in M-406 and M-234p tumor-models. Vascular endothelial growth factor serum levels were lower in treated animals than in controls (p<0.01). Also, CD34⁺ vessels were lower in mouse treated tumors than in controls, showing its antiangiogenic effect. The scarce response to MCT with Cy in L-TACB bearing nude mice indicated the involvement of the immune response in the antitumoral effect. There were no general, renal, hepatic, hematologic or cardiac toxicities. These results suggest the implementation of MCT at the clinical setting in the near future, increasing survival and life quality of patients.

8.

MODULATION OF GALECTIN-1 (GAL-1) EXPRESSION BY THE IMMUNE SYSTEM IN THREE MURINE TUMOR MODELS

Zacarías Fluck MF*, Rico MJ*, Rozados VR, Rabinovich GA, Scharovsky OG.

Instituto de Genética Experimental, Facultad de Ciencias Médicas, Universidad Nacional de Rosario, Rosario, Argentina (MFZF, MJR, VRR, OGS). Laboratorio de Inmunopatologia, IBYME-CONICET, Buenos Aires, Argentina (GAR). * Contributed equally.

We have demonstrated that the kinetics of Gal-1 expression is different in tumors with different immunogenicity.. We also showed that Gal-1-induced immunosuppression is a mechanism used by tumors to evade the immune response (IR). We hypothesized that the kinetics of Gal-1 expression would depend on the relationship between tumor immunogenicity and host IR. The aim of this study was to validate the postulated hypothesis. Euthymic inbred e and m rats and Balb/c mice were s.c. challenged with L-TACB (rat lymphoma), S-E100 (rat sarcoma) and M-234p (mouse mammary adenocarcinoma). Simultaneously, nude mice were s.c. challenged with the same tumors. Rats and mice were sacrificed periodically. Primary tumors and spleen were excised, homogenates prepared and Gal-1 expression determined by Western Blot. Tumor growth rate in nude mice did not differ from that of euthymic hosts in the three tumor models. Gal-1 expression was lower in nude mice than in euthymic hosts (p<0.05) during tumor growth. Splenic Gal-1 was only quantifiable in euthymic hosts. Since Gal-1 expression in nude mice remained constant or decreased during tumor growth, we conclude that the antitumor IR strongly modulates Gal-1 expression. Hence, the tumor would regulate Gal-1 expression according to the cross-talk established with the Immune System.

INTESTINE HISTOLOGIC CHANGES ASSOCIATED TO NATURAL ENTEROPARASITOSIS (*NE*) IN MICE OF DIF-FERENT GENOTYPES FROM THE CBI STOCK

Buccalossi G^1 , Gorosito M^2 , Valenti J^2 , Vasconi MD^3 , Di Masso $R^{1,4}$, Font $MT^{1,4}$, Hinrichsen $L^{1,4}$

¹IGE, ²Anat Pat, FCM; ³Parasit, FCByF; ⁴CIC-UNR, U.N.R.

A chronic parasitic infection constitutes a balanced ecologic system in which host and parasite are controlled by a complex group of interacting forces that result in the host developing resistance, susceptibility and/or resilience to the infection. The analysis of NE in lines of mice of the CBi-IGE colony showed qualitative and quantitative disparities, evincing genetic differences among lines in the host - parasite relationship; yet, no symptoms and/or pathology due to the parasites were observed. To determine whether the parasite type or burden induced changes in the intestine, even in the absence of clinical manifestations, its histology was studied in mice of each genotype. Adult males and females from lines CBi+ (+), CBi- (-), CBi/L (L) and CBi/C (C) were examined. As expected, only (C) mice were susceptible to Trichomonas muris while (-) was the less resistant genotype to Spironucleus muris. The helminth burden (Syphacia obvelata and Aspiculuris tetraptera) showed differences between sexes within genotypes (males had higher burdens than females) and among genotypes (L males had the lowest burden). The histology of the small and large intestine did not show alterations that could be ascribed to any specific host (genotype x gender) - parasite pair. Some mice presented a decrease of the cytoplasmic mucin in the superficial intestinal epithelium, associated to helminths. The inflammatory infiltrate in the lamina propria, the lymphoid follicles of the small and large intestine as well as the villi of the small intestine did not show important modifications. These results suggest that host resilience, in this model, is independent of the parasite type or burden.

10.

RESISTANCE/SUSCEPTIBILITY (R/S) TO NATURAL ENTEROPARASITOSIS (*NE*) IN PRE-PUBESCENT MICE OF THE CBI STOCK

Vasconi MD^2 , Buccalossi GG^1 , Di Masso $R^{1,3}$, Font $MT^{1,3}$, Hinrichsen $L^{1,3}$.

¹IGE, FCM; ²Parasitología, FCByF; ³ CIC-UNR, U.N.R.

In a conventional animal facility mice are exposed to infective agents capable of contaminating them. Enteroparasites are among these "natural pathogens". The genotype of the host is one of the factors responsible for the progression of the parasitic infection because it delimits the host-parasite interactions. The analysis of NE in adult mice of the CBi-IGE colony showed significant differences among lines, attributed to the genotype of the hosts. Our aim was to analyze NE in pre-pubescent CBi-IGE mice to determine whether the qualitative and quantitative differences seen in adults were also observed in young mice. NE and nitric oxide (NO), a mediator of the immune response, were studied in thirty days old males and females from lines CBi+ (+), CBi- (-), CBi/L (L), CBi/C (C) and CBi. Trichomonas muris was found parasitizing line (C) and a low proportion CBi mice (p<0.01); lines (-) and CBi showed a high proportion of animals infected with Spironucleus muris while (+), (L) and (C) had a lower percentage (p<0.01). At this age the parasitic burden (PB) was high and independent of the percentage of infected animals. The helminth (Syphacia obvelata and Aspiculuris tetraptera) PB only showed differences among genotypes (p<0.001), males had a higher PB than females, except CBi. NO was significantly lower than in adults (p<0.05), except in CBi females. It can be concluded that the genetic differences in R/S to NE are expressed early in development, in this model. The sex differences observed in adults were unapparent in 30 days old mice. As in adult mice, there was no association between the degree of the infection and the serum levels of NO.

11.

BREAST PROPORTION IN TWO THREE WAY EXPERI-MENTAL HYBRIDS OF FREE RANGE BROILERS: TRANS-VERSAL ANALYSIS

Advínculo S¹, Librera JE¹, Álvarez M¹, Canet ZE¹, Font $MT^{2,3}$, Dottavio $AM^{1,3}$, Di Masso $RJ^{1,2,3}$

¹Cátedra de Genética, Facultad de Ciencias Veterinarias; ²Instituto de Genética Experimental, Facultad de Ciencias Médicas; ³CIC-UNR. E-mail: rjdimasso@ciudad.com.ar

The meat conformation of two experimental three way hybrids of free range broilers proposed as alternative to Campero INTA was characterized. Breast proportion at slaughter of Casilda Doña Teresa [Rhode Island Red males x CP (Cornish Blanco x Plymouth Rock Barrado) females], Casilda Don Manuel [Plymouth Rock Barrado males x CR (Cornish Blanco x Rhode Island Red) females] and Campero INTA (reference genotype) was weekly determined between the 1st and 11th week of age. The effect of age, genotype and (age x genotype) interaction was evaluated by means of an analysis of variance corresponding to an 11x3 (eleven ages x three genotypes) factorial experiment. A significant effect of age (F = 11.1, P < 0.0001) indicative of an increase in breast proportion as the chicken reaches the target slaughtering weight was observed. Non significant effects of genotype (F = 0.14, P = 0.86) and genotype x age interaction (F = 1.07, P = 0.38) were seen. Hybrids with 25% of White Cornish genes, proposed as an alternative to Campero INTA, do not differ from the latter in breast proportion indistinctly of the age at which comparisons are made along the breeding cycle.

12.

GROWTH DYNAMICS OF EXPERIMENTAL HYBRIDS OF FREE RANGE BROILERS

Álvarez M¹, Dottavio AM^{1,3}, Antruejo AE¹, Galvagni A¹, Font MT^{2,3}, Di Masso RJ^{1,2,3}

¹Cátedra de Genética, Facultad de Ciencias Veterinarias; ²Instituto de Genética Experimental, Facultad de Ciencias Médicas; ³CIC-UNR. E-mail: rjdimasso@ciudad.com.ar

With the aim of characterizing the growth pattern of two experimental hybrids of free range broilers: Casilda Doña Teresa [Rhode Island Red males x CP (Cornish Blanco x Plymouth Rock Barrado) females] and Casilda Don Manuel [Plymouth Rock Barrado males x CR (Cornish Blanco x Rhode Island Red) females], and of the reference genotype (Campero INTA), body weight and shank length behaviour in relation to chronological age were studied. Longitudinal body weight/age and shank length/age data were adjusted with Gompertz and logistic models respectively. The effect of genotype on the estimates of A and k parameters in each model was evaluated with a one way analysis of variance. Campero INTA showed a higher asymptotic size (p<0.05) (Mean \pm SEM – Campero: 4955±137; Casilda Doña Teresa: 3427±74; Casilda Don Manuel: 3815 ± 129) and a faster speed to reach it (p<0.05) (0.2163\pm0.0052; 0.1911±0.0028; 0.1850±0.0037, respectively), and an equal final shank length (Campero: 15.3 \pm 0.32; Casilda Doña Teresa: 15.1 \pm 0.22; Casilda Don Manuel: 14.9 ± 0.34) and a faster speed to reach it(p<0.05) (0.1780±0.0041; 0.1475±0.0030; 0.1515± 0.0037, respectively) than the alternative hybrids. Even though the higher adult biomass of Campero INTA chickens was not accompanied with a higher skeletal development (estimated from shank length) indicating a certain imbalance between biomass to support and supporting bone structure, the faster speed of approximation to mature body weight (high k) showed a favorable modification of the maturity rate for shank length.

ABDOMINAL FAT IN EXPERIMENTAL HYBRIDS OF FREE RANGE BROILERS WITH CORNISH GENES: TRANSVER-SAL ANALYSIS

Martines A^1 , Romera BM^1 , Dottavio $AM^{1,3}$, Font $MT^{2,3}$, Di Masso $RJ^{1,2,3}$

¹Cátedra de Genética, Facultad de Ciencias Veterinarias; ²Instituto de Genética Experimental, Facultad de Ciencias Médicas; ³CIC-UNR, U N de Rosario. E-mail: rjdimasso@ciudad.com.ar

The behaviour of abdominal fat content [(fat weight/eviscerated body weight) x 100] in males of two three-way experimental hybrids: Casilda Doña Teresa [Rhode Island Red males x CP (Cornish Blanco x Plymouth Rock Barrado) females] and Casilda Don Manuel [Plymouth Rock Barrado males x CR (Cornish Blanco x Rhode Island Red) females] and in the reference genotype (Campero INTA) was studied after serial slaughtering between the first and the eleventh week of age. The effects of age, genotype and (age x genotype) interaction were studied by means of an analysis of variance corresponding to an 11x3 factorial experiment (eleven ages x three genotypes). A significant effect of age (F=3.09; P=0.001) and genotype (F=12.36, P<0.0001) due to a higher fat proportion in Campero INTA was observed. A non-significant effect of the genotype x age interaction (F = 1.29, P= 0.20) was seen. Results show that the inclusion of White Cornish genes in the hybrids proposed as an alternative to Campero INTA for semi-intensive systems does not show unwanted effects on body fat deposition estimated by abdominal fat percentage. Such response would be independent from variations observed in the fat content of poultry with age.

14.

HISTOPATHOLOGICAL LESIONS PRODUCED IN THE BRAIN OF INOCULATED RABBITS WITH BOVINE HER-PESVIRUS 1 AND 5

Pidone CL¹, Galosi CM^{2,3}, Poli GL¹, Brion C¹, Anthony LM¹, Riganti JG¹.

Faculties of Veterinary Sciences ¹National University of Rosario and ²National University of La Plata; ³Researcher of Scientific Research Commission, Buenos Aires Province, Argentina.

Some researchers demonstrated that Los Angeles strain, bovine herpesvirus 1 (BHV-1), could induce neurological signs in 30-35 days old rabbits after the intranasal inoculation, indistinguishable of those induced by bovine herpesvirus 5 (BHV-5). However, the histopathological study revealed vasculitis and thrombosis, focuses of neuronal necrosis in brain although they were not compatible with encephalitis, suggesting that the nervous lesions were probably consequence of the tissue anoxia. The objective of this investigation was to characterize the type of lesions that BHV produced in brain of 30-35 days old rabbits after the intranasal inoculation and to determine if there are differences among those that are produced by strains of BHV-5 and by BHV-1. Los Angeles strain (BHV-1) and Argentinean strains A663, A178 and A54 (BHV-5) were used. When the animals showed nervous signs they were euthanized, necropsied and brain samples were introduced in formol 10% and processed through graded alcohols and xylene before being embedded in paraffin wax. Sections (5 µm) were cut and stained with haematoxylin-eosin. The histopathological exam revealed non purulent meningoencephalitis, with multifocal gliosis and perivascular cuffing by mononuclear infiltration in all cases. We concludes that the neurological signs are a consequence of the lesions mentionated and they can be produced both BHV-1 and BHV-5.

15.

PHENOTYPES RESISTANT TO MACROLIDES IN ISO-LATED *Staphylococcus* spp. FROM CANINES WITH DER-MATITIS. PRELIMINARY RESULTS

François S, Tártara G, Marro AV, Sarradell J, Anthony L. Fac. de Cs. Veterinarias, UNR. Bv. Colón y Bv. Spangemberg (2170) Casilda. E-mail: silvinafrancois@uol.com.ar

Dermatitis in dogs is caused mainly by Staphylococcus intermedius. To determine antimicrobial sensitivity, betalactamic antibacterials are used, however, habitual to find resistant strains, so macrolide antibacterials are also used. The aim of this work is to study resistance phenotypes to microlides, lincosamides and streptogramins B of Staphylococcus spp. isolated from dogs with dermatitis. Twenty isolates of Staphylococcus spp. resistant to erythromycin from dogs with dermatitis were analyzed. They were identified through conventional biochemical tests. Sensitivity determination was performed through an antibiogram with agar diffusion according to NCCLS. A two plate test was performed placing an erythromycin plate (ery) at 2 cm from a clydamycin plate (cly). The phenotypes analyzed were: constitutive resistance phenotype, MLS; inducible resistance phenotype, MLS; macrolide resistant phenotypes, M; and lincosamides resistant phenotypes, L. Sixteen isolates of S. intermedius and four of negative coagulase Staphylococcus (NCS) were identified. Twentyfive percent of S. intermedius isolates showed a MLS phenotype, 69% an MLS and 6% an M phenotype. Among NCS, three showed an MLS and 1 M phenotype. These results lead us to conclude that within the studied population isolates show mainly the MLS phenotype. This phenotype indicates crossed resistance between ery and cly, making it important to constantly monitor the various resistant phenotype profiles of staphylococci from canine.

16.

EFFECT OF VEGETAL EXTRACTS ON EQUINE RED BLOOD CELL MEMBRANE DEFORMABILITY AND FLU-IDITY

Catalani G¹, Dottavio ME², Miglietta MC³, Spengler MI², Bertoluzzo SM², Rasia M².

¹Fisiología, FCV, UNR. ²Física Biológica. FCM, UNR. ³Anatomía Veterinaria. FCV, UNL. E-mail: galena@arnet.com.ar

Erythrocytes (Es) are specially adapted to microcapillary circulation. Their deformability capacity is based on several factors, i.e., membrane fluidity (MF), cytoskeleton, surface/volume ratio, & cytoplasm viscosity. In thorough bred horses (TBH) different degree of training induced nasal hemorrhage (TINH). Clinical improvement occurred after treatment with vegetal lysates. Objective: EDTA anticoagulated whole blood TBH samples (n=10) treated with VL were withdrawn to study erythrocyte deformability (ED) and MF. Controls: Healthy TBH healthy, same managing and feeding. Samples were obtained 10 days before & after oral treatment with 50 ml 30% LV hydro-alcohol crude extract, from Portulaca sp (Portulacaceas) foliar parenchyma, 30% Agavia sp (Liliaceas) & 40% Opuntia sp. (Cactaceas), & 5% dry residue. ED expressed as its inverse, rigidity index (RI): E filtration suspension through 5 µm diameter polycarbonate membrane/medium E suspension ratio. MF (fluorescence polarization): Es were labeled with fluorescent TMA-DPH (Trimethylammonium-Diphenylhexatriene). Anisotropy coefficient (r), inversely related with MF was calculated. Statistics: Wilcoxon's test for paired samples. Results (median; range): show RI decrease (p<.05) before [5.85 (2.70-17.80)] & after treatment [4.42 (1.73-10.80)]; r decreased with VL: before [.205 (.143-.206)] & after [.180 (.131-.204)] treatment. Conclusion: VL treatment produces E deformability & MF increase, enhancing their capillary circulation.

DYNAMIC BEHAVIOR OF HEMATOCRIT IN HYBRIDS OF FREE LINE RANGE BROILERS

Fernández R¹, Rozados VR^{1,2}, Font MT^{2,3}, Dottavio AM^{1,3}, Di Masso RJ^{1,2,3}.

¹Genética, FCVeterinarias; ²Instituto de Genética Experimental, Facultad de Ciencias Médicas; ³CIC-UNR.

In commercial broiler chickens modifications of the packed cell volume or hematocrit (H) represents one of the adaptative responses to the high necessity of oxygen imposed by the increased metabolic demands that the intensive selection for rapid growth has fixed in these chickens. Campero INTA is a meat type chicken, with a growth rate lower than that of the commercial broiler strains. As White Cornish is the breed used as the paternal genotype in the commercial broilers, the aim of this work was to evaluate the behaviour of H in males of two experimental hybrids of free range broilers with 25% of White Cornish genes and the reference genotype (Campero INTA). The two hybrids were: Casilda Doña Teresa [paternal Rhode Island Red x maternal Casilda CP (Cornish White x Barred Plymouth Rock]; Casilda Don Manuel [paternal Barred Plymouth Rock x maternal Casilda CR (White Cornish x Rhode Island Red)]. H was determined weekly (1st-11th week of life) in samples collected before euthanasia. Values of H were significantly affected by age (P<0.0001), since H showed oscillations in time, and by genotype x age interaction (P=0.032) because each group behaved differently with age. There were no significant effects of genotype (P=0.098), though the experimental hybrids showed a lower H than the Campero INTA, a result similar to that observed in growth rate (Casilda Doña Teresa<Casilda Don Manuel<Campero). We conclude that inclusion of Cornish genes in these hybrids does not produce a metabolic stress, as evaluated by measure of H value.

18.

CONTAMINATION OF WORKERS PRESENT IN PRODUC-TION LOTS WHILE PHYTOSANITARY PRODUCTS ARE APPLIED

Panelo M, Bulacio L, Giuliani S, Giolito I.

Facultad de Ciencias Agrarias UNR. CC 14 (S2125ZAA) Zavalla, Pcia. Santa Fe, Argentina. E-mail: invest-agr@unr.edu.ar

The objective of the work was to determine on the bodies of the personnel present at the time of a foliar treatment with phytosanitary products, the areas where the residues would deposit considering the position and distance of the workers from the operator applying the product. The work was on a French bean (Phaseolus vulgaris L.) crop, with plants 2,20 m high, set out at 0,70 m between rows and plants, staked with cane. Four foliar applications of phytosanitary products were simulated using a manual backpack sprinkler and replacing the agrochemical product for a phenolphthaleine solution (0,5 gr.l⁻¹). Each block of treatments was made of 2 persons to the N and 2 persons to the S, placed in the rows next to those where the application was being performed, one 0,70 m and the other 1,40 m away from the operator applying the product. Previous to each treatment, their bodies were covered with 23 white 10 cm x 10 cm cotton patches. At the laboratory, the residue of colorant was recovered with sodium hydroxide 0,1 N and it was valued in a spectrophotometer at 540 nm. The statistical design was of random complete blocks, with 4 repetitions. An average of 57,55 µg.cm⁻² of colorant was detected on the operators, distribuited as per patches groups 11,99% on the head, 20,94% on the torso and abdomen, 35,19% on upper limbs and 31,88% on lower limbs. These preliminary results showed the risk of exposure for the operators that stay near the areas being treated, a practice that should be advisable to prohibit in horticultural systems.

19.

IMIDAZOLINONE RESISTANCE (IMI-R) EVALUATION IN SUNFLOWER HYBRIDS AND LINES AT SEEDLING STAGE Breccia G, Accastello C, Ceccarelli N, Maiocco V, Spirolazzi M,

Vega T, Nestares G. Cátedra de Genética. Facultad de Ciencias Agrarias. UNR. CC14 (S2125) Zavalla.

The IMI-R in sunflower is controlled by two genes and can only be achieved when both loci are in the homozygous state. The objective of this work was to evaluate IMI-R during development of plantlets of six sunflower genotypes. Three inbred lines differing for their IMI-R: susceptible (S), intermediate (I) and resistant (R) and the crosses beetwen them were evaluated. Achenes were seeded in speedlings filled with pearlite and watered by capillarity with a 25% strength MS saline solution (Murashige and Skoog, 1962) with different doses of imazapyr: 0 (control)-5-7,5-10 uM. For each treatment 2 replications of 5 seeds each were incubated under controlled conditions for 8 days. The variables hypocotyl and root length, aerial and root fresh and dry weight were analysed through ANOVA. The R, RxI and RxS genotypes did not present significant differences among control and herbicide treatments for any variable. Conversely, for S line mean values significantly diminished as imazapyr doses increased for all variables. Moreover, S plantlets grown in presence of herbicide showed short tip-necrosed roots and no secondary roots. Principal root of I and SxI genotypes elongated but secondary roots were poorly developed in the presence of imazapyr 10 uM. It is concluded that different resistance degrees can be distinguished through this method. This test could be useful to assist conventional breeding programs for the development of herbicide resistant hybrids.

20.

GENOTYPE X ENVIRONMENT INTERACTION IN EXPERI-MENTAL ASPARAGUS HYBRIDS BY BIPLOT ANALYSIS

Espósito MA^{2*} , López Anido F^1 , Milanesi L^2 , Martin E^1 , Cravero V^2 , Cointry E^2 .

¹Genética. ²Mejoram. Veg. Cs. Agr.- UNR. CC14 S2125ZAA.*Email: mesposi@unr.edu.ar

Genotype x Environment (GxE) interaction evaluation allows the knowledge of the performance of different genotypes in several environments. In asparagus the system of production (white or green asparagus) as well as the period of harvest (1st and 2nd year), can be considered as different environments. 34 experimental hybrids were evaluated in a DBCA design with three replicates. Two productive traits were considered: total yield (total weigh of the harvested spears, in g) and market yield (total weigh of spears which diameter overcomes 12 mm for white asparagus and 10 mm for green ones). ANOVA and (GxE) analysis were carried out using GGEbiplot software. Significant differences (P<0,001) between genotypes, environments and (GxE) interaction were found. A polygon with the hybrids 4, 6, 22, 30, 34 and 28 was conformed. These hybrids showed the best and the worst performances according to their location. Hybrids 4, 17, 16, 23, between others, showed high performance for white and green production in 2ndyear. The environment "green production in 1st year" was the fewer discriminate. Hybrids 2, 18, 13 shown the upper stability but the lowest yields. Biplot analysis allowed to identify hybrids with high performance and stability in different environments, as well as, to determine the most favorable environments for cultivars identification.

21. SAPLING BANK DESCRIPTION AND COMPARISON IN THREE TYPES OF FORESTS, FORMOSA-ARGENTINA

Sender MB, Maturo H, Prado D, Barberis I, Lewis J. Botánica. Fac. Cs. Agrarias, UNR, Zavalla.

E-mail: belsender@yahoo.com.ar

E-mail. beisender @yunob.com.ur

The forests in eastern Formosa province, Humid Chaco District, are determined by a climatic and topographic gradient, generating a high floristic and physiognomic heterogeneity of plant communities. In south-east Formosa, three types of forests are distributed forming a coenocline spreading from riparian ridges of fertile land to topographically lower, floodable areas (Riparian Forest-RF, Transitional Forest-TF, Hardwood Forest-HF). Abundance, richness, diversity and floristic composition of the saplings in these three types of forests were compared, establishing the Indicator Value (IV) of saplings species. Five 50 m long transects were established in each forest. Saplings of woody species were identified and recorded. The total sapling richness and diversity, at forest scale, were compared using rarefaction curves; whereas these parameters in plots were compared using ANOVA. Differences in the floristic composition were evaluated using permutation methods (MRPP). An analysis of species Indicator Value was done using a PC-ORD programme. Differences in total richness and diversity in the forests were not found. Abundance in RF was higher than that in HF and richness in RF was higher than that in TF and HF, at plot level. Sapling floristic composition was different in the three forests. Myrcianthes pungens, Piper amalago, Brunfelsia australis, Trichilia catigua, Holocalyx balansae, Trichilia elegans, Capparis flexuosa, Patagonula americana and Syagrus romanzoffiana species were RF indicators. Aspidosperma quebracho-blanco was an HF indicator and Eugenia moraviana and Acacia praecox were TF indicators.

22.

INTRODUCTION TO THE STUDY OF *CENTROSPERMAE* IN THE SANTA FE PROVINCE: *PORTULACACEAE*

Galetti L, Oakley LJ, García RCA, Prado DE.

Cátedra de Botánica, Facultad de Ciencias Agrarias (UNR), C.C.Nº14, S2125ZAA Zavalla.

The object of the present contribution is to start the taxonomical study of the Family Portulacaceae for Santa Fe. Placed in the order Centrospermae (=Caryophyllales), it includes 20 genera and about 500 cosmopolitan species. In Argentina is represented by 11 genera and 66 species, out of which 14 endemics. They consist of succulent plants, usually herbaceous, rarely shrubs or subshrubs. Leaves alternate or opposite; the flowers are actinomorphic, perfect, solitary or in racemose inflorescences, hypogynous or semiepigynous. Fruit capsule with transversal or longitudinal dehiscence. The methods consisted in field observation, analysis of plant collections (Herbaria SF, SI and UNR) and reviewing existing literature. Our preliminary results show that the family is represented in Santa Fe by two genera. These are the genus Portulaca L. with eight species and two varieties: P. amilis Speg., P. cryptopetala Speg., P. fluvialis D. Legrand, P. gilliesii Hook., P. grandiflora Hook., P. mucronulata D. Legrand var. mucronulata, P. mucronulata D. Legrand var. robusta D. Legrand and P. oleracea L.; and the genus Talinum Adans. with three species: T. fruticosum (L.) Juss., T. paniculatum (Jacq.) Gaertn. and T. polygaloides Gillies ex Arn. A distribution map, dichotomic keys for genera and species and some photographs are presented.

23.

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

Mogni V, Oakley LJ, García RCA, Prado DE.

Cátedra de Botánica, Facultad de Ciencias Agrarias (UNR), C.C.Nº14, S2125ZAA Zavalla.

The Lythraceae, in the Order Myrtales, is a tropical and subtropical family of over 25 genera and 450 species. It comprises herbaceous, shrubby or arboreal plants with opposite leaves, perfect flowers, actinomorphic or zygomorphic, solitary (axillary or interpetiolar) or in raceme. 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. Eight genera and 37 species are known to occur in Argentina, of which only four genera are present in Santa Fe. These are the genera Ammannia L., with two herbs: A. auriculata Willd. and A. latifolia L. founds in the north of the province; Cuphea P. Browne with three species: C. glutinosa Cham. & Schltdl., C. racemosa (L.f.) Spreng. subsp. racemosa -both very frequent- and C. laeviuscula Bacig. known only through the presence in the Dpt. Gral Obligado; the genus Heimia Link. with the 'quiebra arado' H.salicifolia (Kunth) Link has been collected, appearing in Santa Fe in areas under high anthropic activity and intensive cultivation; and Lythrum L. with two species: L. hyssopifolia L. and L. maritimum Kunth, both herbs typical of the wetlands in the Paraná River floodplain. Some authors refer the presence of Pleurophora saccocarpa Koehne, not confirmed yet.

A species distribution map, a brief description of each species, botanical ilustrations and a key for field recognition are provided.

24.

THE FAMILY *LEMNACEAE* IN THE PROVINCE OF SANTA FE (ARGENTINA)

Galetti L, Oakley LJ, Lusardi M, García RCA, Prado DE. Cátedra de Botánica, Facultad de Ciencias Agrarias (UNR), C.C.N°14, S2125ZAA Zavalla.

The object of the present contribution is to start the taxonomical study of the Family Lemnaceae for Santa Fe (Order Arales), which includes four genera and about 26 cosmopolitan species. In Argentina is represented by all its genera with 10 species. They consist of aquatic floating plants, with a flattened or globose shoot called 'fronde'. Flowers 1-2 per shoot, perfect or diclinous, achlamydeous. Fruit utricle with 1-5 seeds. The methodology consisted in observation in the field, analysis of plant collections (Herbaria SF, SI and UNR) and existing literature. Our preliminary results show that the family is represented in Santa Fe by two genera. These are the genus Lemna L. with two species: L. disperma Hegelm. and L. minuta Kunth; and Spirodela Schleid. with only one species: S. intermedia W. Koch. However, there exist unsupported citations for three genera: Wolffia Horkel ex Scheleid.-with two species: W. brasiliensis Wedd. and W. columbiana H. Karst.; Wolffiella (Hegelm.) Hegelm., represented by two species: W. lingulata (Hegelm.) Hegelm. and W. oblonga (Phil.) Hegelm.; and three additional species of the genus Lemna L.: L. aequinoctialis Welw., L. gibba L. and L. valdiviana Phil. A distribution map, dichotomic keys for genera and species and some photographs are presented.

FLORA OF SANTA FE (ARGENTINA): THE FAMILIES COMBRETACEAE AND MELASTOMATACEAE

Espiñeira M, Jordán V (ex-aequo), Bianchi MB, Oakley LJ, García RCA, Prado DE.

Cátedra de Botánica, Facultad de Ciencias Agrarias (UNR), C.C.Nº14, S2125ZAA Zavalla.

The closely related families Combretaceae and Melastomataceae are both placed in the Order Myrtales. The main characteristics are their actinomorphic, perfect, epigynous or perigynous flowers. The Combretaceae is a tropical and subtropical family of over 20 genera and 350 species; two genera and six species are known to occur in Argentina. It comprises shrubs, vines or trees with simple leaves, alternate or opposite, andromonoecious flowers, always epigynous, with apendicular hypanthium. The Melastomataceae is a tropical family with about 200 genera and 4500 species; seven genera and 23 species in Argentina. It comprises herbaceous, shrubby or arboreal plants with single and opposite leaves, flowers epigynous or perigynous with free or attached hypanthium. The object of the present contribution is to set off the taxonomical study of the both families for Santa Fe, through observation in the field, analysis of plant collections (Herbaria SF, SI and UNR) and revision of the literature. In Santa Fe both families are represented only for one genera and one species each: Combretaceae with the tree Terminalia triflora (Griseb.) Lillo called locally 'guayaibí amarillo'; and the Melastomataceae with the perennial herb or suffrutex Tibouchina gracilis (Humb. & Bonpl.) Cogn., both species known only through their presence in the Dpt. Gral Obligado. A species distribution map, botanical ilustrations and a key for field recognition are provided.

26.

DISCRIMINATING NINETEEN TOMATO GENOTYPES USING FRUIT QUALITY TRAITS

Mahuad SL^{1*}, Liberatti DR^{2*}, Marchionni Basté E^{2*}, Rodriguez GR², Pratta GR², Zorzoli R³, Picardi LA³.

¹FONCyT; ²CONICET; ³CIUNR; ^{*}ex-aequo. Cátedra de Genética, Facultad de Ciencias Agrarias UNR, CC 14 S2125ZAA, Zavalla, Argentina. E-mail: smahuad@unr.edu.ar

Several traits were measured in six recombinant inbreed lines (RILs, TOUNR1, TOUNR5, TOUNR8, TOUNR9, TOUNR15, TOUNR18), eleven hybrids among them, the cultivated parent (C), the wild genotype (PL) and the hybrid (CxPL). These traits were: Weight (W, g), Fruit Shelf Life (FSL, days until the first symptoms of deterioration and softening appeared), Soluble Solids content (SS, °Brix) and Titleable Acidity (TA, g of citric acid /100 g of homogenised juice). Field design was completely randomized $(N_{Enuit}=2660)$. A cluster analysis was performed to discriminate these genotypes (Ward method). The first discriminating trait was W (t=-51.0; p<0.01) segregating the ancestor C (average = 64.2g), from the rest of the genotypes (average=7.4g). The FSL and SS traits (t=5.3; p<0.01 and t=-2.4; p<0.01 respectively) allowed to segregate this last group in others two group. The first one including 6 genotypes with an average for FSL of 23.7 days and for SS of 6.7°Brix and the second group had an average of FSL=17.7 days and SS=7.7°Brix. PL and CxPL genotypes were included in this last group. Finally, this last group was subdivided (being AT the discriminating trait) into a new cluster that included ancestor PL, whereas the CxPL was integrated in another one. This analysis allowed to establish the similarity of these genotypes respect of these fruit quality traits.

27.

RELATIONSHIP BETWEEN BODY WEIGHT AND BODY CONDITION IN MAGRARIO FEMALES

Passarini M, Vilalta E, Ghirardi G, Kerps G, Ferrari S, Mahuad S, Amelong J¹, Picardi L².

Cátedras de Genética ex acquo. ¹Cátedra de Anatomía y Fisiología Animal-Ciencias Agrarias. UNR. ²CIUNR-Campo Experimental Villarino, Zavalla.

Magrario is a new genotype that was obtained by backcrossing the Ideal breed to Texel breed. Body condition (BC) is an indicator of the biomass state of the animal and it could be an useful tool to design strategies for food supply during the gestation period. The BC method is based on the evaluation of several animal body areas and these assessments could be influenced by the judge that are making them. A trial was conducted to test the degree of concordance between judges using 66 Magrario females during the beginning of the gestation period. Three groups were performed, Group A: an expert judge and others two (B and C) with undergraduate students that were trained by this expert. Evaluations made by each group were compared by the non parametrics Kendall test (W). The W was 0.86 $(\chi^2 = 168, p < 0.001)$ showing the high degree of concordance between groups. A correlation coefficient was estimated between the BC and female weight obtained at the same moment. The following results were obtained: Group A, r=0.32; Group B, r= 0.48 and Group C, r=0.47: All of them were highly significant (p<0.01). The Fisher tests applied to compare these coefficients were no significant indicating the similarity of these correlationst. These results suggested that EC method could be appropriate to evaluate Magrario mothers state during the gestation period. Consequently it could useful to define the right food supply.

28.

VARICOCELE. EVALUATION OF THE POST SURGERY SPERM RECOVERY

Bouvet BR¹, Paparella CV¹, Feldman RN².

¹Area Química Analítica Clinica. Fac. Cs. Bioq y Farm. ²Serv Rep. Fac. Cs Méd. Universidad Nacional Rosario UNR. E-mail: beatrizbouvet@arnet.com.ar

The varicocele is a frequent pathology in the centres of male reproductive medicine. Our aim was to study in infertile patients with varicocele the seminal parameters in pre and post surgical correction. A retrospective study was carried out in 39 men between 22 and 35 years old with diagnosed varicocele through usual techniques, they undergone a corrective surgical and were evaluated after a year with a sperm study. The patients with other pathology able to alter the seminal quality were discarded. A sperm study according to WHO (1999) was carried out. The sperm mobility, viability and concentration were researched with subjective methods using optical microscope; the morphology with Papanicolaou. The statistic analysis of the pre (X_{pre}) and post (X_{post}) surgical variables was determined with the *t* Student Test. Significant differences were found (p<0.05) in progressive mobility (p=0.0454), sperm morphology (p=0.0004) and sperm viability (p=0.03914). There wasn't a significant statistically difference in the sperm concentration (p=0.3894). The results show that the sperm mobility, viability and morphology are altered in patients with varicocele but they change to reference values (WHO) after one year's surgery. The concentration of spermatozoids is the seminal parameter less altered by the varicocele. Although this pathology damages the reproductive capacity, it offers a simple treatment with excellent results in male infertility.

29. PROSTATIC ACID PHOSFATASE ACTIVITY IN BASAL SEMINAL PLASMA AND POST EXPOSITION TO HIGH TEMPERATURE

Pavesi A, Paparella C, Bouvet B, Pituelli N. Fac. Cs. Biog y Farm. Química Analítica Clínica.

Sexual crimes are a matter of great social concern. Acid phosphatase is the parameter that desbribes phosphatase activity at pH bellow 7. The detection of the prostatic isoenzyme is taken as a legal medical indicator of the presence of seminal plasma. The objective of this work is to compare the prostatic acid phosfatase activity in basal seminal plasma and post exposition to high temperature. The samples of fresh seminal plasma (n=25) were divided in two fractions, F1=1:10000 dilution and F2=1:200 dilution. These samples were subjected to 54°C±2 in a stove to simulate a critical environmental exposure. The activity was detected by a kinetic method using α naftilphosphate as substrate and citrate buffer (pH:5,2). The t Student test was applied to compare the obtained enzymatic activity in the F1 fraction (not exposed) versus F2 (exposed). The result shows statiscally significant differences between both fraction (x1 = 87.92 \pm 45.0 ; x2 = 27.1 \pm 1.9 ; p = 0.012). The decreased activity post exposition is not an obstacle for its detection because of its high activity in fresh samples and the sensitivity of the developed method (0,8 U/l). This determination has great relevance to analyze samples of azoospermic or oligospermic suspects. Therefore the investigation of prostatic acid phosfatase activity is a good indicator of the presence of semen in forensic samples.

30.

ANTI-OXIDIZED LOW-DENSITY LIPOPROTEIN ANTI-BODIES AND LIPID PROFILE IN NORMO- AND DYS-LIPEMIC PREGNANT WOMEN. A PRELIMINARY STUDY Valdés M¹, Pelusa H¹, Daniele S¹, Almará A^{1,3}, Caille A¹, Dimónaco R², Renzi J², Ghersevich S¹, Arriaga S¹.

¹Fac Ciencias Bioquímicas y Farmacéuticas. ²Fac. Ciencias Médicas. ³CIUNR. UNR. E-mail: mdavaldes@hotmail.com

Dyslipidemia and oxidative stress associated to pregnancy would increase oxidized low-density lipoproteins (oxLDL) thus favouring abortion or eclampsia. OxLDL are more immunogenic than native LDL, so anti-oxLDL autoantibodies (Ab) reflect *in vivo* native LDL oxidation. Previously, we determined Ab levels in normal population (NP). Aim: to establish Ab values in normo (NPW) and dyslipemic (DPW) pregnant women according to ATPIII. Sample: 19 pregnant women (10 NPW; 9 DPW) in 1st-2nd trimester of pregnancy (Obstetrician Service Hosp. Prov. Centenario).Total cholesterol (C), HDL-C, LDL-C and triglycerides (TG) were determined in plasma. Ab was measured by ELISA formerly developed by us. Results: Lipid profile (media±SD):

	GROUP	C(mg/dl)	HDLC(mg/dl)	LDLC(mg/dl)	TG(mg/dl)
	NP	171 ± 30	50 ± 12	99 ± 23	91 ± 43
	NPW	186 ± 24	58 ± 13	102 ± 18	117 ± 46
	DPW	264 ± 45	63 ± 12	161 ± 46	187 ± 75
1					

Significant (p<0.05) increases: C, LDL-C and TG for DPW vs NP and DPW vs NPW, and HDL-C for NPW vs NP and DPW vs NP. Ab levels (UAb, median/range): NP=0/0-26; NPW=2.5/0-29; DPW=11/3-26, significantly (p<0.05) higher for DPW vs NP and increasing tendency in DPW vs NPW. Conclusion: increased C-HDL in pregnancy would provide antioxidant effect enough to prevent oxidation of native LDL in NPW but not in DPW, reflected by elevated Ab in DPW. This determination would be useful for the evaluation of a pregnancy at risk.

31. BACTERIOPHAGES APPLICATION TO DISSEMINATION CONTROL OF ESCHERICHIA COLI HARBORING STX1 AND STX2 GENES

Floreani F, Balagué C, Larini S, Fernández L. FCByF, UNR.

The importance of Shiga toxin-producing Escherichia coli (STEC) non-O157 strains, as food contaminants or zoonotic pathogens, has been widely documented. Considering that enterobacteriaceae interchange easily antibiotic treatment resistence genes, it is interesting to use other alternative controls against STEC strains dissemination. Owing to this we have decided to determine conditions of isolation and amplification of phages that allow the biocontrol of STEC strains. We have proceeded to study, isolate, titrate and determinate the optimum multiplicity of infection (MOI) of phages proceeding from 31 pediatrics patients feces. We have determinate de phage specificity in the following species: Escherichia coli (19 iso-lates and 6 ATCC strains), Shigella flexneri (2 isolates), Proteus mirabilis (6 isolates), Citrobacter freundii (3 isolates), Citrobacter diversus (1 isolates) and Salmonella typhimurium (1 isolate). In the isolated phages the genes stx1, stx2 and eaeA have been amplified by polymerase chain reaction (PCR). We have isolated 4 phages whit the following titles: gf1 5.8×10^9 PFU/mL; gf2 1×10^8 PFU/ mL; gf3 5 x 10⁸ PFU/mL and gf4 1 x 10⁷ PFU/mL. The gf1 phage has shown specificity to E. coli (3 isolates and 1 ATCC strain) and S. flexneri (1 isolate); the gf2 phage has shown specificity to E. coli (3 isolates); the gf3 phage has shown specificity to E. coli (3 isolates and 1 ATCC) and *S. flexneri* (2 isolates); and the gf4 phage has shown specificity to *E. coli* (3 isolates and 1 ATCC) and *S. flexneri* (1 isolate). The phage optimum MOI has been the same for all of them but it depended on the studied strains, for *E. coli* DH5 α strain has been 10^1 , for the *E. coli* 4823 strain has been 10^2 and for the *E. coli* 20 strain has been 10³. None of the 4 phages has shown pathogenicity genes. The obtain results let us see the importance of the wild phages as bacteriolitics elements against STEC non-O157 strains and infer their utility as alternative therapy.

32.

ISOLATION OF *MALASSEZIA GLOBOSA* FROM PATIENTS WITH PITYRIASIS VERSICOLOR

Ramadán S¹, Ramos L¹, Sortino M¹, Bulacio L¹, López C¹, Cabrni JM²

¹CEREMIC, FCByF, UNR. ²Ctro. Espec. de la piel-Rosario.

The eleven species of *Malassezia*, emergent patogen had adquired importance for their association to Pityriasis versicolor (PV), folliculitis, atopic dermatitis and seborrheic dermatitis. The epidemiology of *Malassezia* genus has not been yet completely dilucidated and there are variation in the isolated specie according with the geographic zone or the population studied.

The aim of this work is communicate the first case of PV caused by *M.globosa* in our Center. A male patient, 27 years old, with skin lesions was treated empirically with oral antifungals. He had a relapse when the treatment had just finished. He presented hipochromic lesions in neck and torax, presuntives of PV, when the sample was taken.

Short mycelium was observed in direct examination from skin scraping and yeast colonies were obtained in cultures. Phenotipic assays were performed in order to identified this yeast: catalase activity, β -glucosidase, Tween assimilation and termotolerance at 37° and 42°C, which ones confirmed the isolation of *M.globosa*. The patient is under antifungal treatment.

The finding of this species in our zone is highly showy, since in previous studies of our work group it had not been detected. Although *M.globosa* presents an important prevalence in Japon and Europe; in regional studies in our country *M.sympodialis*, *M.furfur* and *M.sloffiae* appears to be the main causal agents of PV. It is important to carry out the species identification in our region in order to deepen the knowledge of ecology, epidemiology and the role of the different species of *Malassezia* genus like pathogenic agents.

PRELIMINARY STUDY OF PHAGOCYTOSIS OF YEASTS OF GENUS *Candida*

Echenique C^{1} , Tosello ME^{2} , Biasoli M^{2} , Luque A^{2} , Racca L^{3} , Magaró H^{1}

¹Área Parasitología.²CEREMIC.³Área Estadística. Fac. Cs. Bioq. y Farm. UNR. E-mail: cgechenique@yahoo.com.ar

The objective of this work was to evaluate the phagocytosis of Candida yeasts, by the cellular line RAW 264.7 macrophages (RM) and peritoneal macrophages (PM) of CBi mice previously inoculated with the yeasts. Two strains of yeasts were used: Candida albicans 787 (Ca787) and Candida dubliniensis 79MF (Cd79MF). Six mice of line CBi were inoculated by oral route with each yeast and 3 weeks after inoculation they were sacrificed and PM were extracted. PM and the RM were cultivated in MEM with bovine fetal serum and antibiotics, at 37°C with 5% CO₂ during 24 hs. Phagocytosis test in vitro: suspensions of 1 x106 of RM and PM and were incubated in MEM at 37°C with 5% CO, during 24 hs. After that, suspensions of 2.8 x 106 lev/ml of Ca787 and Cd79MF were added. Macrophages and yeasts were incubated 30 minutes at 37°C in 5% CO₂. The % of macrophages with yeasts phagocytated (MYP) was determined. When RM were used, the % of MYP was 29% for Ca787 and 12% for Cd79MF. For the PM the % of MYP was 45% for Ca787 and 26% for Cd79MF. Phagocytosis of Ca787 was bigger than phagocytosis of Cd79MF (p < 0.005). The% MYP of the PM was bigger than the % MYP of the RM (p < 0.01). These preliminary results suggest that phagocytosis was greater when macrophages were previously stimulated by yeasts inoculated in mice. A greater number of strains of C. albican and C. dubliniensis would be required to elucidate the importance of phagocitosis on pathogenicity processes of both species.

34.

RENAL ELIMINATION OF P-AMINOHIPPURATE IN GLU-TATHIONE DEPLETED RATS

Di Giusto G, Torres AM.

Area Farmacología, Fac. Cs. Bioq. y Farm. CONICET, UNR.

Glutathione (GSH) is essential for maintaining an adequate renal structure and function. The aim of this study was to evaluate the renal elimintation of p-aminohippurate (PAH, prototypical organic anion) and the expression of the organic anion transporter 1 (Oat1) in GSH depleted rats with diethylmaleate (T, 4 mmol/kg b.w., i.p., n = 6, 2 h before the experiments). A parallel group of control rats was processed (C, n = 7). A pharmacokinetic study of PAH was performed after a single dose (30 mg/kg p.c., i.v.). Data were adjusted to a bicompartimental model and the main pharmacokinetics parameters were calculated. Oat1 expression in renal cortex homogenates was performed by Western blotting. (*P<0.05). PAH systemic clearance (ml/min/ 100 g b.w., C: 2.95 ± 0.13 , T = $1.43 \pm$ 0.19*), elimination rate constant from the central compartment (K $_{1-0}$, min⁻¹, C: 0.494 ± 0.094, T = 0.215 ± 0.035*). As PAH metabolization and biliary excretion are negligible, the decrease in K 1-0 suggests a lower renal excretion of this anion. No significant differences were observed in the volume of the central compartment (mL/100 g b.w., C: 7.48 \pm 1.57, T: 6.97 \pm 0.27). The total volume of distribution was decrease in T rats as consequence of the decrease in the volume of peripheral compartment (mL/100 g b.w., C: 23.07 ± 1.64 , T: $11.47 \pm 2.38^*$). Oat1 abundance (%) was increased in T rats (C: 100 ± 11 , T: $267 \pm 30^*$). Nevertheless, the increase in Oat 1 expression observed in renal cortex homogenates from GSH depleted rats, they showed a decrease renal capacity for the elimination of organic anions. Impairments in the Oat1 subcellular distribution and in the renal hemodynamics might explain this tubular alteration.

35.

RENAL TUBULAR FUNCTION IMPAIRMENT AND ITS TIME EVOLUTION IN RATS WITH BILATERAL URE-TERAL OBSTRUCTION (B)

Villar S, Brandoni A, Torres AM.

Area Farm. Fac de Cs. Bioq. y Farm. UNR. CONICET.

Previous studies showed a decreased fractional reabsorption of sodium and water in rats with B. In this study, we evaluated the time evolution of water and electrolite renal handling in rats with B. Adult male Wistar rats were employed. Both ureters were ligated during 24 h, then they were released and the studies were done after 1 (B1,n=5), 2 (B2,n=6) and 7 days (B7,n=5). A parallel group of sham rats (S,n=16) was employed. Na,K-ATPase activity (umolPi/h/mg prot) was evaluated by spectrophotometric technique (NK). Fractional excretion of sodium (EF%Na), potassium (EF%K) and H₂O (EF%H₂O) were determined employing conventional clearance techniques. The abundance of the renal AQP2 (%) was determined by Western Blotting. Data were analysed with ANOVA and Newman-Keuls P<0.5: [a]vsS, [b]vsB1, [c]vsB2, [d]vsB7.

	NK	NK NK		EE0/ V	EE%/ 1120	AQP2	AQP2
	cortex	medulla	EF 70INA	EF 70K	EF 70H2U	cortex	medulla
s	15.9±0.81	17.3±0.9	1.28±0.1	30±2.5	0.87±0.12	100±4	100±6
B1	11.9±0.7 a,c,d	11.7±0.5 a	50.3±11.6 a,c,d	88±5.5 a,d	9.86±1.57 a,c,d	64±5 a	72±5 a
B2	16.4±1.1 b	11.9±0.8 a	2.96±0.2 b	78±8.2 a,d	5.04±0.93 a,b,d	52±6 a	52±6 a
B 7	15.5±1.1 b	14.4±2.6	1.46±0.1 b	39±3.1 b,c	0.89±0.10 b,c	68±6 a	62±11 a

These modifications in Na,K-ATPase activity and AQP2 expression could explain, at least in part, the impairment of urinary concentrating capacity associated in rats with B.

36. SEX AND AGE INFLUENCE IN THE RENAL EXPRESSION OF OAT1 AND OAT3 TRANSPORTERS IN RATS Brandoni A, Torres AM.

Área Farmacología. Fac. Cs. Bioq. y Farm. U.N.R. CONICET.

Organic anion renal transporters contribute to the excretion of a variety of substances from endogenous and exogenous origin. We have demonstrated sex and age-related differences in paminohippurate (PAH) pharmacokinetics. The aim of this work was to evaluate the abundance of two organic anion transporters, OAT1 and OAT3, involved in PAH renal tubular secretion in male (M) and female (F) rats of 120 days ($M_{120} n=6$; $F_{120} n=6$) and 365 days $(M_{_{365}} n=6; F_{_{365}} n=6)$ old. Renal homogenates were prepared from kidney cortex from each group. Abundances of OAT1 and OAT3 were analyzed in these samples by Western Blotting. Statistical analysis was performed using ANOVA (p<0.05: a vs. M_{120} ; b vs. $\begin{array}{l} \text{Hardyon performed using friction (p) order a torm (p) order a to$ days-old rats, a decrease in OAT1 expression was observed in F. Also a decrease in this transporter expression, in females, was evidenced in rats aged 365 days. $M_{_{365}}$ showed a lower expression of OAT1 than $M_{_{120}}$, the same was evidenced in $F_{_{365}}$ compared with F_{120} . For OAT3, a slight decrease in the abundance was observed in F_{120}^{120} compared with M_{120} , and a great decrease in F_{365} compared with M_{365} . On the other side, a slight decrease in OAT3 expression was observed in M₃₆₅ compared with M₁₂₀, and an important decrease in F_{365} compared with F_{120} . Gender and age-linked modifications in the pharmacokinetics of PAH could be related to the observed differences in the expression of OAT1 and OAT3. These sex and age-related differences could be relevant especially in drug administration, classified as organic anions, with a narrow therapeutic index.

EFFECT OF ERYTHROCYTIC ANIONIC CHARGE DE-CREASES BY *IN VITRO* GLICOSILATION

Re A, Lebensohn N, Valverde J, D'Arrigo M, Foresto P. Lab. Inmunohematología y Hemorreología. Depto. Bioquímica Clínica. Fac. Cs. Bioquímicas y Farmacéuticas. UNR.

The aim of present investigation was to study the variation of membrane erythocytic anionic charge (CAE) and erythrocyte aggregation in glycosylated samples *in vitro* and healthy subject. Twenty five samples from healthy donors were glycosylated *in vitro* through the incubation of the erythrocytes in PBS containing glucose.

CAE was determinate by binding of the cationic dye Alcian Blue, which is an expression of the anionic charge on the cell surface. The cells were subjected to separation in an aqueous two-phase system containing 5% (w/w) Dx and 4% (w/w) PEG in PBS which was defined as the Partition Coefficient (PC). Erythrocyte aggregation was studied by digital image analysis from the morphology eritrocytaire through an Aggregate Shape Parameter (ASP).

The values of CAE (%), PC and ASP for healthy subject showed: 100; 0.95 ± 0.03 ; 0.25 ± 0.07 and in glycosylated samples showed: 41±8; 0.38 ± 0.12 ; 0.69 ± 0.09 . Our results show a significative decreasing of CAE by *in vitro* glicosilation in an attempt of model the non-enzymatic glicosilation process in diabetic. The reduction of CAE glycosylated *in vitro* samples contribute to the study of the erythrocyte aggregation alterations found in diabetic patients.

38.

COMPARISON OF THE EFFICIENCY OF TWO METHODS OF SEPARATION OF POPULATIONS ERITROCITARIAS OF DIFFERENT AGES

Luján M, Rucci A, Cotorruelo C, García Borrás S, Racca L, Acosta I del L, Biondi C, Racca A.

Facultad de Ciencias Bioquímicas y Farmacéuticas. Universidad Nacional de Rosario. (2000) Rosario.

The methods more commonly used to separate Senescent Red Blood Cells (SeRBC) are based on the increase of the density that they present as they age. The objective of this work was to compare the efficiency in the separation of RBC populations of different ages, being used two preparatory methods: differential centrifugation (CD) and gradients of Percoll (GP). ACD anticoagulated blood samples were processed. Young (Y) RBC and SeRBC were obtained by CD (n=15) and GP (n=12). In aliquot of both cellular suspensions were analyzed. Mean Corpuscular Volume (MCV) using a SYSMEX SF3000 counter, % of Reticulocyte using brilliant cresil blue and % of active monocitos (MA) by means of the eritrofagocitosis test. The differences of the values obtained between YRBC and SeRBC by both methods of separation were for VCM (CD: 1.72±3.54 vs GP: 2.35±0.57); R (CD: 1.69±0.37 vs GP: 1.66±0.24) and MA: (CD: 14.10±1.89 vs GP: 14.25±1.83). When applying the t of student for independent samples, this values does not differ significantly between both methods (p > 0,05). The similar results obtained using both preparatory methods, indicate that its efficiency of separation is comparable. Because GP allows to obtain a greater yield of the populations of YRBC and SeRBC, it would be recommendable in studies of the mediators involved in the aging of RBC which demand the use of great amount of sample.

39.

FETAL *RHD* GENOTYPING USING FREE DNA FROM MATERNAL PLASMA

Cotorruelo C¹, Gaspari C, Biondi C, García Borrás S, Di Mónaco R, Racca A².

Area Inmunología. Dpto. de Bioquímica Clínica. Fac. de Ciencias Bioquímicas y Farmacéuticas. ¹CONICET. ²CIUNR. UNR.

Prenatal determination of fetal RHD is required when the fetus is at risk of haemolytic disease of the newborn due to maternal alloimmunisation. Invasive methods, such as amniocentesis, to obtain fetal DNA for genetic analysis pose a risk to the fetus and mother. Free fetal DNA has been recently found in maternal plasma providing new possibilities for prenatal RHD determination by non-invasive PCR based procedures. The aim of this work was to assess the feasibility of RHD genotyping of the fetus by PCR amplification of fetal DNA obtained from maternal plasma. We analyzed plasma samples from 9 RhD negative pregnant women. Fetal DNA was extracted from maternal plasma through absorption and elution in silica gel membranes. RHD genotype was determined by a nested PCR strategy that analyses intron 4 of the RHD gene. The fetal origin of the DNA was confirmed by amplification of the SRY gene in cases of male fetuses. Six fetuses were RHD positive and 3 were RHD negative. The SRY gene was amplified in 5 plasma samples from the pregnant women. For all cases, 100% correlation was observed with RhD phenotype performed at birth. Fetal RHD status may be determined in DNA extracted from maternal plasma of pregnant women by the nested PCR strategy developed. These preliminary results suggest that this non-invasive procedure may be useful for the management of RhD negative mothers. A large scale evaluation is needed before considering application in clinical situations.

40.

GLYCOSILATION ABNORMAL OF ABH ANTIGENS IN ORAL CANCER

Campi C¹, Escovich L¹, Racca L, García Borrás S, Racca A, Cotorruelo C, Biondi C.

Fac. Cs. Bioquímicas y Farmacéuticas. ¹Fac. Odontología. UNR.

In most human carcinomas, including oral carcinoma, a significant event is decreased expression of histo-blood-group antigens A, B and H. The mechanisms of aberrant expression of blood-group antigens are not clear in all cases. The aim of this work was to investigate the association of ABO blood groups and oral cancer, we conducted a study of premalignant lesions and malignant tumors diagnosed. We studied the antigen expression in tissue sections of patients with precancerous and cancerous oral lesions (n=50) using a modified specific red cell adherence technique (SRCA-test). The SRCA test results were graded from negative adherence to very strongly positive adherence. Negative adherence was defined as a complete absence of adhered indicator erythrocytes. In 17 of the50 samples analyzed the SRCA test showed slightly positive results on atypical areas, and there was a complete antigen deletion in areas affected by neoplasia. Eight samples showed a total absence of ABH antigens in both histologically normal and pathological areas. Further it is suggested that areas of blood group isoantigen negative epithelium showing atypia, or in some instances near normal histology, may give rise to relatively low grade carcinomas. Considering these results we suggest the use of this method to monitor probable preneoplastic lesions in risk population, especially in those with no secretor status.

A13

41.

ALLELES *HLA-DRB1* IN PATIENTS WITH LEPROSY IN A ROSARIO POPULATION

García Borrás S, Cotorruelo C, Recarte M¹, Biondi C, Racca L, Bottasso O², Racca A.

Area Inmunología. Facultad de Ciencias Bioquímicas y Farmacéuticas. ²Facultad de Ciencias Médicas. UNR. ¹Servicio de Dermatología. Hospital Carrasco. (2000) Rosario.

Leprosy is the clinical syndrome that occurs in genetically predisposed individuals as a result of infection with Mycobacterium leprae. Although the immunopathology of leprosy has received much attention over the years, the proper reasons underlying individual differences in resistance and response to the bacilli are less characterized. The study included 71 unrelated individuals with diagnostic of leprosy, based on clinical assessment and detection of acid-fast bacilli in skin slit smears and skin lesion histopathology. Additionally, 81 non-related healthy individuals with neither symptoms nor previous diagnosis of leprosy disease were included as a control group. Non-infected individuals were ethnically matched patients. Genomic DNA was extracted from peripheral blood using the standard salting out method and used as a template to amplify by the PCR the polymorphic second exon of the HLA-DRB1. PCR products were hybridized separately with sequencespecifics oligonucleotides (SSOP). In comparison with the control group, leprosy patients revealed a greater occurrence of the DRB1*1401 and DRB1*1406 alleles, for which they may be implied in leprosy susceptibility. By opposite, DRB1*0808 and DRB1*1103 alleles were less frequent among patients and hence more involved in disease resistance. The HLA-DRB1 alleles could act alone or in combination with other genes to confer differential susceptibility and also protection to leprosy disease in endemic areas of the American continent.

42.

PRELIMINARY HISTOCHEMICAL STUDY OF THE HEPATOPANCREAS OF THE ESTUARINE CRAB Chasmagnathus granulatus

Longo MV, Goldemberg AL, Díaz AO.

Dpto Biología. FCEyN. UNMDP. Funes 3250, 3° Piso. (7600) Mar del Plata. E-mail: mvlongo@mdp.edu.ar

In crustaceans, the hepatopancreas is the main organ of synthesis of digestive enzymes, absorption, storage and distribution of minerals and organic substances. The aim of the present study was to analyze the histochemical distribution of glicoconjugates (GCs) in the epithelium of the hepatopancreas of C. granulatus. Adult male crabs in intermoult, collected from Mar Chiquita Lagoon (Buenos Aires, Argentina), were acclimated in aquarias. The hepatopancreas were fixed with Davidson solution and subjected to histochemical techniques: 1- PAS: GCs with oxidizable vicinal diols and/or glycogen; 2- α-amylase/PAS: as a control of glycogen; 3- Alcian Blue (AB) pH 2.8: GCs with carboxyl groups and/or with sulphate esters; 4- AB pH 1: GCs with O-sulphate esters; 5- AB pH 0.5: very sulphated GCs. Immature R cells showed high glycogen contents in their cytoplasm and vacuoles, whereas the mature stages had weak to moderate amounts of GCs with carboxyl and sulphated groups in their apical zone. The same pattern was observed in the cytoplasm of B cells. However, scarce glycogen was found in the vacuoles of immature B cells, prevailing the GCs with oxidizable vicinal diols. Mature B cells exhibited vacuoles with very sulphated GCs. The cytoplasm of E and F cells showed scarce GCs with oxidizable vicinal diols. This type of GCs was also found in the brush border of all cell types, associated with moderate contents of very sulphated GCs. The composition and distribution of GCs in the epithelium of the hepatopancreas of C. granulatus would be related to the role that each cell type carries out in the digestive cycle.

43.

MORPHOMETRIC AND STRUCTURAL DIFFERENCES OF MYENTRIC PLEXUS IN DISTAL COLON STUDIES BY NADPH TECHNIQUE IN RATS FED WITH SOY FIBER, CELLULOSE OR COMMERCIAL BALANCED FOOD

Geuna JD¹, Hisano N¹, Martínez C², Posada M³, Olguin MC⁴. ¹Cátedra de Histología y Embriología, FCM, UNR. ²Área Estadística y Procesamiento de Datos, FCB y F, UNR. ³Cátedra de Biología, FCM, UNR. ⁴Cátedra de Bromatología, FCB y F, UNR.

Auerbach's plexus are formed by nerves, neurons and glias that constitute ganglia conforming a reticular image characteristic of each gut segment. Neurons from distal colon in beta rats fed with different diets have been analyzed. Male beta rats (obese and diabetic), 21 days of age, were separated in 3 experimental groups: A) fed with sov fiber. B) fed with cellulose, and C) a control group fed with commercial balanced food (Cargill). At 80 days of age, they were sacrificed with ether overdose. Colon were weighed, measured and fixed in 4% paraformalaldehyde in PBS, incubated in nitroblue tetrazolium and NADPH: NADPH-positive neurons were counted under graticulated grid attached to ocular. Statistical studies as Kruskal-Wallis and Dunn's contrasts were done. Neurons were counted: A) 44.31 ± 17.69/mm²; B) 35.58 ± 10.93/mm²; C) 27.43 ± 12.63/mm² (n.s). Rats weight: A) 276.32 ± 40.88g; B) 284.60 ± 40.86g; C) 314.55 \pm 36.32g (n.s.). Colon weight: A) 1.29 \pm 0.12g; B) 1.27 \pm 0.22g; C) 1.66 ± 0.22 g (C >B) (P<0.05). Colon length: A) 15.73 ± 0.91 cm; B) 16.68 ± 0.46 cm; C) 17.08 ± 3.59 cm (n.s.). There were no significant differences in NADPH-positive neuron count between groups fed with different diets. Only colon weight of cellulose fed rats was higher than commercial balanced food.

44.

MULTIVARIATE ANALYSIS WITH INCOMPLETE INFOR-MATION. CHARACTERIZATION OF CHILDREN WITH LEUKEMIA

Quaglino MB, Vitelleschi MS.

IITAEE. FCEyE.UNR. E-mail:mvitelle@fcecon.unr.edu.ar

Principal Component Analysis (PCA) is a very helpful technique for analysing the information contained in the data matrix. Its application requires all measurements to be recorded for the entire set of individuals and the number of individuals to be significantly larger than the variables. Wold S. proposed an algorithm for the calculation of principal component (Non-linear Iterative Partial Least Squares, NIPALS), that can be used even when these conditions are not fulfilled. In this paper the technique of PCA by NIPALS algorithm is applied to the information from thirty-one children with Acute Lymphoblastic Leukemia (ALL) treated at the Hospital «Victor J. Vilela», which had a 20% missing information. The application of PCA on the information available allowed us to evaluate which factors established the greater differences among the studied children, as well as to analyse the possibility of grouping them in subgroups of similar characteristics. The collected variables were: age, white blood cell count, not differentiated cell counts (blasts) in peripheral blood, the amount of hemoglobin, platelet count, the increase of the normal size of the liver and the increase of the normal size of the spleen. The PCA shows two differentiating factors that account for 70% of the total variability among these children. The first factor identifies the degree of progression of the disease, which is associated in a positive way to a greater number of leukocytes, a larger number of blasts and an enlarged liver and spleen. The second factor separates children according to the amount of hemoglobin and platelets. The use of NIPALS algorithm allowed us to analyse, with a classic technique of multivariate analysis, the incomplete information corresponding to a small biological data set, without additional losses.

45

RELATIONSHIP BETWEEN OXIDATIVE STRESS AND ATP-ASA ACTIVITY IN RATS WITH PARTIAL HEPATECTOMY *González MA, Contini M del C, Millen N, Mahieu S.*

Lab. de Investigaciones Fisiológicas Experimentales. FBCB. UNL.

Oxidative stress level (balance between pro-oxidant and antioxidant) is under genetic control and increases with cell differentiation. The objective was to assess oxidative stress parameters and membrane ATPase activity of regenerating livers. Adult male Wistar rats (n=5/group), were divided into three experimental groups: A: controls; B: hepatectomized 48 hours prior to the experience, C: hepatectomized 168 hours before the experiment. It was determined catalase (CAT) activity (Beers method), glutathione peroxidase (GSH - Px) (Paglia and Valentine method), lipoperoxidation (LPO) through dosaje of thiobarbituric acid reactive substances (Ohkawa method), ATP-ase activity was expressed as the difference between the amount of inorganic phosphate in the presence of K (total AT-Pase) and in the absence of ClK and oubaina, and the content of glutathione (GSH) (Ellman method). The results were expressed as the mean±SEM* significant difference p<0.05). LPO(nmolMDA/ g liver. A: 275.51±38.5, B:573.2±75.5*, C:205.51±48.2; GSH(μmol/ g liver.) A:3.47±0.14, B:2.01±0.12*, C:3.07±0.11; GSH-Px(U/mg prot) A: 82.84±7.05, B: 45.2±3.31*, C:93.84±7.11; CAT(U/mg prot), A: 138.8±3.46, B: 11.33±0.3*, C: 110.7±2.3; ATPase(umol Pi/ h mg prot) A:5.44±0.05, B:2.11±0.02*, C:4.25±0.17. The partial hepatectomy at 48 hs. significantly decreased the activities of GSH - Px and CAT antioxidant enzymes as well as the content of GSH with a significant increase in the LPO, showing the presence of oxidative stress. Moreover, the activity of the enzyme membrane ATP-ase declined 61% at 48 hours while at 168 hs their values were normal. These findings might suggest that the harmful effects on this membrane enzyme in regenerating livers could be related to oxidative stress induced liver injury.

46.

OXIDATIVE STRESS AND ALUMINIUM: EFFECT ON HEPATIC ATP-ASE ACTIVITY IN FEMALE RATS *Contini M del C, González MA, Millen N, Sain J, Zacarías F, Mahieu S.*

LIFE. FBCB. UNL.

The aim of this work was to evaluate the relationship between oxidative stress produced by Al, the activity of the membrane Na/K ATP-ase and the protective function of estrogens. We used Wistar female rats, divided into 4 experimental groups (n=5): control (C); ovariectomized (OVX); treated for 3 months with aluminium lactate (0.62mg Al/100g pc.ip)(Al); and ovariectomized and treated with Al (OVXAl) The evaluated parameters: activity of catalase (CAT)-Beers's method, glutathione peroxidase (GSH-Px)- Paglia and Valentine's method, glutathione reductase (GR)-Horn's method, lipoperoxidation (LPO)-Okhawa's method, and content of glutathione (GSH)-Ellman's method. The activity of ATP-ase by Koshier-Summer method's. Data were analyzed by ANOVA and expressed as the mean±SEM (*p <0.05). LPO (nmolMDA / gwt) C: 202 ± 12 ; OVX: 226 ± 13 ; AL: 238 ± 15 ; OVXAL $336 \pm 18^*$. GSH (µmol / gwt) C: 3.1 ± 0.2; OVX: 3.9 ± 0.2*; AL: 3.8 ± 0.09*; OVXAL: 3.7 ± 0.1*. GSH - Px (NADPH nmol / min.mgprot) C: 54 ± 6; OVX: 44 ± 2; AL: 86 ± 8; OVXAL: 80 ± 5*. CAT (U / min.mg.prot) C: 183.3 ± 57.5; OVX: 248.8 ± 42.8; AL: 127.3 ± 6.7; OVXAL: 108.3 ± 12.1, GR (NADPH nmol / min.mgprot) C: 0.91 ± 0.21 ; OVX: 2.78 ± 0.68 ; AL: 5.19 ± 0.94 ; OVXAL: $4.78 \pm$ 1.09*; ATPase (μ mol Pi/h.mgprot) C: 4.35 ± 0.37; OVX: 3.16 ± 0.24; AL: 4.60 \pm 0.62; OVXAL: 2.16 \pm 0.06*. Lack of estrogens caused Al to produce oxidative stress, which could be noted by an increase in LPO and a decrease in antioxidant systems. This in turn caused the activity of membrane Na/K ATP-ase to decrease. These results suggest that estradiol would act by protecting the membrane from the deleterious effects of the metal.

47.

ANTI-OXIDATIVE/OXIDATIVE STATUS OF RENAL TISSUE IN FEMALE RATS WITH CHRONIC EXPOSURE TO ALUMINIUM: RELATION WITH TOTAL CELL MEM-BRANE ENZYMES

Mahieu S, Millen N, Contini M, Saín J, González M. LIFE.FBCB.UNL.

We investigated the impact of ovariectomy (OVX) on rats chronically treated with aluminium, specifically, on the antioxidative systems of renal tissue and total cell membrane (TCM) enzymes. Female Wistar rats were divided into four groups: non-ovariectomized rats were used as controls without any treatment (C, n=6), ovariectomized group (OVX, n=6), Al rats group (Al, n=6), treated during 3 months with Al lactate (0.57 mg Al/100 g bw, three times a week)and OVX + Al (n=6). We evaluated Glutathione peroxidase (GSH-Px), catalase (CAT), GSH content, glutathione reductase activity (GR.) and lipid peroxidation (LPO). In TCM we assessed the ac-tivities of Na/K ATPase and gamma-glutamyl transferase (γ GT). The data were analyzed with ANOVA. Ovariectomy induced a significant increase in GSH content as well as CAT activity, and a decreased GSH-Px activity. Al treatment led to a decrease in GSH content in OVX rats and an increase in GSH-Px activity, but there was no change in the CAT activity. GSH (μ mol/g.tej.h) C: 2,4±0,10; Al: 2.47±0,04; OVX: 3,24±0.07*; OVX±Al: 2,47±0.03. GSH-Px (nmol NADPH/min. mg prot): C: 51,2±3,9;Al: 92,8±9,0; OVX: 40,1±0.4*; OVX±Al: 75,1±5,84. CAT (U/min.g prot): C: 124,4±7,8 To: 107,7±5,8; OVX 151,1±6,3*; Ovx ± Al 153,3±6,1*. A significant decrease in yGT activity in both groups treated with Al was observed. The data indicate that in intact females, estrogens could be playing a protective role when the prooxidating agent is present. Ovariectomy leading to estrogen insufficiency is also reflected in some variation of anti-oxidative/oxidative parameters. However, no differences were noted for either the LPO levels or the Na/K ATPasa activity that would suggest the presence of very effective anti-oxidative systems at renal level. The effect of the Al on yGT activity could be independent of its pro-oxidant action.

48. THE USE OF INTRINSIC ESTIMATOR IN THE ADJUST-MENT OF AGE-PERIOD-COHORT MODELS FOR THE STUDY OF MORTALITY RATES RELATED TO MALIG-NANT BREAST TUMOUR

Arnesi N, Hachuel L. IITAE. Fac. Cs. Econ. y Estad. U.N.R.

E-mail: narnesi@fcecon.unr.edu.ar

The so-called age-period-cohort models (APC) have been proposed in order to analyze and summarize trends in certain diseases incidence and mortality rates. Such models use the information provided by a table showing disease or mortalility rate classified by age and period groups, called cohorts table. They pretend to resolve the effects associated to the age and birth cohort of those people who show the event and the moment or period the event was registered. Recently, a new method for the estimation of APC model parameters known as Intrinsic Estimator has been introduced in the literature of this field. This paper introduces the application of this methodology in the study of mortality rates due to malignant breast tumour in women in 1979-2003 period in Argentina. Results show that the estimations of the effects associated with age follow the change pattern already known during life, i.e. the older the person the higher the risk. The association among the rates and the cohort is characterized by a gradual and persistent decrease in mortality rates for early cohorts. This decrease may reflect the progressive development in nutrition, health care and accessibility to health care for the younger generation, to development of medical research and the increase in women"s educational level. In comparison, estimations of the effects associated to period show a relatively less net variation in mortality rates in relation to that factor. Even though the application of the proposed methodology has been satisfactory, the availability of a series of longer five-year period would enable to find results more relevant regarding cancer problem.

A15

49.

ISOLATION AND IDENTIFICATION OF MICRO ORGAN-ISMS IN LIMONCELLO FROM DIFFERENTS SOURCES *Felix MML¹, Amestoy I², Yeannes MI^{1,2}*

Preservación y Calidad de Alimentos. Dto. Qca. FCEyN, UNMDP. Funes 3350, Mar del Plata. Argentina. ¹UNMDP, ²CONICET. Email: mfelix@mdo.edu.ar

The limoncello is an Italian liquor elaborated industrial and artisanal. Alcoholic graduation is between 30 to 42°C. Since its simple manufacture, in our country the elaboration at industrial and artisanal level has proliferated. There is a great diversity of quality of them. The objective of work was determining the microbiological quality of limoncellos from different sources. Seven samples were analyzed, 1 industrial (A), 3 artisanal (B; C; D) and 3 domestic (R; F; G). The analysis was performed with aliquots from each sample. 0,1 ml from dilutions was inoculated on surface on HyL agar and on count agar by triplicate. For moulds it was incubated at 25°C for 10 days and for aerobics at 37°C for 48 h. Identification was done by macro and microscopic examination. The results were: samples A and D without growth. Counts in ufc/ml in samples were: 300 yeasts, 500 moulds and aerobics 350 in B; 300 moulds and yeasts and 200 aerobics in C; 550 aerobic and 400 moulds and yeasts in E; 100 moulds in F; 45 aerobic, 350 moulds and 700 yeasts in G. Molds identified were: Alternaria spp in B, C & D; Cladosporium spp in C, E & F; Moniliella spp and Crysonillia spp in E. They may be present in soil, seed and fruits as contamination from raw material & process and their growth could diminish the shelf life of products. There is difference between of quality in industrial and artisanal limoncello.

50.

BACTERIAL FLORA PREVALENCE ON REFRACTORY PERIAPICAL LESIONS OF ENDODONTIC ORIGIN

Spoleti P, Siragusa M, Spoleti MJ.

Endodontics Department. Faculty of Dentistry UNR. Santa Fe 3160; Bacteriology Service. Zona Norte Children's Hospital. E-mail: pspoleti@unr.edu.ar

When the dental pulp is exposed by caries, bruxism, iatrogenic preparations, and/or fractures or fissures, bacteria from the mouth can overrun and cause pulp inflammation. Toxic and metabolic products of these bacteria can be responsible for inflammatory reactions and pulp disintegration. If root canal infection persists, a chronic inflammation of the periapical area appears with loss of bone. This infection is usually poly-microbial, and contains anaerobic and facultative bacteria. In most cases endodontic infection have a good evolution after conventional endodontic therapy (among 89 and 90%). Even so, this signifies that is there a 10 or 20 % of periapical lesions which does not respond to local dental treatment, they receive the name of refractory periapical lesions. The objective of this study was to isolate and identify the bacterial flora involved on refractory periapical lesions of endodontic origin. Material obtained from ten root canals of single rooted teeth with refractory periapical lesion diagnosis was cultivated, aerobic and anaerobic flora was identified, and the data recollected was statistically analyzed.

Among the aerobic microorganisms the prevalent ones were *Strep-tococcus viridans Staphylococcus aureus*, and *Escherichia coli*. The anaerobic prevalent ones were Prevotella, Clostridium and Veilonella. Microorganisms found coincide with those described on the literature. The microorganisms recollection technique proved to be successful, although some details should be kept in mind, a small exposition to the air during the sample recollection can be enough to affect some anaerobic bacteria.

51.

IDIOPATHIC HYPERCEMENTOSIS DETECTED IN DENTAL ROOT

Obelli JJ, García Montero M, Espejo T, Gonzalez A, Kohli A. Histol., Anat. Pathology. School of Dentistry. UNR. E-mail: aliciakohli@hotmail.com

The excess of cementum in middle third of root found after a dental extraction with coronal destruction, rises the question of whether it is hypercementosis or a variety of cementoma, with scant vascularization of the bone tissue surrounding the affected piece, negatively influencing infection resistance and causing osteitis that may spread to neighboring dental pieces. Our aim was to identify this type of lesion in order to include it in future clinical findings. The seventy two-year-old patient was not a carrier of any general pathology and didn't present acute symptomatology. Once the root of the upper left second premolar was removed, well-defined round protrusions of various sizes appeared, hard to the touch, in the middle third of mesial, vestibular and palatine surfaces, of different tonalities and textures. The piece was photographed and radiographed to analyze tissue characteristics. Based on the clinical history, we discarded local as well as general causes. The root was transversely cut at the level of the most prominent lesion into two segments: one sample was submitted to abrasion and the other decalcified and stained with hematoxylin-eosin to observe the inorganic and organic phases. The abrasion a 10X showed the lesion was formed by an accumulation of secondary cementum. The 45X analysis revealed two adjacent areas in the main deposit, separated by a resting area. The decalcified and stained tissue showed a small amount of secondary cementum outside the main deposit. Our results indicate it is secondary cementum hyperplasia, known as idiopathic hypercementosis, considering the irregular tissue and its rare localization in the middle third of the root.

52.

EARLY WEANING OF HOLSTEIN CALF

Maiztegui J, Rodríguez Armesto R, Romano G, Sarchioni C. Facultad de Ciencias Veterinarias, Universidad Nacional del Littoral. Argentina. E-mail: jmaizte@fcv.unl.edu.ar

In Argentina the weaning of calf of 30 or minor days age is increassing. The aim was to evaluate the calf development early weaned replacing the milk with high quality extruded feed. A total of 20 calves from 3 to 5 day of age were used in the trial. From day 3 to 56, they were fed with 220 g of milk replacer diluted in 4 litres that contained 21% protein and 12% fat. The milk replacer was given during 4 weeks in two equal feedings at 0600 and 1800 with buckets and comercial starter (CP=18% and EM=2,9 Mcal/kg of DM). Water was supply 2 hours milk replacer. Afterwards, 2 grups of 10 calf of each were assigned at 2 treatments: (1) T BC: milk replacer 4L/d plus comercial starter and water; (2) T EX: extruded concentrate (pelleted, CP=24% and EM=3,6 Mcal of DM) and water until the calves reach 8 weeks old. The calves were weighted and the wither height, heart girth size were measured at the beginning and every 7 days until the end of work. Dry matter (DM) intake of concentrate and water was measured during 2 consecutive days. The daily gain of body weight (kg) was 0.58 ± 0.12 and 0.61 ± 0.15 , total DM intake (kg/a/d) 1,36±0,32 and 1,40±0,35 (p<0.05), water (L/d) 7,2±0,39 and 4,6±0,20 for T EX and T BC, respectively. The increase of wither height (cm/d) was 0.25 ± 01 and 0.22 ± 0.12 the total heart girth increase (cm) was 13,1±4,6 and 12,5±4,1 for T_EX and T_BC, respectively. Thus, it is possible to wean the calf with 28 days old replacing the milk with high quality feed. The calf development was similar compared with those ones weaned before the 30 days old.

53. DEVELOPMENT OF HOLSTEIN CALF AND CROSS-BREEDING HOLSTEIN X JERSEY DURING REARING Maiztegui J, Rodríguez A, Romano G, Zimermann R, Rosa R. University of Littoral. Argentina. E-mail: jmaizte@fcv.unl.edu.ar

The crossbreeding of Holstein x Jersey (HxJ) is frecuent in Argentina. The HxJ calves have less body wheigt than Holstein and to date, only few data of their growth is available. The objective of this study was to evaluate the rear of Holstein (HC) compared to (HxJ) weaned calves from 1 week up to 8 week old. A total of 28 bull calves (14 HC and 14 HxJ) from 3 to 5 days old were used. From day 3 to 56, they were fed with 4L of milk replacer per day (21% protein and 12% fat), in two equal feedings at 0600 and 1800 with buckets. Water was suply 2 hours after milk replacer. The calves were measured at the beginning and every 14 days for weigh, wither height and heart girth. Samples about 2 cm x 2 cm were taken from the most ventral portion of the floor of the cranial sac. Papillary height and thickness of the keratine were measured. The initial body weight (BW) was used as covariable for the ANOVA analysis of BW total gain. The other variables were analized with student Test. The initial BW was 43 ± 2.3 and 34.9 ± 5.6 ; the daily BW gain (kg) was 0.610 ± 0.09 and 0.595 ± 0.17 , the daily BW gain as % of initial BW was 74.4 ± 0.11 and 97.5 ± 0.21 , the dry matter intake (kg/a/d) was 1.33±0.13 and 1.26±0.12, for HC and HxJ, respectively. The wither height (cm) increased significantly (p<0.05) for HxJ (9.50 ± 2.2) compared to HC (8.78 ± 1.7) . Regarding the heart girth (cm) was 21.7±4.2 y 21.3±2.7 for HC and HJ, respectively. On the contrary, the ruminal papillae lenght and keratine thickness were both significantly different (p<0.05) for HC and HxJ: 0,911±0.11 and 0.709±0,13 and 0,062±0,01 and 0.079±0.009, respectively. In conclusion, the HxJ calves have greater increase of BW measured as percent of initial BW(p<0.05). Moreover, the ruminal developments for the breeds analyzed here were also different (p<0.05).

54.

EXPRESSION OF THE IGF-1, IN GOATS MAMMARY AND RELATIONSHIP WITH PLASMATIC COMPONENTS DUR-ING LACTATION

Romano G, Dallard B, Maiztegui J, Moreyra E, Miller RJ.

Cátedra de Fisiología e Histología y Embriología, Facultad de Ciencias Veterinarias, Universidad Nacional del Litoral. Esperanza, Santa Fe.

The IGF-1 it is correlated with plasmatic components in a different way according to the species and the tissue. The aim of this study was to determine the relationship among the plasmatic components with the expression of IGF-1 in goats mammary under conditions of natural grass (cynodon dactillun and c. irsutus). Mammary tissue was obtained from 10 goats in distinct periods of lactation. Parenchymal tissue was aseptically excised using the "quoin" incising technique. The biopsies were processed according to the technique used to reveal the presence of IGF-1 in the mammary tissue (IHQ) method (ABC). Simultaneously blood was extracted in the following periods: 5; 10-15; 20-25; 70-80 and 120 days post childbirth. The correlation coefficient was obtained by the method of Pearson (n=50, 10 animals during 5 periods) and the determination of the coefficient R2. The correlation between percentage of immunostaining area for IGF-1 and the plasmatic components cholesterol, glucemia, uremia, total proteins and albumin was -0.961 (R2=0,92), -0.968(R2=0,94), 0.373 (R2=0,14), -0.536 (R2=0,29), 0.178 (R2=0,03), respectively. Opposite to other tissue behavior, the glucemia presents negative correlation, effect that could be linked with the action of IGF-1 in animals with negative energy balance. PT and albumins presented inverse relationship with uremia during the nursing, that could indicate alimentary influence, while the evolution of the expression of IGF-1 was not affected and it maintained a descending tendency.

55

DETERMINATION OF PLASMATIC COMPONENTS IN SHEEP AND THEIR RELATIONSHIP WITH THE EXPRES-SION OF THE IGF-1 IN MAMMARY GLAND DURING LACTATION

Romano G, Dallard B, Maiztegui J, Moreyra E, Miller RJ. Cat de Fisiología e Histología y Embriología, Fac de Cs Vet, Universidad Nacional del Litoral. Santa Fe.

The IGF-I belongs to the super family of the insulin and it completes a series of functions among those that are considered of bigger importance, the regulation of the proliferation and cellular differentiation, synthesis and degradation of proteins, and transport of substances. The IGF-1, it is correlated with plasmatic components in a different way according to the species and the studied tissue. The aim of this study was to determine the relationship among the percentage of immunostaining area for IGF-1 in mammary gland with the concentration of plasmatic variables. Mammary tissue was obtained from 10 sheep in distinct periods of lactation. Parenchymal tissue was aseptically excised using the "quoin" incising technique. The biopsies were processed according to technique used to reveal the presence of IGF-1 in the mammary tissue (IHQ) method (ABC). Simultaneously blood was extracted in the following periods: 5; 10-15; 20-25; 70-80 and 120 days post childbirth. The correlation coefficient was obtained by the method of Pearson (n=50, 10 animals during 5 periods) and coefficient R2. The correlation between percentage of immunostaining area for IGF-1 and the levels of cholesterol, glucemia, uremia, total proteins and albumin was of 0,424 (R2=0,18); -0.972 (R2=0,95); -0.683 (R2=0,47); 0,259 (R2=0,07); 0,458 (R2=0,21); respectively. The biggest correlation in the glucemia could be appropriate with the differentiated action mechanism of the IGF-1 in animals with negative energy balance. Also, the IGF-1 during the nursing, diminishes its expression in mammary tissue because its action goes to other tissue, what would be linked with the control of the glucemia and not to its direct activity in the mammary gland to stimulate its production, like one observes in other species.

56.

EFFECT OF ASCORBIC ACID (VIT C) IN THE CO-TREAT-MENT WITH INTERFERON-α2B (IFN) ON RATS WITH PRENEOPLASTIC LIVER (PL)

Pisani GB¹, Quiroga AD^{1,2}, Lugano MC¹, Alvarez ML^{1,2}, Carnovale CE², Carrillo MC^{1,2}.

¹Area Morfología. Fac. Cs. Bioquímicas y Farmacéuticas. UNR. ²IFISE .CONICET.

IFN reduces the number and volume of altered hepatic foci (AHF) on rats with PL, inducing apoptosis through the production of TGF $\beta_{..}$ It has been proved in others models that TGF $\beta_{.}$ produces reactive oxygen species (ROS)-mediated apoptosis. We studied the IFN+VIT C (scavenger of ROS) co-treatment on rats with PL. We used 4 groups of adult male Wistar rats (n=10 each one). G1: subjected to the preneoplastic liver model initiation-promotion (I-P); G2: I-P and IFN treated; G3: I-P and VIT C treated and G4: I-P and IFN+VIT C co-treated. We determined by immunohistochemical staining the AHF (positives to rat glutathione S-transferase placental form); Apoptosis Index (AI by TUNEL and H&E) and Proliferation Index (PI by PCNA). IFN (G2) reduced AHF number (p<0.05). VIT C (G4) abolished this effect. Liver volume occupied by AHF diminished in G2 vs G1 (p<0.05). IFN+VIT C (G4) increased this volume fraction vs G2 (p<0.05). AI in AHF of G2 reached the highest values (p<0.05). There were no differences between groups in PI in AHF. There was an accumulation of G, phase of the cell cycle in AHF of G2 (p<0.05). We demonstrated the role of ROS in the increased apoptosis induced by IFN in AHF. The inhibition of ROS production with VIT C may be not a beneficial effect because it caused an increase in the liver volume occupied by AHF probably by a diminution of the apoptosis rate.

57. STEREOELECTRONIC FACTORS THAT CONTROL THE ACTIVITY OF BIOMIMETIC ANTIOXIDANTS

Daier V, Palopoli C, Biava H, Moreno D, De Gaudio M, Signorella S.

División Química Inorgánica, IQUIR-Instituto de Química Rosario, CONICET - UNR. Suipacha 531 (S2002LRK) Rosario.

Catalases (CAT) and superoxide dismutases (SOD) constitute the main line of defense of living organisms for controlling O₂ - y O₂. However, exogenously administered SOD/CAT has been tested with limited success, especially because of immunogenic responses. This led to an interest in developing synthetic SOD/ CAT mimics with more favorable pharmaceutical properties. We examined the CAT/ SOD activities of MnL¹⁻² and [Mn_L³⁻⁵(µ-OAc)(µ-OMe)] complexes, using 1,3-bis(5-SO₂-salicyliden amino)propan-2-ol (H₂L¹), 1,4-bis(salicylidenamino)butan-2-ol (H,L²), 1,3-bis[(2-hydroxy-5-X-benzyl-2-methylpyridyl)amino] propan-2-ol (H₂L³), 1,5-bis(5-SO₃-salicylidenamino)pentan-3-ol (H₂L⁴) and 1,5-bis[(2-hydroxy-5-X-benzyl-2-methylpyridyl) amino]pentan-3-ol (H,L5), where X = phenyl substituent. The catalase activity was determined by measuring the oxygen concentration of H₂O₂+ catalyst mixtures with a Clark-type oxygen electrode. The SOD activity was assayed by measuring the inhibition of the photoreduction of nitro blue tetrazolium in the presence of the catalyst. These studies reveal that (i) for MnL1-2 complexes, the ability of the ligand to act as an intramolecular base to assist electron transfer is a key factor in the SOD activity; (ii) in $[MnL^{1-2}]_2$ and $[Mn_2L^{3,5}(\mu\text{-OAc})(\mu\text{-OMe})]$, where the metal coordination sphere is saturated by non-labile ligands, peroxide binds Mn as a terminal ligand; (iii) [Mn₂L⁴(µ-OAc)(µ-OMe)] with two substitution-labile sites, employs Mn^{III}-µ-O₂-Mn^{III} and [Mn^{IV}=O], species for CAT activity.

58.

OPTIMIZATION OF THE ADSORPTION OF CHROMIUM (III) ON ORANGE PEEL

Pascucci F, Bellú S, García S, González J, Signorella S, Sala L. Área Inorgánica. IQUIR- CONICET. Facultad. de Ciencias. Bioquímicas. y Farmacéuticas. UNR. Suipacha 531 (S2002LRK). Rosario.

Chromium is a highly toxic contaminant. Cr(VI) species interfere the development and growing of plants, while Cr(III) species are less toxic but can be oxidized to Cr(VI). The objective of this work was to optimize the capacity of orange peel to absorb Cr(III). The material was obtained from commercial orange, air-dried for 7 days, and additionally dried at 40°C for 24 hours. Orange peel was grounded and sieved to retain the fraction of particles in the range of 0.3 - 1.2 mm size. A 2³ full factorial design was used to analyze the main factors that influence the adsorption process (pH, adsorbent mass, contact time). Only pH and adsorbent mass were significant, with probability lower than 0.05 (confidence level > 95%). The adsorption process was optimized applying a central composite response surface design. The optimal conditions found for the adsorption of 5.03 mg of Cr(III) were pH = 5 and adsorbent mass = 0.42 g. In these conditions, 80% adsorption was achieved in 15 min.

Acknowledgments: FONCyT- ANPCyT (PICT 10625), CONICET PIP 03016.

59.

OXIDATION OF D-GLUCOSAMINE WITH Cr^{vi}. REAC-TION CONDITIONS

Frascaroli MI, Andreu V, Signorella S, Sala LF.

División Química Inorgánica, IQUIR-Instituto de Química Rosario, CONICET - UNR. Suipacha 531 (S2002LRK) Rosario.

N-acetyl-D-glucosamine and D-glucosamine (Nglc) are the products of the hydrolysis of chitin, the most abundant natural aminopolysaccharide. It occurs mainly in the exoskeleton of crustaceans, insects, and in the cell walls of some microorganisms, and its annual production is almost as abundant as that of cellulose. Nglc, a natural component of glycoproteins found in connective tissues and gastrointestinal mucosal membranes, has therapeutic potential for the treatment of a variety of diseases. It is known that heavy metals can cause environmental pollution and compounds of Cr^{VI} are among the chemicals first classified as carcinogens for humans. In this work we direct our investigation to the interaction of Cr^{VI} with Nglc. Previous studies on the oxidation of D-glucose (Glc) and 2-deoxy-D-glucose (dGlc) with Cr^{VI} showed that the C1-OH hemiacetalic function reacts faster than the primary or any of the secondary alcohol groups, giving aldonic acid as the only reaction product. The study of the interaction of this aminosugar with Cr^{VI} provides information on the role of C2-NH₂ in the reaction of aminosugars with $Cr^{\mbox{\tiny VI}}.$ We analyzed the effect of temperature, ionic strength and reactant concentrations on the reaction rate. The reactions were followed under pseudo-first-order conditions, using an excess of Nglc over Cr^{VI}. The disappearance of Cr^{VI} was followed spectrophotometrically at 350 nm. When comparing the rate of the $Nglc + Cr^{VI}$ with $Glc/dGlc + Cr^{VI}$, we observe that Nglc reacts with Cr^{VI} 15 times slower than Glc and 20 times slower than dGlc, at 50°C. It is possible that the presence of the NH, group diminishes the affinity of the substrate for Cr^{VI} and, therefore, the redox reaction becames slower.

60.

THE ACTION OF VANADIUM IN COMPOUNDS WITH BIO-LOGICAL RELEVANCE

Santoro MI, Disetti ME, Cereda YA, Ceruti MJ. División Química Inorgánica, IQUIR, FCByF, UNR. Suipacha 531. (S2002LRK) Rosario.

Vanadium is found in different oxidation states, in about 65 minerals and in carbon deposits. It has been determined that vanadium(IV) and vanadium(V) interact with many molecules in the biological systems. Inside the cell, vanadium (IV) can potentially interact with carboxylate, phosphate and amine groups of the biomolecules. In the last years, they have been synthesised some organic and inorganic complexes of great value in diabetes treatments. Besides, some complexes are used in experimental models as antitumoral agents. Taking into account the growing interest in vanadium interactions with different ligands, and the multiple metal ion oxidation states, it is necessary a detailed understanding of the redox reactions of vanadium(V) with some ligands of living organisms. We study the kinetics of oxidation of 2-alpha-hydroxyacids, as L(+)-tartaric acid and D-glucono-1,5-lactone, by action of V(V). The redox reactions involves kinetic studies at low pH, intermediates and products detection and the formation equilibria of V(V)-substrate complex. The kinetics were carried out by monitoring the V(V) consumption at 350 nm and the formation of V(IV) at 760 nm. In order to provide the reaction mechanisms, the effect of substrate, vanadium and acid concentrations has been analysed, at constant ionic strength and temperature. The presence of free radicals has been detected with acrylamide in the course of the reaction. The kinetic profiles were adjusted considering a sequence of reactions of the type $V(V) \rightarrow V(V)$ -S $\rightarrow VO^{2+}$. The results will serve as model system in the search of simple vanadium complexes with biological relevance.

CHEMOPREVENTIVE ACTION OF ASCORBIC ACID ON THE MUTAGENICITY OF SULFATHIAZOLE-NITRITE REACTION MIXTURES IN ACIDIC MEDIUM: PRELIMI-NARY STUDIES

Pontoriero A^{l} , Trossero C^{l} , Gorr C^{l} , Hure E^{l} , Monti L^{l} , Mosconi N^{l} , Caffarena G^{2} , Rizzotto M^{l} .

¹Área Inorgánica, Facultad de Ciencias Bioquímicas y Farmacéuticas, Suipacha 531; ²Cátedra de Microbiología, Facultad de Ciencias Médicas, Av. Francia y Santa Fe. UNR, 2000 Rosario. Email: mrizzott@fbioyf.unr.edu.ar

Sulfonamides -or their derivatives- are widely used by their properties (antibacterial, antifungal, antidiabetic, herbicides, analytical reagents, etc.). One risk of sulfas is the possible formation of mutagenic products by reaction with nitrite in stomach. Ascorbic acid (Asc.) can react with nitrite, diminishing or removing the nitrosation risk. According to the Ames test a compound would be a mutagen if the reversion coefficient, R.C. (R.C.: revertant number per tested plate/revertant number per control plate) is ≥ 2 . Previously we proved mutagenic action of reaction mixtures of sodium sulfathiazole and NaNO, in acidic medium with the S. typhimurium TA98 strain. In the present work it was proved the chemopreventive action of one dose of Asc. on the mutagenicity of such reaction mixture. Different doses of the reaction mixture were assayed in the presence of a constant dose of Asc. with the TA98 strain. In all cases the R.C. was 1 ± 0.20 . Using the following equation: % inhibition of mutagenicity = (1 - R.C. of the reaction mixture+Asc./ R.C. of the reaction mixture without Asc.).100, it was obtained a 66% of inhibition which, as a preliminary level, suggests a strong inhibitory effect.

62.

ANTIBACTERIAL ACTION OF SULFA-Co(II) COM-PLEXES: STRUCTURE-ACTIVITY RELATIONSHIP

Monti L^1 , Hure E^1 , Mosconi N^1 , Trossero C^1 , Gorr C^1 , Pontoriero A^1 , Lima B^2 , Feresin G^2 , Tapia A^2 , Rizzotto M^1 .

¹Área Inorgánica, Facultad de Cs. Bioquímicas y Farmacéuticas, UNR, Argentina. ²IBT, UNSJ, San Juan, Argentina. E-mail: mrizzott@fbioyf.unr.edu.ar

Among the many and so different families of organic-inorganic chemicals being currently investigated due to their applications, sulfonamides are part of the outstanding ones. The minimum inhibitory concentration of sulfa-Co(II) complexes (sulfas: sulfanilamide, SA; sodium sulfathiazole, NaST; phthalylsulfathiazole, PhST) was determined with different bacterial strains: ATCC, Malbrán Institute, Pasteur Institute and Laboratory of Microbiology (LM), Cuyo Univ. Gram (-) strains: Escherichia coli ATCC 25922, LM E. coli and Pseudomonas aeruginosa ATCC 27853. Gram (+) strains: Staphylococcus aureus methicillin-sensitive ATCC 29213 and S. aureus methicillin-resistant ATCC 43300. The Co(II)-SA complex showed higher activity than the ligand against all the tested strains; similar results were showed by Co(II)-ST with P. aeruginosa, but the activity of the Co(II)-PhST complex was always lower than the ligand one. Trying to correlate these results with the structure of each complex, electronic spectra were recorded (solvents: ethanol, DMSO; range: 400-800 nm). The ligand strength in the spectrochemical serie would be in an inverse relationship in respect of the antibacterial power of the respective complex. This results, in conjunction with the analysis of the structure of the ligands and of the Co(II)-ST complex, suggest that the antibacterial power would be related to the coordination of the metal with atoms of N non heterocyclic.

63.

VASCULAR FLORA OF THE PROVINCE OF SANTA FE: Cannaceae, Marantaceae AND Zingiberaceae

Mc Cargo J¹, Di Sapio O¹, Oakley L², García R², Prado D² ¹Botánica, Fac. Cs Bioquímicas y Farmacéuticas, UNR, Suipacha 531, S2002RLK Rosario. ²Botánica, Fac. Cs Agrs-UNR, CC N°14, S2125ZAA. Zavalla.

The families Cannaceae, Marantaceae and Zingiberaceae belong into the Order Zingiberales (= Scitamineae), comprising species of tropical and subtropical marshy regions of both hemispheres. They are herbaceous, rhizomatous plants with asymmetric flowers, due to the progressive transformation of stamens in staminodes. This work contributes to the knowledge of these families providing keys based on morphological characters, and a distribution map. A thorough bibliographical review, consultation of national herbaria with important collections of the province (SF: Esperanza; SI: San Isidro; UNR: Zavalla), field work experience of the authors and lab work to confirm their identity were performed. The Family Cannaceae is represented by one genus, two species and three varieties: Canna glauca L. var. angusta J.W. Richardson; Canna glauca L. var. glauca; Canna glauca L. var. rubro-lutea Hook. and Canna indica L. They are ornamental and known as "achiras", whose distribution comprises departments of NE Santa Fe. The Family Marantaceae comprise one genus and two species: Thalia geniculata L. and Thalia multiflora Horkel; common in wetlands of the provincial center-north. As regards Family Zingiberaceae, the exotic species Hedychium coronarium J. Köning was collected in neighboring provinces (Cba, E.Ríos, Bs.As.) and even observed as sub-spontaneous in Santa Fe, but not supported by herbarium material. Keys, distribution map and illustrations are provided.

64.

IN VITRO EFFECT OF DIFFERENT CONCENTRATIONS OF PROANTHOCYANIDINE FROM *Ligaria cuneifolia* ON ERYTHROCYTE SHAPE

Crosetti D¹, Ferrero M¹, Dominighini A¹, Alvarez L³, Ronco MT³, Wagner M², Gurni A², Carnovale C³, Luquita A¹.

¹Biofísica, FCMédicas-UNR. ²Farmacobotánica, Fac Farma y Bioquímica-UBA. ³Fisiología, FCByF-UNR-CONICET, Suipacha 570. 2000 Rosario. Argentina. E-mail: luquitale@hotmail.com

Ligaria cuneifolia (Lc) is popularly known as "muérdago criollo" Infusions of this plant are used in folk medicine to increase blood fluidity lowering the plasma cholesterol levels. Aqueous extract of Lc is constituted by flavonoids (quercetine, proanthocyanidine and categuine). In vitro studies made in our laboratory showed a direct influence of aqueous extract of Lc which altered the erythrocyte shape from dyscocyte to spherostomatocyte and decreased both erythrocyte deformability and osmotic fragility. In this work we analysed the effect of the proanthocyanidine from Lc (PLc) on erythrocyte shape. Blood from adult male Wistar rats, was obtained by cardiac puncture. Blood samples were divided in: Control (C) (n=8) and Treated (T). PLc was added to T blood (mg/ml): 0.10 (T1) and 1.00 (T2) (n=8 of each one), and incubated 30 minutes at 37°C. The erythrocyte shape was evaluated by direct microscopy, the Morphological Index was estimated (IM = Σ [index of shape (discocyte:0; stomatocyte I:-1; stomatocyte II:-2; stomatocyte III:-3)x number of cells / total number of cells]): C: -0.49 ± 0.03 ; T1: - 0.62 ± 0.07 ; T2: -2.64 $\pm 0.28^*$, (*p=0.05 vs. C). Percentage of stomatocyte III: C: -2.62 ± 0.95 ; T1: $-10.75 \pm 3.01^*$; T2: $-54.22 \pm$ 2.21* (*p<0.001 vs. C). No changes were observed in Mean Corpuscular Hemoglobin (C: $33,08 \pm 1.13$) nor in Mean Corpuscular Volume (C: $72,20 \pm 5.78$). These data suggest a direct effect of proanthocyanidine on the erythrocyte membrane that induces a change in cellular morphology.

A19

65.

DEVELOPMENT OF THE EXTERNAL GENITALIA DUR-ING THE EARLY HUMAN FETAL PERIOD *Tellez TE, Carrera LI, D'Ottavio AE.*

Performing a morphological study of external genitalia during the early human fetal period, serial sections (7 µm thick) were obtained from the pelvic-crural zone of 9th and 10th weeks-old human fetuses. Specimens underwent histological and histochemical techniques and were later exposed to planimetry and spatial reconstruction. A well developed conic genital tubercle (GT), with a ventral groove extending the penial portion of the urogenital sinus, was seen. Its axial region distally showed a solid cellular blastema which was proximally bifurcated and deeply connected. Numerous vessels and nerves, joining and crowning the cellular bud, were detected in the loose mesenchyma separating that blastema from the epidermal sheath. Two sharp urethral folds (UF) continued both lips of the ventral groove in the genital tubercle whilst the genital folds (GF) represented long, thick and blunt-edged elevations. The UF and GF histologically revealed an avascular blastema (possible sign of immaturity) and profuse collagen III condensed around vessels and nerves and beneath the epithelium. With the exception of the distal epidermal coat of the GT showing a fence like basal sheath and abundant PAS positive cells, probably associated with its differentiation rate, the rest of the epidermal coating resembled those covering the entire organism. In sum, no differential details were observed in external genitalia during the studied ages.

66.

EFFECT OF CALCIUM (Ca) ON THE ACTIVITY AND THE EXPRESSION OF INTESTINAL ALKALINE PHOS-PHATASE (IAP)

Brun LR, de Candia LF, Brance ML, Di Loreto V, Rigalli A. Laboratorio Biología Ósea. Fac Medicina. UNRosario.

IAP suffers structural and functional changes at high concentrations of Ca, co-expresses with the channel CaT1 and the expression is regulated by calcitriol in the same way as other proteins involved in Ca absorption. The expression decreases from duodenum to ileum, as transcellular transport of Ca does. However, the role of IAP in Ca absorption remains unknown. In situ isolated intestinal loop experiments were carried out in 200-g IIM/Fm rats. The lumen was filled with 0, 10, 50 y 100 mM Ca (5 repetitions). After 20 minutes samples were obtained from the luminal solution and IAP activity was measured. Samples of mucosa were obtained, the activity was measured (histochemistry) with BCIP and the expression of IAP was measured (immunohistochemistry) with an anti-IAP primary antibody and a peroxidase conjugated secondary antibody. Luminal activity and brush border expression of IAP were not different between groups (ANOVA, p>0.05). In contrast, IAP activity in the brush border was higher when the mucosa was exposed to 50 y 100 mM Ca (ANOVA, p<0.05). Conclusion: High luminal Ca concentrations stimulate IAP activity at the brush border without modification in IAP expression. Experiments where the activity of IAP and the absorption of Ca be simultaneously measured, will clarify this results.

67.

VARIABLES THAT INFLUENCE BONE MASS (BM) DE-VELOPMENT IN RATS WITH SODIUM FLUORIDE (NaF) TREATMENT

Brun LR, Armendariz M, Aguirre MC, Rigalli A. Laboratorio Biología Ósea. Fac Medicina. UNRosario.

NaF produces an increase in BM after 30 days of treatment, but no further increases were found in longer treatments. The aim of this work was to identify the causes of the lack of response of bone to NaF stimulus. Female 21-day old rats drank for 100 days distilled water (controls, n=40) or 100 ppm NaF solution (treated, n=40). Every 10 days 4 rats of each group were killed. Bone mass (BM: weight skeleton*100/body weight), bone fluoride content (BFC: μ g F/g bone), water intake (WI: μ l/g body weight/day) and urinary excretion of fluoride (UFE: µg/day) were measured every 10 days. BFC was higher in treated with a maximum at 40 days (Two way ANOVA). BM increased in treated rats after 30 days but the difference between groups did not increase after 100 days of treatment. WI decreased in both groups along the experiment (1,48±0.02, r=-0.63). UFE increased after 2 days in treated rats (278±42) respect to controls (26±15, P<0,05) and the difference remained unchanged throughout the experiment. The decrease in WI and the constant UFE appear to be the causes of the decrease in bone fluoride content, which could be the cause of no further response of bone. This results suggest that in long treatments the concentration of NaF in drinking water should be adjusted to produce a constant fluoride intake.

68.

EFFECT OF NIFEDIPINE ON ERYTHROCYTIC DEFORMABILITY IN PATIENTS WITH SYSTEMIC SCLERODERMA

Van Isseldyk F¹, Leroux B², Svetaz MJ³, Parente F¹, Spengler MI¹ ¹Cátedra de Física Biológica, ²Cátedra de Dermatología, Facultad de Ciencias Médicas; ³Sección Inmunidad Celular, Departamento de Bioquímica Clínica, Facultad de Ciencias Bioquímicas y Farmacéuticas, Universidad Nacional de Rosario. E-mail: isabelspengler@hotmail.com

Raynaud's phenomenon and obliteration of micro capillaries found in systemic scleroderma produce ischemia and peripheral tissue damage. It's precisely in the capillaries where the deformability of the erythrocyte (damaged in scleroderma), becomes a crucial factor in the maintenance of the blood flow and oxygen supply to the tissue. The objective of this study is to asses the effect of the nifedipine treatment (vasodilator and calcium channel blocker) over the erythrocytic deformability, in patients with systemic scleroderma. Rigidity index (inverse of the deformability) was determined by filtration through 5µm-diameter polycarbonate membranes. 47 female patients were assessed: 22 treated with colchicine (1mg\day) and nifedipine (30 mg\day), and 25 patients treated only with colchicine (1mg\day). The statistic analysis of the data using Student's t, showed that RI mean value were significantly lower (p<0,001) in the nifedipine group $(13,4\pm2,00)$ than the colchicine-only treatment group (17,40±2,42).

It's highly probable that nifedipine can inhibit the entrance of Ca⁺⁺ and prevent it's intracellular accumulation, improving patient's damaged erythrocytic deformability.

MORE FOOD OR BETTER QUALITY?

Marchegiani M, Cesolari JAM, Calvi BJ.

Facultad de Cs. Médicas (UNR), Inst. Gral. San Martín, IUNIR. E-mail: ceso@cimero.org.ar

Food quantity and diet quality are concepts whose analysis could better explain how they influence the principal symptoms of malnutrition in infants. Objective: To analyze the nutritional quality of feeding in children and its relation with energetic intake. Material and Methods: Data of 5 studies (n=100) aged 12 to 48 months of different socioeconomic levels, in a pediatric Service in San Nicolas (Prov. Of B.A.) were analyzed, utilizing a 24 h recall questionnaire. Results: 78% of the participants have an energy intake higher than required, although 20% of them had a lower intake of calcium, zinc, vitamins C and A according to the food daily recommendations (FDR). In case of iron 50%. In low socioeconomic levels the proportion of intake, lower than recommended is greater for Vitamins A, C and calcium, and lesser in iron and zinc. Half of children of low income homes have energy intake above 120% of recommended. The proportion of children with low density diets (nutritional density lower than recommended) is higher than those with less intake that FDR recommendations. Two thirds of children with iron and zinc low density diets have 20% or more than recommended diet energy intake. Conclusion: If the excess diet energy is more frequent that deficit, then it must be emphasized the concept that educational strategies and nutritional program designs priority should be quality and diversity of nourishment against food global quality.

70.

YOGHOURT, MILK AND ENRICHED JUICES (FOSHU) IN UNIVERSITY STUDENTS

Calvi BJ, Cesolari JAM, Busmail LI, Martínez C, Taborda M. Facultad de Cs. Médicas (UNR), IUNIR, UNSAM. E-mail: ceso@cimero.org.ar

FOSHU (food for specific health use) are foods added to a diet to increase their quality, providing substances that improve well-being, they are also called functional foods. Objectives: To evaluate consumption and frequency consumption (FC) of some enriched foods, such as yoghourt, milk and, natural and artificial fruit juice, in a group of University students. Methodology: 231 students (150 females and 81 males) were submitted to a questionnaire regarding voghurt, milk and fruit juice consumption and their frequency, and determining the presence or absence of special added nutrients. Results: 18.5% out of the total admitted consumption of yoghourt enriched with Ca, A and D vitamin, soy and fiber, as well as micronutrients and probiotics. FC: daily 82% and occasional 18%; 20% drink fermented milk with omega3 and oleic acid: 95% daily consumption and 5% occasionally; 16% consumes natural and artificial fruit juice or soy juice, added with vitamins, micronutrients and fibers. FC: daily 67% and occasionally 33%.

Conclusion: The tendency to functional food consumption is higher at the University, due to the fact that they provide substances improving life quality, and since they are diet or light products are more appalling for young people.

71.

EVALUATING SUNFLOWER SEED SUPPLEMENTED DIET ON MALE IIMb RATS

Passerini F, Sgro MV, Spies J, Aimaretti S, Erbayo E, Alet NA, Gayol M del C.

Cátedra de Biología. Facultad Ciencias Médicas. UNR.

The originally eumetabolic inbred IIMb rat line developed a glucose intolerance. Due to deficiencies in the commercial food composition used to feed rats, it had to be supplemented with sunflower seeds to recover body size. A group of male IIMb rats was fed with rat food (C)(n=5) and another with rat food plus sunflower seeds since weaning (C+G)(n=6). Glycemic profile, biomass and adipose panicle weight were compared between groups at 50, 100 and 200 days old. The results are shown as media±SEM: Basal glycemia (g/l): 50ds (p>0.05); 100ds (p<0.05) (C) 1,12±0,04; (C+G) 1,39±0,09; 200ds (p<0.01) (C) 1,25±0,03; (C+G) 1,55±0,08. Glycemia at 120' after a glucose overload (g/l): 50ds (p>0.05); 100ds (p>0,05) (C); 200ds (p<0.05) (C) 1,67±0,12; (C+G) 2,02±0,02. Triglyceridemia (g/l): 50ds (p>0.05); 100ds (p>0.05); 200ds (p>0.05). Cholesterolemia (g/l): 50ds (p>0.05); 100ds (p>0.05); 200ds (p>0.05). Weight (g): 50ds (p>0.05); 100ds (p>0,05); 200ds (p<0.05) (C) 471,2±14,21; (C+G) 530,0±12,75. Adipose panicles weight (g/100g weight): 200ds (p<0,01) (C) 6,10±0,35; (C+G) 7,88 \pm 0,10. Rats fed with the supplemented diet (C+G) developed glucose intolerance since 50 days old and become diabetic by 200 days old. In all cases cholesterolemia and triglyceridemia remained normal. There was an increase in rat weight in diet (C+G) compared with diet (C), and that was corresponded with greater adipose panicle weight in (C+G). Further studies on the fatty acid sunflower seed ω -6/ ω -3 ratio are needed to obtain an adequate body size without neglecting IIMb rat glycemic profile.

72.

INFLUENCE OF SUNFLOWER SEED SUPPLEMENTED DIET ON OBESE DIABETIC MALE $\boldsymbol{\beta}$ RATS

Sgro MV, Passerini F, Spies J, Aimaretti S, Gutierrez S, Alet NA, Gayol M del C.

Cátedra de Biología. Facultad Ciencias Médicas. UNR.

Deficiencies in the usual rat food have compelled to evaluate a sunflower seed supplemented diet to preserve the animal model. Male β rats were fed on either rat food (C)(n=5) or rat food and supplementary sunflower seeds (C+G)(n=5) since weaning. Variables were compared between groups at 50, 100 and 200 days old. Results are shown as media±SEM: Basal glycemia (g/l): 50ds (p>0.05); 100ds (p>0.05); 200ds (p<0.01) (C) 1,82±0,11; (C+G) 2,83±0,19. Glycemia at 120' after a glucose overload (g/l): 50ds (p>0.05); 100ds (p<0.01) (C) 1,86±0,17; (C+G) 3,50±0,30; 200ds (p>0.05) (C) 2,64±0,32; (C+G) 3,09±0,12. Triglyceridemia (g/l): 50ds (p>0.05); 100ds (p<0.05) (C) 2,28±0,43; (C+G) 0,99±0,18; 200ds (p<0.001) (C) 3,84±0,28; (C+G) 0,97±0,20. Within (C) [50ds vs 200ds and 100ds vs 200ds] (p<0.05) and (C+G) among ages: (p>0.05). Weight (g): (p>0.05) [50ds (C) 203,6±9,71 vs (C+G) 187,8±13,48; 100ds (C) 423,5±19,91 vs (C+G) 429,2±23,39; 200ds (C) 511,5±21,22 vs (C+G) 533,8±22,20]. Adipose panicle weight (g/100g weight): (p<0.01) 200ds (C) 6.31±0.20; (C+G) 8.39±0.05. The glucose impairment developed earlier in (C+G). Both groups become diabetic by 200 days old. Triglyceridemia was higher in (C) than (C+G). In all cases the cholesterolemia remained normal. There was no significant weight difference between diets although (C+G) presented heavier adipose panicles. Maintaining the hypertriglyceridemia that typies the β model, further study in the fatty acid ω -6/ ω -3 sunflower seed ratio is required.

A21

73.

PRODUCTIVITY OF DISMETABOLIC BETA LINE RATS EVALUATED BY EXPLORATORY ANALYSIS OF BREED-ING DATA BASIS

Marani J, López JP, Gayol M del C, Tarrés MC, Montenegro SM. Cátedra de Biología. Facultad de Ciencias Médicas. CIC-UNR.

The animal models have contributed to obesity and diabetes studies. Beta rat shows moderate obesity and glucose intolerance, syndrome that affects fertility. A variable exploratory analysis indicating productivity was done. It was used beta rat breeding data basis generated from mating registers of 779 females. 44% females were infertile (I, n=340) and when data were compared (media ±SD) with the fertile females (F, n=439) it was seen that in these the join for mating age lessened. Frequency distribution showed asymmetry to the right in both groups - greater occurance of the minor values – more marked than in I ($a_r = 1.08$, $a_r = 0.78$). Weight at join for mating was also lighter in F with symmetric distribution. 50% females weighed less than 231g in group F and 260g in I. Male age and weight were also less in group F. Group I distribution was asymmetric to the left (a_i = -0.135) indicating greater frequencies in the heavier weights. In group F litter size was $n=8\pm3$ breedings, with a 14 maximum, 50% mothers gave birth to more than 8 offspring and distribution was slightly asymmetric to the left (a = -0.157). Time passed from joining for mating up to birth oscillated between 30 and 92 days, 50% mothers gave birth in less than 27 days and distribution was asymmetrical marked to the right (a=2.74). Breeding outliving at waning (s/n) was 0.9±0.2 and distribution became highly asymmetric to the left (a=-1.86). Beta productivity contrasts other murine models internationally known to be sterile. Their high infertility, associated with both parent age, would be overcome by joining for mate young animals replaced by both female and male.

74.

IMMUNOMODULATORY EFFECT OF THREE DOSES OF *M.VACCAE* ON INTRACYTOPLASMATIC CYTOKINES (IC) PRODUCTION IN TUBERCULOSIS PATIENTS (TBP) Martinel-Lamas D¹, Selenscig D¹, Fiorenza G¹, Donnari M², Vogué C², Dlugovitzky D¹. ¹Inmunol. FCM-UNR. ²H. Carrasco.

E-mail: diegomartinell@yahoo.com.ar

We previously demonstrated that immunotherapy with three doses of *M. vaccae* (*Mv*) induces changes in clinical and immunological parameters in TBP as Respiratory Burst, CD11b expression, IL8 and TNFa receptors of neutrophils and mononuclear cells. In this trial the effect of this immunomodulatory therapy on IC IL2, IL4 and IFNy production by lymphocytes was studied. Twenty two TB (HIV-) patients, both sexes (39±12.5 years, SD) were studied. 12 TB patients received DOTS therapy and one dose monthy of Mv (0.1 ml of heat killed bacillus (10 mg/ml NCTC 11659 strain), suspended in saline borate buffer, i.d.; provided by Dr. J. Stanford, London University, U.K) for 3 months. Twelve TB patients with the same characteristics of the above described, received only DOTS and Placebo (Pl) 0.1ml saline. After clinical, radiological, and bacteriological diagnosis a blood sample was obtained i.v. (S0), for immunological studies, and TBP received the first Mv or Pl dose at the beginning of treatment. In the 2 following months TBP received the same therapy and the different kinds of exams were also done. Heparinized blood samples were obtained S0, S1, S2 and S3 (posttreatment). To estimate IL2, IL4, IFNy production, 15 ml of heparnized blood was obtained and IC were evaluated by Flow Cytometry (FACS-CALIBUR) previously stimulated or not with PMA (Phorbol Myristate Acetate) and Ionomicine (B: basal, S: stimulated). IL2 and IFNy production increased and IL4 decreased in the sucesives samples in TBP, receiving either Mv or Pl, being higher in TBP receptors of Mv. These data also show the immunomodulatory effect of Mv, as the stimulus induced bigger cytokines production in TBP treated with Mv, B and S.

75.

RELATIONSHIP BETWEEN NITRITES AND IFN γ , IL2 AND TNF α LEVELS IN CULTURE SUPERNATANT (C.S.) OF MONONUCLEAR (MN) AND POLIMORPHONUCLEAR (PMN) CELLS OF TUBERCULOSIS PATIENTS (TBP) TREATED WITH *M. VACCAE*

Martinel-Lamas D¹, Selenscig D¹, Fiorenza G¹, Donnari M², Vogué C², Dlugovitzky D¹. ¹Inmunol. FCM-UNR. ²H.Carrasco.

E-mail: diegomartinell@yahoo.com.ar

It has been previously demonstrated that M. vaccae (Mv) immunotherapy induced in TBP a stimulation of several activation parameters of MN, PMN and lymphocytes. In this trial the effect of the same therapy on nitrites and IFNy, IL2 and, TNF α levels in MN and PMN c.s. and their relationship was analyzed. Twenty four TBP (HIV-), both sexes (36±15.5 years, SD) were studied. Twelve TBP received DOTS therapy and 1 dose/month of Mv (0.1 ml of heat killed bacilli (10 mg /ml NCTC 11659 strain), suspended in saline borate buffer, i.d.; provided by Dr. J. Stanford, London University, U.K) for 3 months. Twelve TBP with the same characteristics of sex and age, received DOTS and Placebo (Pl 0.1ml saline). After diagnosis 25 ml of heparinized blood were obtained i.v (S0), for immunological studies, and TBP received the first Mv or Pl dose at start of treatment. In the following 2 months TBP received the same therapy and clinical and immunological assessments were done in blood samples (S0, S1, S2 and S3 post-treatment). Cytokine levels were evaluated by ELISA (R&D) and nitrites levels by Griess method. Nitrites and cytokines levels significantly increased in the successive samples, and they were also higher than in Pl receptors. Nitrites concentrations have a positive correlation with IFNy, IL2 levels and a negative correlation with TNFα levels (MN: R²: 0.827, 0.809, and 0.815 respectively. PMN: R²: 0.85, 0.7and 0.88 respectively). These correlations were not observed in Pl receptors. The increased production of IFN γ , IL2, TNF α and nitrites and their correlations confirm the immunomodulatory effect M. vaccae in TB.

76.

BIOFLORA IN THE PREVENTION OF INDOMETHACIN INDUCED GASTROINTESTINAL LESIONS, IN RATS

Villarruel R, Laudanno OM, Godoy AR, Cesolari JAM. Experimental Gastroenterology. Histology and Embryology Chair, School of Medicine. University of Rosario (NUR). E-mail address: roro villarruel@hotmail.com

Bioflora probiotic (Bio) is an anti-inflammatory administered orally and subcutaneously (SC), with viable or nonviable bacteria. Aim: to study Bio's mechanism of action in the prevention of severe gastrointestinal lesions produced by Indomethacin (Indo).

Material and method: in randomly selected female Sprague-Dawley rat groups (n:10/group), water ad-lib. and 24h fasting. Experiments: Group I: Indo 30 mg/Kg. SC every 24h for 2 days. II: Bio 1 ml (1.3 x 10.7 alive bacteria) orogastric every 12h for 2 days and Indo. After ether overdose, laparotomy, gastrectomy, and enterostomy were performed; the percentage (%) of the gastric necrotic and intestinal erosive area in mm2 was measured and cuts were obtained for studies with Myeloperoxidase (MPO), intestinal bacteriologic cultures (CFU) and mesenteric lymph ganglions and 4 cm of terminal ileon were resected for histochemical studies - T lymphocytes (CD4+). Statistics: Student's "t" test and ANOVA. Results: % of necrotic gastric area I: 65 ± 7 . II: 7.5 ± 1.3 (p < 0.001). MPO I: 416 \pm 31 mg/protein. II: 30 \pm 7 (p < 0.01). Small Intestine erosions(ID) I: 380 ± 31 . II: 41 ± 6 (p < 0.01). MPO I: 435 ± 45 . II: 51 ± 11 (p < 0.01). Cultures ID I: $7.5 \pm 3.5 \times 10.10$. II: $2.3 \pm 0.8 \times 10.10$ 10.5 (p < 0.01) And in mesenteric lymph ganglions I:. 9 (+) 1 (-). II: 8 (-) 2 (+) (p < 0.02) and T ileon lymphocytes I: 0.5 ± 0.1. II: 4 \pm 1 (p < 0.01). Conclusion: Bio prevented severe gastrointestinal lesions induced by Indo, neutrophil infiltration, intestinal bacteria overdevelopment and increased intestinal T lymphocytes (CD4+).

79.

INFLUENCE OF SERUM FIBRINOGEN AND HIGH AND LOW DENSITY LIPOPROTEIN ON BLOOD VISCOSITY IN HEALTHY MENOPAUSAL WOMEN

Carrara PA, Spengler MI, Mengarelli G.

Cátedra de Física Biológica. Facultad de Ciencias Médicas.UNR. E-mail: npablont@hotmail.com.

During the menopause, the cardiovascular risk is higher. The increase of blood viscosity (η_{i}) could accelerate the atherothrombosis due to fall of blood flow and the extension time of residence of atherogenic particles near the endothelium. Previous studies have shown the effect of fibrinogen (F) on η_{e} and the relation of LDL-Cholesterol and HDL-Cholesterol on η_{a} . The objective of this work was study the influence of F and LDL-Cholesterol and HDL-Cholesterol on η_c . In 40 menopausal healthy women (50 ± 7 age), previous written consent of them, η was measured with cone – plate viscosimeter, F by gravimetric method and HDL-cholesterol and LDL-cholesterol by colorimetric method. We use Pearson coefficient for the statistical analyses. We observed a positive correlation between: F and η_{c} (r=0,4713, p<0,01); LDL-Cholesterol and $\eta_{c}(0,5168, p < 0,001)$. Negative correlation between HDL-Cholesterol and η_c (r=-0,4481, p<0,01). These results show that the F and LDL-Cholesterol have a positive effect on η_{e} , but the HDL-Cholesterol effect is opposite. The increased of F and LDL-Cholesterol are considered cardiovascular risk factors. HDL-Cholesterol is a protective factor. These events do that the menopausal women have a similar cardiovascular risk compared with men of same age.

78.

EFFECT OF NIFEDIPIN ON LIPIDIC FLUIDITY MEM-BRANE ON ERYTHROCYTES IN WOMEN WITH SYS-TEMIC SCLERODERMA

Carrara PA³, Spengler MI¹, Bertoluzzo SM¹, Leroux MB², Svetaz MJ⁴. ¹Cátedra de Física Biológica, ²Cátedra de Dermatología, ³Cátedra de Fisiología, Facultad de Ciencias Médicas; ⁴Sección Inmunidad Celular, Departamento de Bioquímica Clínica, Facultad de Ciencias Bioquímicas y Farmacéuticas, Universidad Nacional de Rosario. E-mail: npablont@hotmail.com

Systemic Scleroderma is an autoimmune disease, due to a connective tissue alteration characterized by extracellular matrix increase in the skin and internal organs. It is already known that the Raynaud's phenomenon and the microcapillary obliteration lead to ischemia and peripheral tissue injury The objective of this work was study the effect of nifedipin treatment on membrane lipidic fluidity of erythrocytes (f) in 47 women with systemic scleroderma $(37 \pm 6 \text{ years})$. 22 of them were taking colchicin (1mg/day) and nifedipin (dihidropiridinic calcium blocker) (30mg/day). The rest only treated with colchicin (1mg/day). f was measured by fluorescence polarization method with TMA-DPH (Trimethylammonium-Diphenylhexatriene) as fluorescence marker. The anisotropy (r) is inversely related with f. r is associated with the restriction movement of marker into lipidic membrane. The results shown that the anisotropy of patients were taking nifedipin and colchicin was lower that the patients were taking only colchicin. (r= $0,164 \pm 0,03$ versus r=0,201 \pm 0,020; p<0,001). It's likely that the nifedipin reduce the intracytoplasmatic calcium and improve the f. This effect, together with the classical vasodilatation action, allows that nifedipin a drug of first choice for the Raynaud phenomenon in this disease.

EFFECTS OF A POSTWEANING ALLOXAN INJECTION ON NADPH NEURONS FROM AUERBACH PLEXUS IN SPRAGUE-DAWLEY RATS THAT AFTER A WEEK DID NOT END IN ALLOXANIC DIABETES

Podestá MF, Hisano N.

Cátedra de Histología y Embriología. Facultad de Ciencias Médicas. Universidad Nacional de Rosario. E-mail: flor_podesta hotmail.com

Alloxan is a well known toxic for β cells of Langerhans islet. A single dose of it could be followed by hyperglycemia, resistance or death. As we have reported a 'toxic' effect of alloxan in 'M' rats, we analyzed the effect of alloxan in NADPH+ neurons of Auerbach plexus, in Sprague Dawley alloxan resistant rats. 10 male Sprague Dawley rats 23 days old were separated into the following experimental groups: 'a' 6 rats injected with alloxan (24mg/100g CBW); 'b' 4 rats injected with destilled water (1cc/100g CBW). Glycemia was performed after 48 h and at autopsy. After sacrificed with ether overdose autopsy was performed at 30 days old. Small intestines were removed and washed with chilled PBS, fixed in 4% paraformaldehyde in PBS, and processed for NADPH histochemistry. Results are expressed in media \pm SEM and were compared with the test 't' of Student, finding no significatives differences for: corporal body weigh 'a' 111.12 \pm 3.7 mg, 'b' 107.33 \pm 2.98 mg; small intestine weigh 'a' 7.68 \pm 0.31 mg, 'b' 8.43 \pm 0.9 mg; small intestine length 'a' 109.33 \pm 1.78 mm, 'b' 101.5 \pm 4.93 mm; neuronal count 'a' 39.41 ± 3 neurons/mm2, 'b' 38.27 ± 1.71 neurons/mm2. The histological reticular structure of the plexus and the neuronal count are conserved, this supposed that alloxan do not have a direct toxic effect on those structures.

80.

AUERBACH PLEXUS STRUCTURE WITH NADH HIS-TOCHEMISTRY IN A LINE OF OVERWEIGHED RATS. EFFECTS OF DIETARY RESTRICTION

Hisano N¹, Posadas M².

¹Cátedra de Histología y Embriología. ²Cátedra de Biología. Facultad de Ciencias Médicas, UNR.

Beta rats are overweighed, and became diabetic rats at 8 months old. We report results from Auerbach's structure with ageing and dietary restriction. 1.- Male beta rats from 2,4,8,12 and 18 months old, fed ad libitum, with balanced food. 2.- beta male rats fed a restricted balanced food (30%) from wealing to 8 months old. 3.male beta rats fed a restricted balanced food from 60 to 120 days old. Animals were sacrificed with ether overdose. At autopsy, gut was dissected out, washed with PBS, weighed and measure their lengths. Segments of yeyunum, distal and proximal colon were NADH histochemically stained. 1.- In ageing studies, from 2 months old small disruption of plexual structure are described, disruption augmented with ageing. Alteration in yeyunum are more severe than colon. 2.-Dietary restriction till 8 months old: reticular structure of Auerbach plexus were conserved, similar to "control rats" (i.e. Wistar). 3.- Dietary restriction from 60 to 120 days old, produced reticular disruption from Auerbach plexus, lesser than mature animals.

In beta rats, Auerbach plexus histochemically stained with NADH, disruption of reticular image are observed, more severe and extended with ageing. Dietary restriction delays such phenomenon, more effective when early dietary restriction are started.

FUNTIONAL PROPERTIES OF ENZYMATIC HYDROLY-SATES FROM AMARANTH HIPERPROTEIC FLOUR *Ballerini G, Ciappini MC, Martinet R.*

Cidta UTN FRR. Zeballos 1341. Rosario. Sta. Fe. E-mail: gballerini@ciudad.com.ar

Enzimatic protein hydrolysis is a common procedure for improving functional properties of proteins. It improves nutritional characteristics and solubility compared to the original proteins. Amaranth is a pseudocereal wich contains 18.0% of protein. From this small seed, a high hiperproteic flour is obtained by differential milling. Amaranth defatted hyperproteic flour was hydrolyzed using proteases, Alcalase and/or Flavourzyme, at 50°C and pH of 7.5. Treatments with proteases were of 20, 60, 120 and 180 min; when both proteases were used, Alcalase was added at the beginning and Flavourzyme was added after 1/3 of the total treatment time had passed. Hydrolysates with 58.3% degree of hydrolysis was achieved. The increase of solubility is particularly high for pH between 3 and 6. The lowest increase was observed with Flavourzyme and the highest with Alcalase plus Flavourzyme treatments. In this case, small structure were formed which improved the foam capacity, although the stability of foam are poor. The total protein was around 93 and 95% in all treatments. The amino acid compositions of hidrolysates obtained with proteases were very similar but they showed an amino acid pattern with important diferences compared to Amaranth defatted hiperproteic flour. The protein hydrolysates obtained were poor in lysine and leucine, although they reached the FAO/WHO requirements for 2-5 year old.

82.

ACTION OF R-MET-HU-G-CSF AND R-HV-ALFA-ERITROPOYETIN IN THE REPARATION OF THE GAS-TRIC ULCER BY ACID ACETIC IN RATS

Bedini O, Traverso M, Naves A, Laudanno O, San Miguel P, Cesolari J, Godoy A.

Exp. Gastroenterology. FCM, UNR. E-mail: bedinioa@arnet.com.ar

Aim: to evaluate the action of two drugs stimulating the progenitor cells of the MB, injected SC before the induction of the GU by AA in different doses. Method: groups of wistar rats n=3, 200g, fasted 24 hours, water ad libitum, were submitted to these experiments: A) NEU,30 ng/Kg SC and waited for 5 days. B) ERI, 400 ug/kg SC and waited for 5 days. C) NEU and ERI SC and waited for 5 days. D) NEU, SC, 30 ug/Kg and waited for 8 days. E) ERI 400ug/kg and waited for 8 days. F) NEU and ERI SC and waited 8 days. Laparotomy (L) was performed previous anesthesia, The E was externalized and GU was induced, with AA in perithonel gastric sub serous antral form closed the L. For 3 days, the rats received food and water ad-libitum. Then, they were sacrificed by ether overdose. E was removed, % of the necrotic area was established, histology (H) was done. Results: 1) Phys. Sol. 2) AA 25% (3.5 \pm 0.5)mm 3) AA 60% (5.0±1.5) 4) AA 100% (7.5 ± 2.3) Group A) NEU– AA 25% (3.3 ± 0.5) B) ERI – AA 25% (3.2 ± 0.5) C) NEU+ ERI – AA 25% (2.0 \pm 0.5) (P< 0.5) <u>D</u>, <u>E</u>, <u>F</u> (0.0). <u>Conclusions</u>: NEU (A) and ERI (B) did not show macroscopic reparation (MR). NEU and ERI (C) simultaneously reduced the size of the ulcer, while the H showed intense proliferation of fibroblasts (F) and vessels in neoformations. NEU and ERI, A, D in separate form (D and E) or in simultaneous (F) 8 days before, showed complete MR of the GM, the H showed intense reparative process with active F and neoformative vessels. NEU like ERI along stimulating the stem cell provoked MR of the GU by AA producing fibroblastic reactivation.

83.

EFFECT OF A BIOLOGICAL RESPONSE MODIFIER ON CELLULAR DEATH MECHANISMS AT DRYING OFF

Dallard B, Heffel S, Ruffino V, Baravalle C, Calvinho L. Cátedra de Histología y Embriología. Facultad de Ciencias Veterinarias. Universidad Nacional del Litoral.

The objectives of this study were to describe the effects of a single intramammary infusion of a LPS-based biological response modifier (BRM) on cellular death mechanism in uninfected and Staphylococcus aureus-infected bovine mammary glands during involution. Three groups of 12 cows, each one including 6 S. aureusinfected and 6 uninfected, were infused in two mammary quarters with BRM or placebo and sacrificed at 7, 14 and 21 d of involution. In infected quarters, BRM treatment produced a significant increase of Bax staining at every observation period. In uninfected quarters, BRM treatment produced an increase in number of epithelial and stromal cells staining with active caspase-3 at 7 d of involution and a significant decrease in percentages of terminal deoxynucleotidyl transferase dUTP nick end labeling (TUNEL)positive epithelial cells at 14 and 21 d of involution; while in stromal cells the increase in caspase-3 staining and TUNEL-positive cells was significant at every observation period. In infected quarters BRM treatment induced a significant increase in number of epithelial and stromal cells staining with caspase-3 as well as in percentages of TUNEL-positive epithelial cells at 7 and 14 d of drying off; while in stromal cells the percentages were higher than controls at every sampling period. These results provide new insights into the mechanism of mammary cell death in uninfected and S. aureus infected bovine mammary gland during involution and illustrate the effects of LPS-based BRM stimulating non specific defences and promoting mammary involution.

84.

STRUCTURAL 3D ANALYSIS OF AGGLUTINATED ERYTHROCYTES USING CELLSCAN MICROSCOPY

Danieli V¹, Riquelme B¹, Dumas D², de Isla N², Stoltz JF². ¹Física, FCByF y Óptica Apl. Biol., IFIR (CONICET-UNR), Rosario, Argentina. E-mail: bibiana_riquelme@yahoo.com.ar ²Ingénierie Cellulaire et Tissulaire UMR CNRS 7563. Faculté de Médecine. 54500 Vandoeuvre lès Nancy. France.

The interaction mechanisms involved in cellular adhesion are of particular interest for biological and medical sciences, being the cell-cell adhesion a crucial phenomenon for the relationship of the cell with its environment. The objective of this work was to study the human erythrocyte adhesion through anti-glycophorin monoclonal antibodies using CellScan fluorescence microscopy. A. B and C Glycophorins (GP) are abundant transmembrane integral proteins that greatly contribute to the superficial negative charge of erythrocytes. The distribution of four different murine anti-GP antibodies in the interfacial region of an agglutinated of two erythrocytes was studied by means of digital analysis of 12 bit images obtained with an inverted epifluorescence Olympus IX-70 microscope equipped with acquisition system CellScan EPR (Scanalytic's IPLab). A parameter related with the fluorescence intensities in the contact area between both cells and also in their surfaces were determined using IPLab software. The analysis of these intensities for each image (step of 0.25µm) showed that the distribution of these antibodies was 30% higher in the contact region, for all the antibodies studied. This is a useful tool to understand the mobility of antibodies linked to the cellular surface as part of the cellular adhesion phenomenon.

PERCENTAGE COMPOSITION OF FAT AND PROTEIN IN MILK FROM A DAIRY FARM IN THE SOUTH OF SANTA FE PROVINCE

Dumas A^1 , Musso G^1 , Smacchia A^2 .

¹Advisor: ²Laboratorio de Química Biológica Facultad de Cs. Veterinarias. CIUNR. UNR. E-mail: alfredodumas@hotmail.com; asmacchi@hotmail.com

The different feeding systems in dairy farms may have a great impact on protein and fat concentration in milk. Several tools help producers modify milk components. The strategies that improve ruminal activity are capable of rising milk production and its components. To achieve the latter, it is necessary to know the fat and protein composition and variability in each farm. Data from milk production registers allow the identification and selection of animals or groups of animals that have fat and protein at a higher level than the mean in each herd. The aim of this work was to evaluate the composition and variation of milk fat and protein in a dairy cattle herd from a farm in the south of Santa Fe province. Milk samples were taken and fat and protein percentage was determined for two years, being day 0 the calving day of each animal. Registers (n=946) were taken once a month in all milking cows (n=80) that were between their first and third lactation. The dairy farm produced a mean total milk volume 23.1 liters with a 6.71 deviation. The percentage of butter fat (BF) and protein (CP) in the studied milk samples showed a mean and deviation (shown between brackets) of 3.52 (0.61) and 3.30 (0.20), respectively. Results show that both protein and fat percentage in milk had a rising tendency from calving day on and that protein percentage had a lower variation compared to that of butter fat.

86.

DRY MATTER RUMINAL DEGRADATION AND CHEMI-CAL COMPOSITION IN PASTURE

Figallo RM, Faienza HL, Pidello A, Smacchia AM.

Laboratorio de Química Biológica. Facultad de Cs. Veterinarias. CIUNR. UNR. E-mail: rfigallo@fveter.unr.edu.ar

The aim of this work was to study, in pastures, the relation between parameters that describe the kinetics of ruminal degradation of dry matter (DMRD) with its chemical composition. The pastures employed were oats, bromus, mellilotus alba, white clover, and alfalfa, dried at 60°C, ground and sieved through a 2mm mesh. Samples of 3 g DM of each pasture (17mgDM/cm²) were incubated in ASTM 230 nylon bags (pore size: 62 micron) for 0, 2, 4, 8, 12, 18, 24 and 48, in rumen (Meherz and Orskov, 1977). The chemical composition (g/100g DM): Organic Matter (OM), non-structural carbohydrates (NSC), Crude Protein (CP: N x 6.25), Lignine (Lig.), Neutral Detergent Fiber (NDF) and Acid Detergent Fiber (ADF) was determined according to AOAC (2000). DMRD data were adjusted to the Orskov and McDonald (1979) model: DMRD = a + b (1 - e^{-ci}); being: a: soluble fraction, b: slowly degradable fraction, and c: degradation rate. DMRD parameters and chemical composition were studied through correlation. Results are shown in the table.

		DMRE)	Chemical composition (g/100g MS)					
Pasture	а	b	с	OM	NSCs	RP	Lig.	NDF	ADF
Mellilotus Alba	25.8	49.5	0.100	90.0	5.7	20.3	5.6	42.0	31.9
White Clover	39.0	52.7	0.096	86.5	6.3	22.6	4.5	29.8	26.6
Oat	39.8	50.4	0.060	89.0	11.9	18.4	4.6	47.8	30.5
Bromus	36.0	56.1	0.095	81.3	2.8	21.7	7.4	59.2	40.9
Alfalfa	34.1	45.6	0.105	89.3	7.6	20.4	6.1	39.2	28.4

The only significant correlation that was seen was: Lignine with ADF (0.82; P<0.09). In the studied pastures no significant correlations were seen between the estimators of parameters of ruminal degradation kinetics and the chemical composition, as in other type of samples studied.

87.

DRY MATTER RUMINAL DEGRADATION AND CHEMI-CAL COMPOSITION IN RESIDUE CROP

Figallo RM, Faienza HL, Pidello A, Smacchia AM.

Laboratorio de Química Biológica. Facultad de Cs. Veterinarias. CIUNR. UNR. E-mail: rfigallo@fveter.unr.edu.ar

The aim of this work was to study, in residue crop, the relation between parameters that describe the kinetics of dry matter ruminal degradation (DMRD) and the chemical composition. The samples consisted of wheat straw, corn, sorgum and soya residue crop, dried at 60° C, ground and sieved through a 2mm mesh. 3 g DM of each sample ($17mgDM/cm^2$) were incubated in ASTM 230 nylon bags for 0, 2, 4, 8, 12, 18, 24 and 48, in rumen (Meherz and Orskov, 1977). The chemical composition (g/100g DM): Organic Matter (OM), non-structural carbohydrates (NSC), Crude Protein (CP), Lignine (Lig.), Neutral Detergent Fiber (NDF) and Acid Detergent Fiber (ADF) was determined (AOAC, 2000). DMRD data were fitted to the Orskov and McDonald (1979) model: DMRD = a + b (1 - e^{-ct}). DMRD parameters and chemical composition were studied through correlation.

		DRMS	5	Chemical composition (g/100g DM)					
Crop Residue	а	b	с	OM	NSC	СР	Lig.	NDF	ADF
Wheat Straw	3.6	93.1	0.010	91.8	0.9	4.2	6.0	81.9	57.3
Corn	17.8	68.3	0.016	92.0	1.3	5.2	6.0	80.7	50.2
Soya	16.9	29.9	0.031	87.0	3.5	10.7	9.6	60.5	54.8
Sorgum	14.8	58.4	0.017	89.7	2.9	7.2	5.2	70.0	45.1

The significant correlations were: fraction b with CP (-0.97), NDF (0.93), NSC (-0.92) and fraction c (-0.97); c with OM (-0.94); NDF (-0.93) and CP (0.98). The b fraction related straightforwardly with NDF and inversely with CP and NSC and the degradation speed with CP and inversely with OM and NDF. These results show that NDF is the main component of the slowly degradable fractions and that it is responsible for the slower degradation speed of crop residues.

88.

STATISTIC NONLINEAR STUDY APPLIED TO PHOTO-METRIC SERIES OF RBC UNDER SHEAR STRESS

Korol A¹, Riquelme B¹, Forest P¹, Darrigo M¹, Rosso O², Valverde J¹. ¹Facultad Cs. Bioquímicas, IFIR (CONICET-UNR). ²Facultad Cs. Exactas, UBA. E-mail: korol@ifir.edu.ar

By using nonlinear methods and time-frequency analysis we characterized the cell behaviour under shear stress. On the basis of fractal approximation for ordinary (OBM) and factionary 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.

We have the time series but we have not the differential equations for the process so it could be modelled by a trajectory in phase space, using delay coordinates we generate two different phase space for the photometrically recorded series to diabetics patients and also for healthy donors samples.

We used time delay suggested by Takens, May algorithm, used Pearson for the correlation evaluation, and maximum Ljapunov.

Here we report studies on 11 donors: 6 diabetic patients and 5 healthy donor non smokers and non alcoholic individuals. The photometrically recorded series were obtained by ektacytometry during creep and recovery process over several millions of cells. On calculating May algorithm, the healthy donor samples show white noise behaviour while diabetic patient samples were deterministic chaos. On obtaining Ljapunov exponent for diabetic patients: $0.022 < L_{\rm E} < 0.266$, while for healthy donors: $0.232 < L_{\rm E} < 0.266$, that is 10 times more.

This information could be used in evaluating clinical aspects of erythrocyte rheological properties. Further studies with larger and well defined patient populations are in process.

A COMPARATIVE STUDY OF MUSCULAR INSERTIONS IN SHOULDER GIRDLE AND UPPER MEMBERS IN TWO PREHISTORIC HUNTER-GATHERER POPULATIONS

Kozameh L^{1,2}, López M², Testa N², Tessone A³. ¹CIUNR, ²Department of Bioanthropology, School of Humanities and Arts, UN de Rosario. ³INGEIS-CONICET.

 $E\text{-mail: liviak} @arnet.com.ar \ ; \ bioantropologia @unr.edu.ar \\$

Human populations exhibit two inseparable natures, the biological and the sociocultural. The most remarkable aspect is perhaps that they have a unique adaptive mechanism, namely culture. Activities carried out in daily life are reflected in multiple cultural and biological characteristics. Evidence of the latter is found in the skeletal remains of individuals who formed extinct societies. In the case of mobile societies, the ergological evidence is scant because mobility is a limiting factor, which is why skeletal analysis becomes fundamental as a source of information in adaptive studies of this type of society. The objective of this study has been to carry out a comparative functional analysis of anatomical and biomechanical traces of the shoulder girdle and upper members in the human remains of two prehistoric hunter-gatherer populations exhumed in archeological excavations. One of these populations inhabited Isla Grande in province of Tierra del Fuego, and the other the southern part - humid soil - of the province of Entre Rios. The methodology applied involved macroscopic observations and measurements with sliding and spreading calipers and graduated steel tape. While it is true that highly developed muscular insertions are found in both populations they occur in different anatomical positions and reveal differential actions and movements that are possibly related to the activities involved in the subsistence of these populations.

90.

SILAGE OF DISCARDED SWEET POTATO (Ipomoea batatas L) OR ANIMAL FEEDING. PRELIMINAR STUDY

Manresa N, Peronja N, Smacchia AM.

Lab. Quím. Biol. FCVeterinarias CIUNR-UNR. E-mail: asmacchi@unr.edu.ar

The aim of this work was to study the viability of preserving sweet potato in the form of silage, employing the Arapey variety which is mostly grown in San Pedro (Argentina), to feed animals. A preliminar sweet potato ensilage test was carried out at a laboratory scale. The pH evolution was monitored during the process and the nutritious property of sweet potato was compared before and after 60 days of ensilage. The chemical composition: humidity (H), fiber (FC), ether extract (EE), ashes (As), Protein (SPP), carbohydrates (CH), total digestible nutrients (TDN) was analyzed and expressed as % of dry matter (DM). The energetic contribution was estimated: gross energy (GE), digestible energy (DE) and metabolisable energy (ME), and it was expressed as Kcal/kg DM. The pH mean value and deviation (shown between brackets) recorded on days 4. 9, 51 and 62 were: 4.14 (0.12); 4.40 (0.03); 3.74(0.01) and 4.15 (0.13), respectively. The sweet potato composition and its silage are:

			\sim										
Sweet	mII	DM	Н	FC	EE	As	TN	SPP	CH	TDN	GE	DE	ME
silage	рп					%]	Kcal/K	g
Initial Final	—	30	70	6	0.8	3	1.25	7.9	82	84	3970	3710	3220
(day 60)	4.15	31	69	5.5	0.7	4.4	1.7	10.6	79	83	3970	3660	3190
Initial		33	-14	-83	-12.5	46.6	36.0	34.2	-37	-12	0.00	-13	-0.9

Arapey sweet potato ensilaged showed a lower FC, EE concentration and a higher SPP and As concentration. TDN, DE and ME values were slightly lower. These preliminary results encourage the repetition of this study at another experimental scale and the continuation of studies to evaluate the nutritional quality of the preserved product. 91.

PHYSIOLOGICAL PROFILE OF Pseudomonas fluorescens 92 AND THE STRAINS P. fluorescens 92^{RT} AND P. fluorescens $92^{RTcG/p}$

Perotti EBR¹, Fracchia L², Pidello A¹, Martinotti MG². ¹LQB-FCV-CIUNR- Universidad Nacional de Rosario, Argentina. ²DiSCAFF, Università degli Studi del Piemonte Orientale "Amedeo Avogadro", Italia. E-mail: eperotti@unr.edu.ar

The aim of this work was to study the functional physiological profile (FPP) of the bacterium that increase the plant growth (PGPR, plant growth promoting rhizobacteria), that produce siderophore, produce indolacetic acid and act as phosphate solubilizer. Previously, we observed that in the Argiudoll soil inoculated with *P*. fluoresces 92, the FPP was lower in relation to control soil. In this work the FPP was studied by BIOLOG EcoPlate[™], a system that use 31 different carbon sources frequently utilized by soil rhizospheric microorganisms. The results indicated that: (i) the three strains have the same FPP; (ii) substrates that contribute to the modification of the FPP profile in the Argiudoll ecosystem are metabolizated by the inoculated bacterium, in special the following singly compounds: L-Threonine, L-Asparagine, L-Glutamine; the D-Galactonic and Pyruvic methyl ester acids and Mannitol. The results suggest that the characteristics of these compounds could affects the genetic expression of P. fluorescens 92, 92RT and P. fluorescens 92^{*RTcGfp*}, and also produce anti-microbial factors that affects the FPP profile of the total microbial community.

This work was realised in the framework of the scientific bilateral Co-operation between Argentine and Italy supported by SeCyT and the Italian Ministry of Foreign Affairs N°IT/PA05-BVII-085.

92.

LOCALIZATION OF TYPE II TGF-BETA RECEPTOR IN MESENCHYMAL STEM CELLS FROM BONE MARROW BY 3D FLUORESCENCE MICROSCOPY

Riquelme B¹, Dumas D², de Isla N², Muller S², Stoltz JF². ¹Física, FCByF y Óptica Aplic. Biol., IFIR (CONICET-UNR). ²Ingénierie Cellulaire et Tissulaire UMR CNRS 7563. Faculté de Médecine. 54500 Vandoeuvre lès Nancy, France.

Transforming growth factor TGF-beta is a family of multifunctional cytokines controlling cell growth, differentiation, and extra cellular matrix deposition. The biological effects of TGF-beta are mediated by type I (TBR-I) and II (TBR-II) receptors. In this work we presented a preliminary study for characterize the localization and mobility of TBR-II using mesenchymal stem cells derived from bone marrow and grown on "Lab tek" culture chambers. Localization of receptors was attained from indirect immuno-labeling of TBR-II (A11017 Invitrogen, Alexa488TM) on mesenchymal stem cells and using DiI for labeling of membranes and lipid vesicles. The images were obtained with a confocal microscope SP2-AOBS (Leica) and analyzed with LCS Leica and IPLab-BD Biosciences software. The results show that TBRII is localized on cellular surface and in the cytoplasm internalized in cylindrical non-lipid vesicles of 3 to 5 µm of diameter and 8 to 10 µm of longitude. These vesicles can be acting as reservoir or recycle function for TBRII and their internal mobility in relation to the significant mobility of surface receptors could be significant for MSC cells differentiation. These results concerning dimension of vesicles may be very useful as preliminary study for FCS analysis and for modelization of signal transduction during the MSC cells differentiation.

94.

RELATIONSHIP BETWEEN OXIDATIVE STRESS AND ATPase ACTIVITY IN RATS WITH PARTIAL HEPATEC-TOMY

González MA, Contini M, Millen N, Mahieu S. LIFE. FBCB.

Previous studies have shown how injury is associated with hepatic oxidative stress. Oxidative stress .level (balance between pro-oxidant and antioxidant) is under genetic control and increases with cell differentiation. The objective of this study was to assess oxidative stress parameters and membrane ATPase activity of regenerating livers. Adult male Wistar rats (n = 5 / group), average weight 300 g, were divided into three experimental groups A: controls; B: hepatectomized 48 hours prior to the experience, C: hepatectomized 168hours before the experiment. It was determined catalase (CAT) activity, glutathione peroxidase (GSH-Px), lipoperoxidation (LPO), ATPase activity, and the content of glutathione (GSH). The results were expressed as the mean \pm SEM (* significant difference p <0.05). LPO (nmolMDA/g liver: C 275.5 ± 38.5, HP 48hs 573.2 ± 75.5*, HP 168hs 205.5 \pm 48.2. GSH (µmol/g liver.); C 3.47 \pm 0.14, HP 48hs 2.01 ± 0.12 *, HP 168hs 3.07 ± 0.11. GSH-Px(U/mg prot); C82.84 ± 7.05, HP 48hs 45.2 ± 3.3*, HP 168hs 93.84 ± 7.11; CAT (U/mg prot); C138.8 ± 3.46, HP 48hs 11.33 ± 0.3*, HP 168hs 110.7 \pm 2.3; ATPase (µmol Pi/ h mg prot), C 5.44 \pm 0.05, HP 48hs 2.11 \pm 0.02*, HP 168hs 4.25 \pm 0.17. The partial hepatectomy at 48 hs significantly decreased the activities of GSH - Px and CAT antioxidant enzymes as well as the content of GSHwith a significant increase in the LPO, showing the presence of oxidative stress. Moreover, the activity of the enzyme membrane ATPase declined 61% at 48 hours while at 168 hs their values were normal. These findings might suggest that the harmful effects on this membrane enzyme in regenerating livers could be related to oxidative stress induced liver injury.

EXPRESSION OF IGF-1 IN GOATS AND SHEEP MAMMARY GLAND AND THEIR RELATIONSHIP WITH MILK PROTEIN

Romano G, Dallard B, Maiztegui J, Moreyra E, Miller RJ. Cátedra de Fisiología e Histología y Embriología, Facultad de Ciencias Veterinarias, Universidad Nacional del Litoral. Esperanza, Santa Fe.

The presence of Growth Factors in milk takes the first place in the speculation that they are synthesized and also transported by the mammary epithelium. In recent studies it has been determined that in bovine, plasmatic IGF-1, is related with high concentrations of to lactoalbúmina and other proteins of the milk under handling conditions and controlled feeding. The proteins of the milk are of high biological value and of high impact in the industrialization of milk. The flocks of goats and sheep in our region, they are fed with natural prairies that present seasonal variations without being supplied most of the times. The aim of this study was to determine the relationship among the percentage of immunostaining area for IGF-1 in mammary tissue and milk proteins. Mammary tissue was obtained from 10 goats and sheep in distinct periods of lactation. Parenchymal tissue was aseptically excised using the "quoin" incising technique. The biopsies were processed according to technique used to reveal the presence of IGF-1 in the mammary tissue (IHQ) method (ABC). Simultaneously milk was extracted in form manual, for determination of proteins by means of Kjeldahl in the following periods: 5; 10-15; 20-25; 70-80 and 120 days post childbirth. The correlation coefficient was obtained by the method of Pearson (n=50, 10 animals during 5 periods) and coefficient R2. The correlation between percentage of immunostaining area for IGF-1 and the percentage of total protein in milk of sheep and goats was of 0.826 (R2=0,682) and 0.183 (R2=0,034) respectively. It is observed that in sheep an important relationship exists among both variables.

AUTHOR INDEX

Α		Catalani G	16	Floreani F	31
Accastello C	19	Ceccarelli N	19	Font MT	5, 6, 9, 10, 11, 12, 13, 17
Acosta I del L	38	Cereda YA	60	Forest P	88
Advínculo S	11	Ceruti MJ	60	Foresto P	37
Aguirre MC	67	Cesolari J	82	Fracchia L	91
Aimaretti S	71, 72	Cesolari JAM	69, 70, 76	François S	15
Alet NA	71, 72	Chapo G	3	Frascaroli MI	59
Almará A	30	Ciappini MC	81		
Alvarez L	64	Cointry E	20	G	
Álvarez M	11 12	Contini M	47 93	Galetti L	22 24
Alvarez MI	56	Contini M del C	45 46	Galosi CM	14
Amelong I	27	Cotorruelo C	38 30 40 41	Galvagni A	12
Amestov I	40	Cravero V	20	García Borrás S	38 30 40 41
Androu V	49 50	Crasetti D	20	García Montara N	50, 59, 40, 41
Andreu v	15	Closetti D	04	Careía D	A 51
Anthony L	13	D		Carola DCA	03
	14		27	Garcia RCA	22, 23, 24, 25
Antruejo AE	12	D Arrigo M	37	Garcia S	58
Armendariz M	67	D'Ottavio AE	65	Gaspari C	39
Arnesi N	48	Dater V	57	Gayol M del C	71, 72, 73
Arriaga S	30	Dallard B	54, 55, 83, 94	Gervasoni SI	7
		Daniele S	30	Geuna JD	43
B		Danieli V	84	Ghersevich S	30
Balagué C	31	Darrigo M	88	Ghirardi G	27
Ballerini G	81	de Candia LF	66	Giolito I	18
Baravalle C	83	de Isla N	84, 92	Giuliani S	18
Barberis I	21	De Gaudio M	57	Godoy A	82
Bedini O	82	Decot SV	1	Godov AR	76
Bellú S	58	Di Giusto G	34	Goldemberg AL	42
Bensoussan D	1	Di Loreto V	66	Gonzalez A	51
Berra HH	7	Di Masso R	9.10	González J	58
Bertoluzzo SM	16 78	Di Masso RI	5 6 11 12 13 17	González M	47
Bianchi MB	25	Di Mónaco R	39	González MA	45 46 93
Biasoli M	33	Di Sanio O	63	Gorosito M	9
Biava H	57	Díaz AO	42	Gorr C	61 62
Biondi C	38 39 40 41	Dimónaco R	30	Guadagnoli F	2
Bocapara P	J0, J7, 40, 41	Dinollaco K Disetti ME	50	Gurni A	2 64
Docalicia K	7	Discui ML Diagonitzka D	74 75	Gutiarraz S	72
Dollin KD	/ /1	Daminiahini A	7 4 , 75	Gutteriez 5	12
Doulasso U	41	Dominighini A Domnori M	74 75	п	
Bouvet B	29	Donnari M	/4, /5		40
Bouvet BR	28	Dottavio AM	11, 12, 13, 17	Hachuel L	48
Brance ML	66	Dottavio ME	16	Herrel S	83
Brandoni A	35, 36	Dumas A	85	Hinrichsen L	9, 10
Breccia G	19	Dumas D	84, 92	Hisano N	43, 79, 80
Brion C	14			Hure E	61, 62
Brun LR	66, 67	E			
Buccalossi G	9	Echenique C	33	J	
Buccalossi GG	10	Erbayo E	71	Jordán V	25
Bulacio L	18, 32	Escovich L	40		
Busmail LI	70	Espejo T	51	K	
		Espiñeira M	25	Kadi A	1
С		Espósito MA	20	Kerps G	27
Cabrni JM	32			Kohli A	51
Caferra D	3	F		Korol A	88
Caffarena G	61	Faienza HL	86, 87	Kozameh L	89
Caille A	30	Feldman RN	28		
Calvi BJ	69, 70	Felix MML	49	L	
Calvinho L	83	Feresin G	62	Larini S	31
Campi C	40	Fernández L	31	Laudanno O	82
Canet ZF	11	Fernández M del C	3	Laudanno OM	76
Carnovale C	64	Fernández P	17	Lehensohn N	37
Carnovale CE	56	Ferrari C	27	Leroux P	51
Carroro DA	טנ סר דר	Formero M	∠ / 6 A	Leroux MD	70
Carrora II	11,10	Finalla DM	04	Leioux MD	/0
Camilla MC	03	Figalio KM	00, 07	Lewis J	21
Carrino MIC	20	riorenza G	/4, /3	Liberatti DK	20

A28

AUTHOR INDEX

Librera JE	11	Р		San Miguel P	82
Lima B	62	Palopoli C	57	Sánchez AM	7
Longo MV	42	Panelo M	18	Santoro MI	60
López C	32	Paparella C	29	Sarchioni C	52
López JP	73	Paparella CV	28	Sarradell J	15
López M	89	Parente F	68	Scharovsky OG	6, 7, 8
López Anido F	20	Pascucci F	58	Selenscig D	74, 75
Lugano MC	56	Passarini M	27	Sender MB	21
Luján M	38	Passerini F	71, 72	Sgro MV	71, 72
Luque A	33	Pavesi A	29	Signorella S	57, 58, 59
Luquita A	64	Pelusa H	30	Silva PS	5
Lusardi M	24	Peronja N	90	Siragusa M	50
		Perotti EBR	91	Smacchia A	85
Μ		Picardi L	27	Smacchia AM	86, 87, 90
Magaró H	33	Picardi LA	26	Sortino M	32
Mahieu S	45, 46, 47, 93	Pidello A	86, 87, 91	Spengler MI	16, 68, 77, 78
Mahuad S	27	Pidone CL	14	Spies J	71, 72
Mahuad SL	26	Pisani GB	56	Spirolazzi M	19
Mainetti LE	7	Pituelli N	29	Spoleti MJ	50
Maiocco V	19	Podestá MF	79	Spoleti P	50
Maiztegui J	52, 53, 54, 55, 94	Poli GL	14	Stoltz JF	1, 84, 92
Manresa N	90	Pontoriero A	61, 62	Svetaz MJ	68, 78
Marani J	73	Posada M	43		,
Marchegiani M	69	Posadas M	80	Т	
Marchionni Basté E	26	Prado D	21,63	Taborda M	70
Marro AV	15	Prado DE	22, 23, 24, 25	Tapia A	62
Martin E	20	Pratta GR	26	Tarrés MC	3, 73
Martinel-Lamas D	74, 75			Tártara G	15
Martines A	13	0		Tellez TE	65
Martinet R	81	Quaglino MB	44	Tessone A	89
Martínez C	43, 70	Quiroga AD	56	Testa N	89
Martinotti MG	91			Tomat MF	4
Masoni A	4	R		Torres AM	34, 35, 36
Matar P	7	Rabinovich GA	8	Tosello ME	33
Maturo H	21	Racca A	38, 39, 40, 41	Tozzini R	4
Mc Cargo J	63	Racca L	33, 38, 40, 41	Traore M	1
Mengarelli G	77	Ramadán S	32	Traverso M	82
Menu P	1	Ramos L	32	Trossero C	61, 62
Miglietta MC	16	Rasia M	16		
Milanesi L	20	Re A	37	V	
Millen N	45, 46, 47, 93	Recarte M	41	17-1-14- NA	20
Miller RJ			71	v aldes IVI	30
Mogni V	54, 55, 94	Renzi J	30	Valenti J	30 9
	54, 55, 94 23	Renzi J Rico MJ	30 6, 8	Valenti J Valverde J	30 9 37, 88
Montenegro S	54, 55, 94 23 3	Renzi J Rico MJ Rigalli A	30 6, 8 66, 67	Valdes M Valenti J Valverde J Van Isseldyk F	30 9 37, 88 68
Montenegro S Montenegro SM	54, 55, 94 23 3 73	Renzi J Rico MJ Rigalli A Riganti JG	30 6, 8 66, 67 14	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD	30 9 37, 88 68 9, 10
Montenegro S Montenegro SM Monti L	54, 55, 94 23 3 73 61, 62	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B	30 6, 8 66, 67 14 84, 88, 92	Valees M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T	30 9 37, 88 68 9, 10 19
Montenegro S Montenegro SM Monti L Moreno D	54, 55, 94 23 3 73 61, 62 57	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B Rizzotto M	30 6, 8 66, 67 14 84, 88, 92 61, 62	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H	30 9 37, 88 68 9, 10 19 2
Montenegro S Montenegro SM Monti L Moreno D Moreyra E	54, 55, 94 23 3 73 61, 62 57 54, 55, 94	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B Rizzotto M Rodríguez A	30 6, 8 66, 67 14 84, 88, 92 61, 62 53	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H Vila H	30 9 37, 88 68 9, 10 19 2 27
Montenegro S Montenegro SM Monti L Moreno D Moreyra E Morosano M	54, 55, 94 23 3 73 61, 62 57 54, 55, 94 3, 4	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B Rizzotto M Rodríguez A Rodríguez GR	30 6, 8 66, 67 14 84, 88, 92 61, 62 53 26	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H Vilalta E Vilalta E Villar S	30 9 37, 88 68 9, 10 19 2 27 35
Montenegro S Montenegro SM Monti L Moreno D Moreyra E Morosano M Mosconi N	54, 55, 94 23 3 73 61, 62 57 54, 55, 94 3, 4 61, 62	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B Rizzotto M Rodríguez A Rodríguez GR Rodríguez Armesto	30 6, 8 66, 67 14 84, 88, 92 61, 62 53 26 R 52	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H Vilalta E Vilalta E Villar S Villarruel R	30 9 37, 88 68 9, 10 19 2 27 35 76
Montenegro S Montenegro SM Monti L Moreno D Moreyra E Morosano M Mosconi N Muller	54, 55, 94 23 3 73 61, 62 57 54, 55, 94 3, 4 61, 62 1	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B Rizzotto M Rodríguez A Rodríguez GR Rodríguez Armesto Romano G	30 6, 8 66, 67 14 84, 88, 92 61, 62 53 26 R 52 52, 53, 54, 55, 94	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H Vilalta E Villar S Villarruel R Vitelleschi MS	30 9 37, 88 68 9, 10 19 2 27 35 76 44
Montenegro S Montenegro SM Monti L Moreno D Moreyra E Morosano M Mosconi N Muller Muller S	54, 55, 94 23 3 73 61, 62 57 54, 55, 94 3, 4 61, 62 1 92	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B Rizzotto M Rodríguez A Rodríguez GR Rodríguez Armesto Romano G Romera BM	30 6, 8 66, 67 14 84, 88, 92 61, 62 53 26 R 52, 53, 54, 55, 94 13	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H Vilalta E Villar S Villarruel R Vitelleschi MS Vogué C	30 9 37, 88 68 9, 10 19 2 27 35 76 44 74, 75
Montenegro S Montenegro SM Monti L Moreno D Moreyra E Morosano M Mosconi N Muller Muller S Musso G	54, 55, 94 23 3 73 61, 62 57 54, 55, 94 3, 4 61, 62 1 92 85	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B Rizzotto M Rodríguez A Rodríguez GR Rodríguez Armesto Romano G Romera BM Ronco MT	$ \begin{array}{r} 30 \\ 6, 8 \\ 66, 67 \\ 14 \\ 84, 88, 92 \\ 61, 62 \\ 53 \\ 26 \\ R \\ 52, 53, 54, 55, 94 \\ 13 \\ 64 \\ 64 $	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H Vilalta E Villar S Villarruel R Vitelleschi MS Vogué C	30 9 37, 88 68 9, 10 19 2 27 35 76 44 74, 75
Montenegro S Montenegro SM Monti L Moreno D Moreyra E Morosano M Mosconi N Muller Muller Muller S Musso G	54, 55, 94 23 3 73 61, 62 57 54, 55, 94 3, 4 61, 62 1 92 85	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B Rizzotto M Rodríguez A Rodríguez GR Rodríguez Armesto Romano G Romera BM Ronco MT Rosa R	30 6, 8 66, 67 14 84, 88, 92 61, 62 53 26 R 52, 53, 54, 55, 94 13 64 53	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H Vilalta E Villar S Villarruel R Vitelleschi MS Vogué C W	30 9 37, 88 68 9, 10 19 2 27 35 76 44 74, 75
Montenegro S Montenegro SM Monti L Moreno D Moreyra E Morosano M Mosconi N Muller Muller Muller S Musso G	54, 55, 94 23 3 73 61, 62 57 54, 55, 94 3, 4 61, 62 1 92 85	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B Rizzotto M Rodríguez A Rodríguez GR Rodríguez Armesto Romano G Romera BM Ronco MT Rosa R Rossa A	$\begin{array}{c} & & & & & \\ & & & & & \\ & & & & & \\ &$	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H Vilalta E Villar S Villarruel R Vitelleschi MS Vogué C W Wagner M	30 9 37, 88 68 9, 10 19 2 27 35 76 44 74, 75 64
Montenegro S Montenegro SM Monti L Moreno D Moreyra E Morosano M Mosconi N Muller Muller Muller S Musso G N Naves A	54, 55, 94 23 3 73 61, 62 57 54, 55, 94 3, 4 61, 62 1 92 85	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B Rizzotto M Rodríguez A Rodríguez GR Rodríguez Armesto Romano G Romera BM Ronco MT Rosa R Rossa A Rosso O	30 6, 8 66, 67 14 84, 88, 92 61, 62 53 26 R 52, 53, 54, 55, 94 13 64 53 7 88	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H Vilalta E Villar S Villarruel R Vitelleschi MS Vogué C W Wagner M	30 9 37, 88 68 9, 10 19 2 27 35 76 44 74, 75 64
Montenegro S Montenegro SM Monti L Moreno D Moreyra E Morosano M Mosconi N Muller Muller S Musso G N Naves A Nestares G	54, 55, 94 23 3 73 61, 62 57 54, 55, 94 3, 4 61, 62 1 92 85 82 19	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B Rizzotto M Rodríguez A Rodríguez GR Rodríguez Armesto Romano G Romera BM Ronco MT Rosa R Rossa A Rosso O Rozados VR	30 6, 8 66, 67 14 84, 88, 92 61, 62 53 26 R 52, 53, 54, 55, 94 13 64 53 7 88 6, 7, 8, 17	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H Vilalta E Villar S Villarruel R Vitelleschi MS Vogué C W Wagner M	30 9 37, 88 68 9, 10 19 2 27 35 76 44 74, 75 64
Montenegro S Montenegro SM Monti L Moreno D Moreyra E Morosano M Mosconi N Muller Muller S Musso G N Naves A Nestares G	54, 55, 94 23 3 73 61, 62 57 54, 55, 94 3, 4 61, 62 1 92 85 82 19	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B Rizzotto M Rodríguez A Rodríguez GR Rodríguez Armesto Romano G Romera BM Ronco MT Rosa R Rossa A Rosso O Rozados VR Rucci A	30 6, 8 66, 67 14 84, 88, 92 61, 62 53 26 R 52, 53, 54, 55, 94 13 64 53 7 88 6, 7, 8, 17 38	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H Vilalta E Villar S Villarruel R Vitelleschi MS Vogué C W Wagner M Y Yeannes MI	30 9 37, 88 68 9, 10 19 2 27 35 76 44 74, 75 64
Montenegro S Montenegro SM Monti L Moreno D Moreyra E Morosano M Mosconi N Muller Muller S Musso G N Naves A Nestares G O	54, 55, 94 23 3 73 61, 62 57 54, 55, 94 3, 4 61, 62 1 92 85 82 19	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B Rizzotto M Rodríguez A Rodríguez GR Rodríguez Armesto Romano G Romera BM Ronco MT Rosa R Rossa A Rosso O Rozados VR Rucci A Ruffino V	30 6, 8 66, 67 14 84, 88, 92 61, 62 53 26 R 52, 53, 54, 55, 94 13 64 53 7 88 6, 7, 8, 17 38 83	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H Vilalta E Villar S Villarruel R Vitelleschi MS Vogué C W Wagner M Y Yeannes MI	30 9 37, 88 68 9, 10 19 2 27 35 76 44 74, 75 64 49
Montenegro S Montenegro SM Monti L Moreno D Moreyra E Morosano M Mosconi N Muller Muller S Musso G N Naves A Nestares G O Oakley L	54, 55, 94 23 3 73 61, 62 57 54, 55, 94 3, 4 61, 62 1 92 85 82 19 63	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B Rizzotto M Rodríguez A Rodríguez GR Rodríguez Armesto Romano G Romera BM Ronco MT Rosa R Rossa A Rosso O Rozados VR Rucci A Ruffino V	30 6, 8 66, 67 14 84, 88, 92 61, 62 53 26 R 52, 53, 54, 55, 94 13 64 53 7 88 6, 7, 8, 17 38 83	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H Vilalta E Villar S Villarruel R Vitelleschi MS Vogué C W Wagner M Y Yeannes MI Z	30 9 37, 88 68 9, 10 19 2 27 35 76 44 74, 75 64 49
Montenegro S Montenegro SM Monti L Moreno D Moreyra E Morosano M Mosconi N Muller Muller S Musso G N Naves A Nestares G O Oakley L Oakley LJ	54, 55, 94 23 3 73 61, 62 57 54, 55, 94 3, 4 61, 62 1 92 85 82 19 63 22, 23, 24, 25	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B Rizzotto M Rodríguez A Rodríguez GR Rodríguez Armesto Romano G Romera BM Ronco MT Rosa R Rossa A Rossa A Rosso O Rozados VR Rucci A Ruffino V	30 6, 8 66, 67 14 84, 88, 92 61, 62 53 26 R 52 52, 53, 54, 55, 94 13 64 53 7 88 6, 7, 8, 17 38 83	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H Vilalta E Villar S Villarruel R Vitelleschi MS Vogué C W W Wagner M Y Yeannes MI Z Zacarías F	30 9 37, 88 68 9, 10 19 2 27 35 76 44 74, 75 64 49 49
Montenegro S Montenegro SM Monti L Moreno D Moreyra E Morosano M Mosconi N Muller Muller S Musso G N Naves A Nestares G O Oakley L Oakley LJ Obelli JJ	54, 55, 94 23 3 73 61, 62 57 54, 55, 94 3, 4 61, 62 1 92 85 82 19 63 22, 23, 24, 25 51	Renzi J Rico MJ Rigalli A Riganti JG Riquelme B Rizzotto M Rodríguez A Rodríguez GR Rodríguez Armesto Romano G Romera BM Ronco MT Rosa R Rossa A Rossa A Rosso O Rozados VR Rucci A Ruffino V S Sain J	30 6, 8 66, 67 14 84, 88, 92 61, 62 53 26 R 52 52, 53, 54, 55, 94 13 64 53 7 88 6, 7, 8, 17 38 83 46, 47	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H Vilalta E Villar S Villarruel R Vitelleschi MS Vogué C W Wagner M Y Yeannes MI Z Zacarías F Zacarías F Zacarías Fluck MF	30 9 37, 88 68 9, 10 19 2 27 35 76 44 74, 75 64 49 49
Montenegro S Montenegro SM Monti L Moreno D Moreyra E Morosano M Mosconi N Muller Muller S Musso G N Naves A Nestares G O Oakley L Oakley LJ Obelli JJ Olguin MC	54, 55, 94 23 3 73 61, 62 57 54, 55, 94 3, 4 61, 62 1 92 85 82 19 63 22, 23, 24, 25 51 43	Renzi J Rico MJ Rigalli A Rigalli A Riganti JG Riquelme B Rizzotto M Rodríguez A Rodríguez GR Rodríguez Armesto Romano G Romera BM Ronco MT Rosa R Rossa A Rossa A Rosso O Rozados VR Rucci A Ruffino V S Sain J Sala L	30 6, 8 66, 67 14 84, 88, 92 61, 62 53 26 R 52 52, 53, 54, 55, 94 13 64 53 7 88 6, 7, 8, 17 38 83 46, 47 58	Valdes M Valenti J Valverde J Van Isseldyk F Vasconi MD Vega T Vila H Vilalta E Villar S Villarruel R Vitelleschi MS Vogué C W W Wagner M Y Yeannes MI Z Zacarías F Zacarías F Zacarías Fluck MF Zimermann R	30 9 37, 88 68 9, 10 19 2 27 35 76 44 74, 75 64 49 49 46 6, 8 53