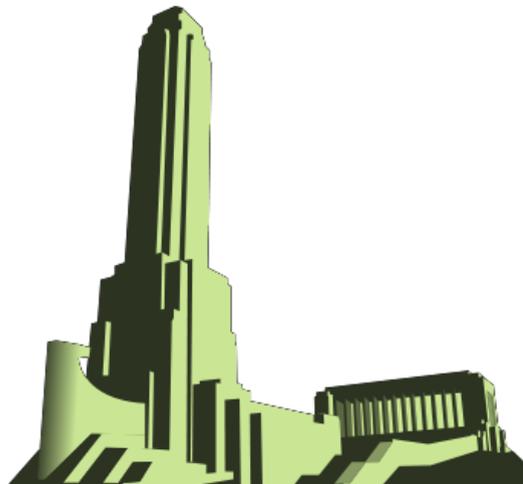




**Sociedad de Biología de Rosario**



**Resúmenes del  
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## A1

### YIELD AND QUALITY IN EARLY AND DELAYED CORN

Cruciani M, Papucci S, González A, González M, Incremona M.

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The objective in this experiment was to study the effect of different sowing moments and fertilization levels on the yield and quality of corn. The trial was conducted in Zavalla, Santa Fe, Argentina, at 60° 53' W, 33° 01' S in 2015. The hybrid sowed was ACA 470 VT3 Pro the 03/09/15 (early sow: ES) and 11/12/15 (delayed sow: DS and on second sow: SS). The treatments consisted in three rates of nitrogen (1: N<sub>0</sub> kg.ha<sup>-1</sup>; 2: N<sub>75</sub> kg.ha<sup>-1</sup> and 3: N<sub>150</sub> kg.ha<sup>-1</sup>). The experiment was set out as a randomized complete block design with 3 replicates per sowing moment. The grain yield (YIELD), weight of 1000 grain (WG1000), grain weight by hectoliters (WH) and prolificity (PROL) were measured. YIELD and WG1000 results were tested with ANOVA and the means were compared using Duncan's test. Both WH and PROL results were analyzed with Friedman's test. In ES, DS and SS, YIELD and P1000 showed increases when nitrogen was supplied. In late maize the differences were not significant for any of the variables (possibly due to the higher N availability). In SS, YIELD and WG1000 values were inferior to those corresponding to the other dates of planting in all the doses of N (unfavorable environment). As regard as WG1000, ES showed significant differences between all doses whereas in SS only the treatment with N150 kg.ha<sup>-1</sup> differed. Both PH and PROL did not present significant variations in any of the dates of sowing. ES and DS can be considered as two viable production alternatives. The advantage of reckon on the water resource and using smaller doses of N to reach its potential as an environment should be added to the DS, with radiation being the limiting factor and determining the ceiling for production

## A2

### MODELS TO DESCRIBE THE ACCUMULATION OF DRY MATTER IN MAIZE GRAIN

(*Zea mays* L.)

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Repeated measures on each experimental unit and non-linear models can be used to describe the kernel's growth process. Mixed non-linear models constitute a powerful tool to analyze the dynamics of growth. The segmented model combines multiple linear models to describe a nonlinear process. They do not contemplate any correlation between observations. To describe this process, nonlinear functions can be used, which allow to model the data with more flexibility and its parameters to have practical interpretation (maximum growth, growth rate, etc.). In this work both modeling approaches are presented to describe the evolution of the dry weight of the grain. The used data come from a trial to evaluate two maize hybrids, which differ in grain texture and yield potential. The dry weight of the grains was measured in 14 moments, from 10 days after the silking until the weight between two measurements was constant. As the data show a different behavior from a point, the evolution of the response is modeled with a bilinear function using a segmented model (M1), with positive slopes up to the cut-off point ( $c = 658.2 \text{ } ^\circ\text{Cday}^{-1}$ ) that are statistically different ( $p < 0.0001$ ) and then a constant slope segment. In the residue analysis it is observed that the assumption of constant variance and uncorrelated errors, required by these models, is not fulfilled. The other model (M2) is a mixed nonlinear model that uses the Gompertz function, showing that the growth rate is the same for the two hybrids while the maximum grain weight reached is different ( $p = 0.0204$ ). Of the two models used the M2 is the one that best describes the process of accumulation of biomass. The choice is made using the AIC information criterion (M1=2604.4 M2=2536.4), the biological interpretation of the parameters and the possibility of using a power function of the adjusted values to correct the heteroscedastic errors in the M2

## A3

### EVALUATION OF THE INCIDENCE AND SEVERITY OF *Fusarium graminearum* IN MAIZE (*Zea mays*) RESULTING FROM PRODUCTION FIELDS, SUBJECTED TO DIFFERENT PRODUCTIVE ENVIRONMENTS AND PLANTING DATE

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The aim of this research was to analyze *F. graminearum* incidence and severity in maize from fields subjected to different planting date (PD) and productive environments (PE). Samples come from fields which belong to small-scale pork producers from the South of Santa Fe (33°23'S, 61°11'W). In 2013/14 (PD: from 03/12 to 04/01), 2014/15 (PD: from 20/09 to 15/12) and 2015/16 growing seasons (PD: from 21/09 to 15/12), after physiological ripeness, four samples of 25 ears were collected from each field. They were evaluated for their incidence (INC) and severity (SEV) of *F. graminearum* according to Reid index et al. (1999). To evaluate if there were differences between INC and SEV of *F. graminearum*, Kruskal-Wallis one-way analysis of variance was used. Significant differences were observed for INC in the evaluated fields. The means of INC in PDxEP interactions varied between 1% and 82% (PE1: 19% to 82%; PE2: 17% to 21%; PE3: 1% to 33%). The PD: 12/11 and 03/12 (PE1) differed from the rest, and PD: 15/12 differed from 21-27/09 (PE3). Similar results occurred in SEV. The median SEV differed from 1-3 (PE1) and 0 (PE2 and PE3). In PE1 the highest SEV values (3 and 2) corresponded to the PD of 12/11 and 03/12. In AP3 the 15/12 PD differed significantly from the early PD. The use of PD which expose the maize to different types of stress condition (environmental and/or biotic), increase the agronomic exposure risk to this pathogen. It is necessary to keep adjusting this and others agronomic practices (density plant, N fertilization) in order to reduce these risks.

#### A4

### ASSIMILATES PRODUCTION AND PARTITIONING DURING SEED FILLING IN LANCEOLATE AND OVATE SOYBEAN ISOLINES

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In previous works, we informed higher seed number per square meter (SN) in lanceolate (L) than in ovate (O) soybean isolines, and that this increase in SN was directly associated to yield. Sustaining the growth of more seeds and yield increase would require higher assimilate production and partitioning to the seeds during the seed filling period (SF). To investigate this hypothesis, a field experiment was performed during the 2015/16 growing season, using a pair of L (FV35-L) and O (FV35-O) isolines. Yield was 11% higher for L than for O isolines (5870 and 5194 Kg/Ha, for L and O, respectively), and this was directly associated to the increase in SN (3792 and 3068, for L and O, respectively;  $P < 0.10$ ). No differences between L and O were observed for intercepted solar radiation and SF. Yet, seed growth rate (SGR) was higher in L than in O (31.1 and 18.0 g.m<sup>-2</sup>.day<sup>-1</sup>, for L and O, respectively;  $P < 0.05$ ). This increase in SGR was directly associated to SN. Moreover, L had higher photosynthesis rates than O (14.7 and 10.1  $\mu\text{moles.m}^{-2}.\text{seg}^{-1}$ , for L and O, respectively;  $P < 0.01$ ) and radiation use efficiency (1.7 and 1.1 g.MJ<sup>-1</sup>, for L and O, respectively;  $P < 0.0001$ ) during SF. These results confirm that assimilate production and partitioning to the seeds can be increased as sink demand increase, and support the hypothesis that sink demand modulate source activity during SF.

#### A5

### INCIDENCE OF *Fusarium* spp., *Penicillium* spp. AND PRODUCTION OF TOXINS IN CORN FROM A CROP WITH NITRO-SULFURED FERTILIZATION

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The object of the work was to determine the incidence of *Fusarium* spp., *Penicillium* spp. and the toxins production on two cultivars of corn with different levels of nitro-sulphured fertilization. The cultivars used were ACA 2000 and ACA 417 RR2. The statistical was in BCAD with 3 repetitions/hybrids/ treatment. The treatments consisted of a factorial arrangement: 3 cycles of culture x 2 hybrid x 2 levels of N (N0: control and N150: 150 kg N ha<sup>-1</sup>) x 2 levels of S (S0: 0 kg S ha<sup>-1</sup> and S40: 40 kg S ha<sup>-1</sup>) x 3 rep, To determine the fungic incidence the grains were incubated on 2 % APG (ISTA, 2009). Were performed ANOVA and Duncan test. Fumonisin were evaluated with the kit Celer Fumo® (Tecna SRL, It.) and DON y Zearalenone with kit Ridascreen Fast AG®, Germany). The absorbance was measure with ELISA reader Metrolab 959 (Metrolab, Ar.). Exist interacción between cycles/hybrids/treatment for *Penicillium* and *Fusarium*. The highest values of *Penicillium* incidence were in 2012/13. ACA 417 N0 S40 were significant differences ( $p < 0,05$ ) with an incidence of 84,6 % in 2012/13. The control (N0S0) of the same cultivar in 2014/15 was the lowest value (29,5 %). In 2014/15 *Fusarium* present the highest incidence (33 and 55%) on both hybrids and all the treatments ( $P \leq 0,05$ ). The lowest incidence for *Fusarium* were in 2012/13 (14 and 27 %). The quantity of *Fumonisin*, *Zearalenone* y *DON* varied with the year. The fertilization there are no the unique condicionant factor for the pathogens incidence.

#### A6

### SURVEY OF ALGAE IN A CREEK IN THE SOUTHERN AREA OF SANTA FE PROVINCE, ARGENTINA

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Though freshwater represents 0.01% of the world's water availability; however approximately 100,000 species, including algae, thrive in different freshwater habitats. Therefore, they constitute a valuable natural resource and as algae communities' composition and distribution of the result of environmental conditions, their presence can be used as an environmental quality indicator. The south of Santa Fe province includes seven creeks (San Lorenzo, Ludueña, Saladillo, Frías, Seco, Pavón and Del Medio) that present different degrees of rural, urban and industrial disturbance. Our objective was to survey algae populations in one of these creeks Ludueña as an initial stage in determining water pollution. Water samples were taken at Funes (32°57'04"S, 60°47'23"W) on April 2014, upstream (S1) and downstream (S2) of a dam. Filamentous algae were collected manually, while 500 mL of the phytoplankton was collected with a plankton net (25  $\mu\text{m}$  mesh size;  $n=5$ ). Taxonomic analyses were performed using a Leica DM500 optical microscope. Cosmopolitan algae, present in several creeks in Latin America, were detected. The genera *Aphanizomenon* (cyanophyta, nitrogen fixing and associated to high concentrations of pollutants such as phosphates), *Stauroneis* and *Cylindrocapsa* (Basilariophyta, class Basilariophyceae, Chlorophyta, Chlorophyceae) were observed in S1, whereas the genera *Mastogloia*, *Gyrosigma* and *Diploneis* (Basilariophyta, class Basilariophyceae) were observed in S2. This is the first report of a community of algae that could be used as indicator of contamination in freshwaters of southern Santa Fe province.

#### A7

### FIRST REPORT OF LICHEN COLONIZATION ON PERIMETRAL RODS EXPOSED TO DIFFERENT AGRO-ENVIRONMENTS OF SOUTHSANTA FE, ARGENTINA

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Presence of lichens associated to plants and other substrates can be useful in air pollution diagnosis. Our objective was to determine abundance and distribution of lichen communities associated to wooden rods that surrounded three different environments (sites) in the south of Santa Fe (Argentina): a field free of farm activity during 34 years, and two plots exposed to activity and impact from pork or agricultural production, respectively. Fifteen wooden posts (four sides exposed to cardinal points) were evaluated for each site. Area colonized by lichens (ACL) was registered using a standard grid (3x14 cells, 2cm<sup>2</sup> each) on each side and post. ACL was calculated as cm<sup>2</sup> of area colonized by lichens / total area (84 cm<sup>2</sup>) of standard grid and expressed as % values. Results were analysed by a factorial ANOVA considering the effects of: 3 sites x 4

sides x 15 posts (replications). Sites ( $F=20.93$ ;  $p<0.0001$ ), sides of the posts ( $F=27.94$ ;  $p<0.0001$ ) and their interactions (sites x sides) ( $F=4.88$ ;  $p<0.0001$ ) were significant. Lichen colonization was higher on the sides exposed to south and west due to the direction of predominant winds. Thus, south sides of the wooden posts that surround the plot under pig production registered the highest ACL= 43.9%; south side of posts from agricultural plot registered ACL= 32.4%; posts from the abandoned field showed high ACL values on sides exposed to the west, south and east: 41.4; 38.3 y 32%, respectively. This is the first report on use of lichens as bio-indicators of air pollution in environments exposed to different agro-ecological systems.

## A8

### SOYBEAN SEEDS GERMINATION: ROL OF EXPANSINS

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Seeds germination requires adequate conditions of water, temperature, oxygen and it is described by a dynamic of water uptake in three successive phases. The first one is the imbibition, which is produced by the difference of the potential water between seeds and its environment, the bigger is that difference the deeper is the imbibition. On the second phase, the seed experiences the most important metabolic changes that will define its germination without considerable water uptake. The last phase initiates with the radicle protrusion throughout the seed coat (*sensu stricto* germination) and implies and increased water uptake. These phases are completed, respectively, at 2, 8 and 12 hours of incubation by soybean (*Glycine max* L Merr.) isolate embryonic axes. The expansive process by water uptake produces the cell wall enlargement on the "elongation zone" of the embryonic axis, thus inducing germination. Expansin proteins (EXP) are responsible for promoting this cell wall loosening during germination. The aim of this work was evaluate the expression by RTqPCR of an identified putative EXP on cells from the soybean elongation zone at 0 (control) and after 1-8, 10 and 12 hours of incubation on distilled water at  $27\pm 1^\circ\text{C}$  and in the dark. The EXP level respect to the control increased for each successive incubation time, reached a significant maximum at 8 h and decreased not significantly at 10 and 12 h. As a conclusion, a critical EXP level would be necessary to initiate cell wall loosening on the elongation zone, thus overcoming the second phase of water uptake and establishing germination of soybean embryonic axis.

## A9

### EFFECT OF GLUTAMINE ON THE ALTERATIONS IN $\text{Na}^+$ , $\text{K}^+$ ATPASE INDUCED BY NEPHROTOXIC DOSES OF ACETAMINOPHEN (APAP) IN RATS

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We have previously reported the development of acute renal failure after the administration of a toxic dose of APAP. A nephrotoxic dose of APAP promoted an increased level of  $\text{Na}^+$ ,  $\text{K}^+$  ATPase and its detachment from its membrane anchoring as well as functional and histological alterations. HSP70 has been associated with cytoprotection in response to several injuries. Our aim was to study the effects of glutamine (GLN) on the alterations in  $\text{Na}^+$ ,  $\text{K}^+$  ATPase promoted by APAP. Male Wistar rats were used. Treatments: Administration of i) single dose of APAP 0.75 g/kg b.w. i.p (APAP, n=6). ii) single dose of GLN 0.75 mg/kg b.w. i.p (GLN, n=4) iii) GLN 30 min before APAP (GLN/APAP n=5). iv) vehicles (Control, n=5). At the end of a 16 h-collection period, animals were anesthetized and blood and kidneys were collected. Renal function, HSP70 cortical abundance and  $\text{Na}^+$ ,  $\text{K}^+$  ATPase cortical abundance and distribution were analysed. GLN protected against the renal functional effects of APAP. HSP70 expression was induced in APAP, GLN and GLN/APAP groups.  $\text{Na}^+$ ,  $\text{K}^+$  ATPase abundance was increased to the same level in the APAP and the GLN/APAP group, but  $\text{Na}^+$ ,  $\text{K}^+$  ATPase maintained its basolateral localization in rats receiving the dual treatment. Our results support the idea that GLN pretreatment could be a therapeutic option to prevent APAP-induced renal injury

## A10

### RENAL FUNCTIONAL REEVALUATION OF SPONTANEOUSLY DIABETIC RATS (eSS) AND ITS CORRELATION WITH ANIMAL AGE

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Rats IIM/FmeSS (eSS-e Stilmann Salgado), produced in the Faculty of Medicine (FM), Central Bioterio, UNR, have been characterized by investigator from (FM) who determined that eSS rats develop Diabetes Mellitus type II (DMTII) spontaneously and symptoms worsen with the age and in males. The objective was to establish if after several generations of the line, it conserved urea (U) and creatinine (Cr) levels and renal morphological alterations that had been described, to continue using eSS rats as a model of DMTII. Three eSS male rats were taken randomly and were subjected to euthanasia every 30 days (d) (100-340 d). Kidneys were extracted and fixed in 10 % formaldehyde and histological processed. Slides were stained with H&E, PAS and Direct Red 80/picric acid. Blood was obtained through cardiac puncture to determinate U and Cr. Urine was also analyzed. U and Cr values were statistically studied. U: 340d > 210d ( $p=0.02$ ); Cr: 340d > 150d ( $p=0.03$ ). Urines: no glucosuria was found. Proteins and hematies were present. Kidney morphological alterations began at 120 d and progress through rat ages. The main damages were: leukocyte infiltrate, positive PAS content in collecting tubes, increased intraglomerular mesangium, thickening of the basal membrane of the tubules and renal corpuscle, and fibrosis. We concluded that eSS rats did not increase significantly U and Cr values until 340 d. Morphological damages worsen with animal age and that eSS rats could be considered a good model to study DMTII.

## A11

### **AUTOMATISED RECOGNITION OF MORPHOLOGICAL ALTERATIONS IN HUMAN ERYTHROCYTES BY GLASS EFFECT THROUGH DIGITAL IMAGE ANALYSIS**

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RBC crenation (glass effect) appears due to the adhesion of RBC membrane to the glass and consists in fine and numerous spicules uniformly distributed on its surface (echinocyte shape). An automatized recognition program was developed using digital image analysis to quantify the percentage of echinocytes. Small concentrations (0.03-0.05%) of fresh RBCs from healthy donors, were suspended in media consisting of physiological solution (PS) and human serum albumin (HSA) at different concentrations (0.002 to 20% v/v). 20µL of each suspension was placed on glass slides and put in an inverted microscope (Union Optical, Japan, 40x lens, Canon Power Shot A640 camera with 1.4 digital zoom), digital photographs of each sample were taken every 3 minutes. Then, the percentage of crenation was calculated in each image by counting cells through the specifically designed image analysis program. Furthermore, form descriptors (area, perimeter, circularity, aspect ratio, roundness, solidity, Ferret Diameter and Minimum Ferret Diameter) were determined in order to identify the morphological states of RBC through a main component analysis followed by a clustering algorithm. From the results, the solution HSA-PS 1% v/v produces a crenation percentage minor to 20%, which accords to that obtained manually. It is possible to conclude that the technique developed is efficient to recognize echinocyte shapes and that, due to its speed and lack of observer subjectivity, it is useful in biomedical research when RBC suspensions are observed through a microscope. In addition, this technique can be extended in the future to the recognition of different erythrocyte morphologies for its application in Hematology.

## A12

### **EFFECT OF SEMINAL TRANSFERRIN LEVELS ON SPERM MEMBRANE AND DNA INTEGRITY**

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Our objective was to determine if testicular transferrin (TfT) levels are related to the fertilizing capacity of spermatozoa. Sperm membrane integrity (SMI) was evaluated by the Hypoosmotic Test (HOST). DNA integrity was assessed by the Acridine Orange Test (AOT) and concentration of TfT levels was measured using the Radial Immunodiffusion method adapted for low concentration developed by our workteam. Twenty semen samples were studied and Pearson Correlation Coefficient was applied. The relationships were as follows: 1) a high degree of negative association ( $r = -0.91$ ) was observed between the TfT concentration (mg / dL) and the SMI alterations ( $p < 0.05$ ). 2) no significant association was observed ( $r = 0.03$ ) between TfT concentration (mg / dL) and DNA integrity ( $p = 0.48$ ). We conclude that low levels of TfT are associated with the loss of SMI, affecting its functionality while it is not related with the integrity of DNA. At present, there is no consensus about techniques that should be used for measuring sperm DNA, tests that evaluate DNA sperm quality should not only identify the ability of sperm to reach the oocyte but also their ability to fertilize it. It is necessary to analyze a greater number of samples, add other techniques for the study of DNA and to evaluate other functional parameters of semen and its relation with TfT levels.

## A13

### **INCORRECT USE OF CLASSIC BIFACTORIAL ANOVA: A NON-PARAMETRIC ALTERNATIVE TO OBTAIN RELIABLE CONCLUSIONS**

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Factorial experiments are widely used because of their efficiency to detect the effects of factors and their interactions. The statistical approach used in this context is Analysis of Variance (ANOVA). This classic method requires the fulfillment of some assumptions (normality, homoscedasticity and independence) which frequently are not met in practice. In these situations, nonparametric and robust methods are alternative approaches. The objective of this work is to show that a parametric approach can lead to erroneous conclusions when the assumptions are not fulfilled. We studied the effect of two factors: Support (soybean, paper) and Time (0,24,48 h) on fungal culture extract pH in cellulase production, under a completely randomized design. Classic ANOVA and Aligned Rank Transformation (ART, a non-parametric method) were performed on a simulated dataset. Normality was not verified (Anderson-Darling test,  $p < 0.005$ ), but treatment variances were homogeneous (Levene's test,  $p = 0.114$ ). The interaction plot showed the existence of interaction between the factors. However, classic ANOVA did not detect it ( $p = 0.108$ ), whereas ART did ( $p = 0.044$ ). Hence, the use of parametric techniques when some of the assumptions are not met can lead to erroneous conclusions. We emphasize the importance of checking assumptions prior to the usage of the classic ANOVA and we recommend using non-parametric methods in cases of non-compliance.

## A14

### **EVALUATION OF THE OCCUPATIONAL EXPOSURE TO SOLVENTS ON THE REPRODUCTIVE CAPACITY OF INFERTILE MEN**

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The solvents used in the manufacture of synthetic paints, the textile industry and in the treatment of wood are endocrine disruptors. Our objective was to study the effect of the occupational exposure to solvents on sperm parameters related with the reproductive capacity of spermatozoid, in seminal samples from infertile men. In the study 90 semen samples of men that attended the Reproduction Service URHMA (Rosario) were incorporated. Smoking men, drug users, exposed to agrochemicals, sources of high temperatures and with pathologies that alter seminal quality were excluded. The sperm study and functional tests (WHO 2010) have been carried out to all samples by subjective method.

Two groups were formed: G<sub>E</sub> (n= 42) men occupationally exposed to solvents and G<sub>NE</sub>(n= 48) men not exposed. The percentage of progressive mobile spermatozooids (PM) and the sperm concentration (C) were determined with optical microscope using Makler camera and platinum thermostated at 37°C. The sperm morphology (M) was assessed with Hematoxylin. The functionality of the sperm membrane was studied with the hiposmotic test (HT), the integrity of nuclear DNA with acridine orange (AN) and the condensation state of chromatin with blue aniline (AA). The t-student test was applied to compare the averages of the analyzed variables. Significant statistically difference between both groups was found to evaluate the sperm functionally: HT, AN, AA and sperm concentration (p< 0.010). The other analyzed variables did not show differences (p> 0.05). It is considered relevant to evaluate the occupational exposure of man and its effects on male infertility.

## A15

### MOLECULAR *RHD* POLYMORPHISM IN D NEGATIVE C/E POSITIVE PHENOTYPES

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*RHD* null alleles express a D negative phenotype on the red blood cells (RBCs) membrane and *DEL* alleles a very low amount of the RhD protein that can only be detected with adsorption elution techniques. The aim of this work was to analyze the molecular basis of 780 D negative C/E positive samples. The presence of *RHD* allele was investigated by a PCR multiplex strategy. Samples carrying *RHD* alleles (D-/RHD+) were studied by exon scanning, microarray and sequencing. We found 117 D-/RHD+ samples. Molecular studies detected the *RHD* null alleles: *RHD-CE(3-9)-D* (n=15), *RHD-CE-D\** (n=58), *RHD-CE(4-8)-D* (n=1), *RHD-CE(4-9)-D* (n=1), *RHDψ* (n=1), *RHD(58IinsG)*(n=19), *RHD(100IT>A)* (n=1), *rG* (n=1) and *RHD-CE(4-7)-D<sub>2</sub>* (n=1) and the *DEL* variants *RHD(M295I)* (n=4), *RHD(46T>C)* (n=12) and *RHD(IVS3+1G>A)*(n=2). *DEL* alleles were detected in 2,31% and *RHD* null alleles in a 12,69% of the D-/RHD+ samples. The characterization of *DEL* alleles will prevent transfusion of this units to patients in risk of alloimmunization.

## A16

### MORPHOANATOMICAL CHARACTERIZATION OF *Euphorbia umbellata* (Pax) Bruynscv. *Rubra* (EUPHORBIACEAE)

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*Euphorbia* is a genus that has significant cytotoxic activity; the cultivar "*Rubra*" of the species *Euphorbia umbellata* (Pax) Bruyns is a small tree, exotic that is cultivated as ornamental by its leaves with several versions of color, from red-purple to homogeneous green with reddish spots. Its latex is related to some properties of ethnopharmacological relevance such as antitumor, anti-inflammatory, anti-allergic and gastric disorders. The aim of this work is to study morphological and anatomical characters of aerial organs of *Euphorbia umbellata* (Pax) Bruynscv. *Rubra* to contribute to the correct botanical identification of the species. Fresh material, fixed in FAA and embedded in paraffin was used to perform cross sections of leaf, petiole and stem with sliding microtome. The transparentations and macerated were carried out according to conventional techniques. The different sections were stained with safranin and cresyl violet. The results show that *Euphorbia umbellata* cv. *Rubra* is a shrub, monoic, latescent, glabrous with succulent stems, the leaves are alternate, simple, with obtuse apex, mucronate, cuneadas at the base and decurrent on the petiole. The anatomical characters are: lamina with homogeneous mesophyll, unistrata epidermis, amphistomatic, with stomatarubioceous type; simple eglandular trichomes, located on the margins near the base. The primary stem is circular in section, with broad cortical and spinal parenchyma with thin-walled cells. The secondary stem has an early development of the secondary xylem but little formation of suber. The laticifers are non-articulated and branched. The analyzed morphoanatomic characters contribute to improve the quality control of the vegetal drug.

## A17

### VASCULAR FLORA OF THE PROVINCE OF SANTA FE: MYRSINACEAE AND PRIMULACEAE

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The families Myrsinaceae and Primulaceae belong to the class Magnoliopsida (=Dycotiledoneae). In the old Engler's classification systems both were located in the Order Primulales. At present, based on morphological and molecular evidence the genera of Myrsinaceae are considered part of Primulaceae; the latter, according to the APG-IV system, is located in the Order Ericales, one of the most notable changes in the classification systems. This contribution is an introduction to the knowledge of these families and their distribution in the province of Santa Fe. The methods consist of a bibliographical review, consultation of national herbaria with important collections of the province (SF: Esperanza; SI: San Isidro; UNR: Zavalla), field work experience of the authors and lab work. Preliminary results show that the genus *Myrsine* L. is represented in Santa Fe by a single species: *Myrsine laetevirens* (Mez) Arechav., "canelón-morotí", "palo de San Antonio", a tree with persistent leaves with dark spots on both sides, common in the valley of the Parana river. The Primulaceae family is represented in the province by three genera with one species each: *Centunculus minimus* L., naturalized, annual grass common in wet meadows of San Jeronimo Dept.; *Anagallis arvensis* L., naturalized, cosmopolitan grass, sometimes invasive in crops, and *Samolus valerandi* L., native, distributed in almost the entire province. Taxonomic information, distribution map and illustrations are provided.

## A18

### EVALUATION OF THE RhD PROTEIN AND OF MEMBRANE-BOUND IgG PARTICIPATION IN THE CORD RED BLOOD CELLS SENESCENCE PROCESS

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While neonatal red blood cells (RBCs) survive in circulation between 60-80 days, adults RBCs survive approximately 120 days. Multiple alterations trigger the selective removal of the Senescent (Se) RBCs from the circulation. In adults, erythrocyte aging may lead to autologous IgG binding followed by recognition and phagocytosis. However, the senescence process in neonatal RBCs is not fully characterized. We evaluated the participation of the RhD protein in the erythrocyte aging and we analyzed membrane bound IgG in Se and Young (Y) RBCs populations obtained from cord blood samples using flow cytometry. For the RhD protein study, samples (n=18) were incubated with APC anti-RhD IgM antibodies. Median fluorescence intensity (MFI) was measured. To assess the membrane bound IgG, samples (n=15) were incubated with an Alexa 488 anti-human IgG antibody. The percentage of opsonized RBCs with IgG was measured in both populations. The mean of MFI in cord YRBCs was significantly higher when compared with SeRBCs (Y=878.9±231.33 vs Se=756.1±166.95, p<0.0001, paired samples Student's t-test). The mean percentage of opsonized RBCs with IgG was: Y=0.4±0.13 vs Se=5.1±3.01. A significant increase of membrane bound IgG was observed in neonate SeRBCs populations (p<0.0001, paired samples Student's t-test). These findings suggest that the RhD protein would be associated with the membrane changes that undergo SeRBCs. Microvesicles formed during senescence may contain the RhD protein explaining the loss of expression observed in SeRBCs. Also, our results reflect the participation of IgG in the selective removal of senescent erythrocytes from neonates.

## A19

### TREATMENT WITH *Ligaria cuneifolia* PROANTOCYANIDINE ENRICHED FRACTION PRODUCES PLASMA LIPIDS CONCENTRATIONS DECREASE AND INCREASE OF BILE SALTS BILIARY EXCRETION IN WISTAR RATS FED WITH A HIGH FAT DIET (HFD)

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Proanthocyanidin enriched fraction from *L. cuneifolia* was purified (*PL.cuneifolia*) to analyze the effect on Cho and triglycerides (TG) plasma levels, blood viscosity, and also biliary excretion. Adult male Wistar rats (n = 24), 70 days old, were fed for 28 days with a HFD (every 100g of standard diet 40% bovine meat juice). Rats were administered with either physiological solution (controls C, n=12) or *PL.cuneifolia* 3mg /100g body weight (treated T; n=12) i.p. each 24hr during 7 days. On day 8 they were anesthetized and the bile duct was catheterized for bile collection each 15 min for 60 minutes, at the end, the blood was obtained by cardiac puncture. In plasma Cho, ChoHDL, ChoLDL and TG were determined by enzymatic and colorimetric methods. Blood and plasma viscosity were determined with rotational viscometer. Relative blood viscosity standardized to 45% of hematocrit (BVrs) was calculated (Blood Viscosity / Plasma Viscosity)<sup>45 / Hto</sup>. Bile flow (BF) was determined by gravimetry, bile salts biliary excretion (EBS) was calculated (Concentration of bile salts in bile x BF). Student t test was applied for unpaired data. Results are expressed as mean±SD. Cho(mg%): C:145.3 ± 8.5, T:62.9 ± 3.2\*\*; ChoHDL: C:25.0±1.3, T:29.5±2.3(NS); ChoLDL: C:28.9±2.4, T:21.4±2.2\*; TG: C:360.1±27.5, T:71.4±11.0\*\*; BVrs: C:5.09±0.45, T:5.41±0.49(NS); BF(μl /min. g liver): C:2.61±0.14, T:3.44±0.21\*; EBS(nmol / min.g liver): C:34.15±3.66, T:54.50±6.50\* (\*p <0.05; \*\*p <0.001 and NS: not significant vs. C). The results showed a lipid-lowering effect of *PL.cuneifolia* in rats fed with a HFD. The decrease in plasma Cho might be due to the increase in the BS biliary excretion (main compound of hepatic metabolism of Cho).

## A20

### UTILITY OF CATALASE ENZYME AS AN INDICATOR OF ACTIVITY IN LUPUS NEPHROPATHY

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Lupus nephropathy (LN), occurring in 60% of patients with systemic lupus erythematosus (SLE), is a clinical complication of poor prognosis. Despite advances in the treatment of LN, 10-15% patients progress to end stage renal failure. Early diagnosis and treatment, however, can significantly improve long term prognosis. Previously, we found that oxidative stress (OS) is increased in patients with active LN (ALN); a pro-oxidant status prevails and antioxidant defenses would not be sufficient to mitigate it. Our aim was to analyze if any of the following OS parameters, tiobarbituric acid-reactive substances (TBARS), superoxide dismutase (SOD) and catalase (CAT), is a better marker of activity in LN. We studied patients with diagnosis of LN and, as a control group, patients with diagnosis of inactive LN (ILN) from Nephrology Service of Hospital Provincial del Centenario. We determined: a) TBARS levels, by a colorimetric technique (n=24 for both groups), b) SOD activity, with a commercial kit (Randox Ltd.) (n=17 for ALN; n=24 for ILN) and c) CAT activity, following H<sub>2</sub>O<sub>2</sub> decomposition at 240 nm (n=13 for ALN; n=11 for ILN). Results obtained for TBARS [pmol malonyldialdehyde/ml; median (range)], SOD (U/gHb; mean±SD) and CAT (U/gHb; mean±SD) in ALN vs ILN were respectively: 40 (0-172) vs 17 (0-420); 1114±710 vs 1069±541 y 1.14±0.75 vs 0.5±0.2. No significant differences between groups were found for TBARS (p=0.25) nor for SOD (p=0.81) but for CAT (p=0.01). We conclude that the activity of CAT would be useful to discriminate between patients with ALN and ILN. Then, determination of CAT, which simple and low cost, could be used as an additional predictor of activity in renal disease together with those parameters commonly used in clinical laboratory.

## A21

### DETECTION OF CIRCULATING TUMOR CELLS IN PATIENTS WITH BREAST CANCER BY ANALYZING THE TWIST-1 GENE

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The neoplastic cells that enter into blood are denominated circulating tumor cells (CTC) and its detection has an emerging role as biomarkers for the prognostic of patients with breast cancer. When increase TWIST1 expression in tumor cells, undergoes mesenchymal-epithelial transition facilitating the metastasis. The aim of this study was to detect CTC determining the TWIST1 expression in patients with breast cancer. It was correlated with the other prognostic factors in breast cancer. From peripheral blood samples of patients with breast cancer (n=34) and healthy donors (n=8) were isolated mononucleated cells and ARN was extracted. A retrotranscription and PCR real time were performed to detect TWIST1 gen and glyceraldehyde 3phosphate deshydrogenase gen as control. Analyzing by comparative method  $2^{-\Delta\Delta Ct}$  was found that 26% of patients showed the expression of TWIST1 increased from 2 to 25 times respect to healthy donors. At the 2 patients that show the greatest increases in the expression of TWIST1, it was detected distance and local metastasis, respectively. The analysis by Fisher test between the expression of TWIST-1 gene and the presence of estrogen and progesterone receptor, the HER2/neu overexpression, the presence of metastatic nodes, the tumor size ( $< 0 > 2$ ), the histologic and the nuclear grade (1-2 o 3) not yield significative association ( $p > 0,05$ ). The increase of TWIST1 expression indicated the presence of CTC in those patients, and would associate a greater risk of metastatic development. Therefore, the detection of the TWIST1 gene levels would be an independent parameter of other prognostic makers and could provide additional prognostic information of clinical relevance.

In healthy controls (n=8) and in the patients (n=34) was calculated  $\Delta Ct = Ct_{TWIST1} - Ct_{GADPH}$ . Expression levels of mRNA TWIST-1 in patients were analyzed, in relation to average expression in healthy donors by the comparing the level of expression metho d  $= 2^{-\Delta\Delta Ct}$ , considering 1 at the expression detected in controls. The controls had a  $\Delta Ct = 3.7 \pm 1.93$  that it used as reference value

## A22

### STUDY OF THE SECRETORY STATUS IN PATIENTS WITH UROGENITAL TUMORS BY SEROLOGIC AND MOLECULAR METHODS

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Anomalous glycosylation of blood group antigens has been studied for different tumors. The common phenomenon found was the partial or total loss of the expression of the blood group isoantigens and the relationship between the loss of expression and the potential malignancy of the tumor was established. The aim of this work was to investigate the Lewis system antigens and the secretor status in patients with urogenital tumors. 64 subjects (50% had prostate and bladder tumors and 50% was the control group) were examined.

Lewis phenotypes were investigated in peripheral blood by hemagglutination techniques. The secretor status was determined in saliva by the hemagglutination inhibition test and the FUT2 gene polymorphism (allele G428) was analyzed by ASO-PCR. The results showed a higher degree of malignancy of urogenital diseases in the non-secretor group OR=3.44. 51% of patients with cancerous lesions were non-secretor with Le (a + b-) phenotype. In the control population the secretor status predominated (78.2%). A marginal association between the secretor status and these lesions was observed. The patients with cancerous lesions showed the absence of the wild-type FUT2 allele. The alterations in ABH and Lewis antigens reflect changes in the level of differentiation of the tumor cell. Our findings indicate that the genetic variants analyzed could be considered as risk markers in patients with urogenital tumors.

## A23

### MOLECULAR ALTERATIONS OF CALRETICULIN IN BCR-ABL, JAK2 AND MPL NEGATIVE MYELOPROLIFERATIVE NEOPLASMS (MPN)

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We investigated the presence of mutations in exon 9 of the CALR gene in patients with ET (Essential Thrombocytemia) and PMF (Primary Myelofibrosis), JAK2V617F, MPLW515L/K negative. We studied genomic DNA from 44 patients diagnosed with ET (36) and PMF (8) according to WHO criteria. The methodology used was PCR amplification of exon 9 of the CALR gene from peripheral blood or bone marrow leukocytes, followed by automatic sequencing. Seven different mutations were detected in exon 9 of CALR in 63.6% of patients with ET and PMF JAK2 / MPL negative. The type 1 mutation (52 bp deletion) and the type 2 mutation (5 bp insertion) accounted for 42.9% and 39.3% of the positive cases respectively. Of the 5 remaining mutations, only one case of each was found. 75% of patients with PMF had mutations in CALR, being the most frequent type 2 mutation (83.3%), the type 1 mutation represented 16.7%. In the patients with ET, these mutations were present in 61.1% of the cases, the most frequent being mutation type 1 (50%). The mutation type 2 was found in 27.3%, whereas the 5 types of remaining mutations had very low frequency since they were detected in a single patient each. The majority of patients with negative JAK2 PMF and ET showed a mutation in the exon 9 of CALR, therefore the detection of these mutations is a very useful diagnostic tool to discriminate a clonal proliferation of a reactive process.

## A24

### HEREDITARY DISORDERS OF HEMOGLOBIN IN ROSARIO AND AREA OF INFLUENCE

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The aim of the study was to know the molecular basis of hemoglobinopathies in Rosario and zone of influence. We studied 313 patients with  $\beta$ Th, 84 with  $\alpha$ Th and 11 with an abnormal hemoglobin. Six  $\beta$ Th: CD39 and IVS I-110, I-6, I-1, II-1 and II-745 mutations were analyzed by ARMS-PCR. The  $-\alpha^{3.7}$ ,  $-\alpha^{4.2}$ ,  $-(20.5)$ ,  $--_{Med}$  and  $--_{SEA}$  were analyzed by GAP-PCR and  $\alpha^{Nco}$  and  $\alpha^{Hph}$  by PCR and restriction enzyme cleavage. In cases where the mutation was not identified and in those that presented an abnormal band, the band/or  $\alpha$ geneS were sequenced. Of the 313 cases of  $\beta$ Th were found to be CD39 carriers 46%, IVSI-110 26%, IVSI-1 9.9%, IVSII-745 6.7%, IVSI-6 5, 7% and IVSII-1 2.6%. By sequencing, IVSI-2 was found in 0.64%, CD6 (-A) and CD41/42 (-TTCT) both in 0.32%. In 0.64% the mutation could not be identified. Of the 84 cases with  $\alpha$ Th, were  $-\alpha^{3.7}$  69%,  $-(20.5)$  6%,  $--_{Med}$  4.7%,  $--_{SEA}$  4.6%,  $\alpha^{Hph}$  was 3.5%,  $\alpha^{Nco}$  2.5% and  $-\alpha^{4.2}$  1.2% and 8.5% homozygous for  $-\alpha^{3.7}$ . In the 11 cases of Hemoglobin variants were found: HbAgenogi, Hb Q India, Hb Constant Spring in homozygosity, Hb Lepore, Hb E, Hb Interlaken, Hb Sabine, Hb Quong Sze (a case of each one) and Hb S (3 cases). In the studied population, mainly of Italian origin, CD 39 and I-110 represented 73% of the  $\beta$ Ths. The frequency of these alleles differs from those in the northwest of the country and are similar to those found in Buenos Aires. In  $\alpha$ Th, the most frequent alteration was the deletion 3,7. Individuals with  $--_{SEA}$  were immigrants of Chinese origin. The knowledge of the mutations found in a specific population and the phenotype (especially in  $\alpha$ Th) allow the selection of the most appropriate strategy for molecular diagnosis.

## A25

### THE TEST OF *Allium cepa* L.: AN EFFECTIVE METHOD FOR THE DETECTION OF FITO, CITO AND GENOTOXICITY OF THE PROMUTAGEN 2-AMINOFLUORENE

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The common onion (*Allium cepa* L.) is considered a standard organism for rapid tests, since there is high correlation with tests in mammals and possesses an enzymatic system that allows the evaluation of promutagens. The promutagen 2-aminofluorene (2AF), aromatic hydrocarbon dye precursor and diagnostic mutagen in the Ames test with the addition of metabolic activation was studied. In case of detecting phyto, cyto and/or genotoxicity, we could infer that the plant is metabolizing 2AF and thus would be a valid indicator for this promutagen, opening the possibility to study other similar ones. This is advantageous with respect to the test in bacteria with metabolic activation because it is simple and much more cheaper. According to the ANOVA and Student's t-test, significantly different mean values were observed with respect to the negative control in all cases of root elongation except in the 2 mg/L 2AF solution. The mitotic index (MI) showed significant differences in the 50 and 25 mg/L solutions, and also chromosomal aberrations were detected, while the MI means of 5 and 2 mg/L 2AF solutions did not differ from the negative control. No mitotic figures were observed at the highest assayed concentrations of 2AF (75 and 100 mg/L). The obtained results suggest that the *Allium cepa* test was a good indicator for the detection of the phyto, cyto and genotoxic properties of the promutagen 2AF.

## A26

### FALSE NEAREST NEIGHBORS DETERMINATION FOR ERYTHROCYTE POPULATION CHARACTERIZATION IN THE PRESENCE OF *Ascaris lumbricoides*

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Red blood cells (RBC) are principal determinants in the rheological behavior of blood. *A. lumbricoides* is a nematode parasite of small intestine of man. In order to get a quantitative description of erythrocyte viscoelasticity and their disturbances in the presence of *A. lumbricoides* a non-linear mathematical tool is applied to photometric temporal series corresponding to RBC membrane fluctuations due to a stationary shear stress. Considering the biosignal as a set of points with delay coordinates in a dimensional space  $D_e$  (Embedding Dimension) then the whole dynamics of the process can be defined uniquely. And, when the chosen dimension  $D$  is  $D < D_e$  then some points appear closer than they really are in  $D_e$  (FNN, False Nearest Neighbors). The %FNN in  $D=3$  was used in order to characterize a population of blood samples (healthy donors) in the presence of larva extracts of *A. lumbricoides*. RBC were suspended in a high viscosity medium (pH 7.4; 295 mOsm/kg; 18 cP) and they were incubated with the parasite under study (30 and 60 min). A blind experiment was performed; 8 samples were labeled as m1-3, m5-7 and m9-10. It could be completely characterized a group 1 (m1 and m7) and a group 2 (m9-10). Also a weekly characterized group 3 (m2-3, m5-6) was determined. Group 1 and 2 were RBC incubated with the nematode for 30 and 60 min, respectively, and Group 3 was the control group. These results corroborate that contact with extracts of *A. lumbricoides* larvae induce disturbances in the RBC membranes that affect their viscoelasticity which could be evidenced by the non-linear parameters determined in this work.

## A27

### SYSTEMIC IRON HOMEOSTASIS: ROLE OF HEPcidIN IN IRON DEFICIENCY ANEMIA, AND ANEMIA OF CHRONIC DISEASE. A PRELIMINARY STUDY

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Hepcidin is considered the central iron homeostasis regulator hormone. It is potentially involved in iron distribution between different organs. Increases in iron storage, and chronic, inflammatory and neoplastic process stimulate *Hepcidin* production, while, hypoxia and anemia, increases erythropoiesis and suppressed *Hepcidin* production. The determination of *Hepcidin* levels could contribute to diagnose iron deficiency (ID) associated to chronic disease anemia (ACD). The aim of this study was to establish the value of Hep-25 bioactive peptide in two groups of patient: iron deficiency anemia (IDA), and anemia of chronic disease (ACD). We also compared these values with a control group. Iron status,

Erythropoietin and Hepcidin were determined in both groups. The mean value of Hepcidin in IDA was significantly lower than in controls ( $p < 0.0001$ ), but the mean value of Hepcidin in ACD was significantly higher than in controls ( $p < 0.0001$ ). We only find a lower than normal Hepcidin value, in one ACD patient with a noncologidisease. *Hepcidin levels* are influenced by hypoxia, erythropoiesis, inflammation and iron storage. Our data support the decrease in Hepcidin values in ID patients. One patient with ACD and concomitant ID had lower Hepcidin levels compared with those who had "pure" ACD. In this case Hepcidin value was effective to diagnose ID associated to ACD.

## A28

### INTERACTION BETWEEN VITAMIN D3 AND BETA-LACTOGLOBULIN MODIFIED BY HEAT TREATMENT

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Dairy protein beta-lactoglobulin (βLG) can bind hydrophobic vitamins, producing nano-complexes which may be suitable vehicles for adding vitamins to food. Moreover, βLG can protect vitamin from environmental factors such as light, temperature, humidity or oxygen, extending its lifetime. The aim of this work was studying the interaction between native βLG, as well as oligomeric βLG formed by heat treatment, with vitamin D3 (vD3) in the production of nano-complexes vitamin-protein. Aliquots vD3 in ethanol at different concentrations were added to solutions of βLG (without heat treatment and treated at 85 °C for different times). Protein conformational changes, either due to effect of heating or ethanol effect, were studied. Different techniques were assayed: turbidimetry, fluorescence quenching, circular dichroism and high performance liquid chromatography (HPLC). The decrease of both turbidity and intrinsic fluorescence confirmed vD3 is captured by βLG; the Stern-Volmer constants decreased at higher temperatures, showing a static quenching; high concentrations of vD3 led to a high relationship vD3:βLG, higher than the currently reported values; the heating treatment of the protein worked against the nano-complex formation, suggesting βLG denatured structures (dimers, trimers and oligomers) would not provide the best structure to capture vD3; ethanol did not modify neither the secondary nor the tertiary protein structure significantly. Further studies will lead to a better comprehension of the mechanisms involved in the interaction vD3-βLG, in order to improve the vitamin supplements in food.

## A29

### THE APPLICATION OF NMR AS A POSSIBLE METHOD OF ECONAZOLE QUANTIFICATION WHICH IS RELEASED FROM SEMISOLID SYSTEMS

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Nuclear Magnetic Resonance (NMR) is used in the elucidation of molecular structures, also for quantitative purposes and in kinetic and thermodynamic studies, so it could become applicable to determine the concentration of active principles released from pharmaceutical systems. We propose to study the employ of NMR, as non official technique, for the quantification of principle active that diffuses from a pharmaceutical semisolid systems to a receptor medium. We work with the NMR300MHz Avance Bruker equipment. We study an emulsion whose oil phase was constituted of stearic acid and liquid vaseline, while the aqueous phase was formulated with triethanolamine, water and econazole (ECZ) 5%. In cellulose nitrate membrane of 0,65µm, were place 40mg of sample with 2mg of ECZ each one. In the NMR tubes were placed the receptor medium formed with propylene glycol and deuterated water and the membranes loaded with the samples were introduced. Once the membrane contact the receptor medium, the concentration of ECZ diffused was quantified by RMN each hour for 4 h. We observed that the systems follows a Higuchi kinetics, with a  $r^2$  of 0,78. Data obtained from this same system performed by the official method and determining the amount of ECZ by HPLC also gave Higuchi kinetics. The method of NMR diffusion and quantification could be an alternative to the official method of using Franz cell to determine the diffusion and subsequent quantification of the active principle of these solid pharmaceutical systems.

## A30

### STUDY OF S100A9 EFFECT ON HUMAN GAMETE INTERACTION AND SPERM CAPACITATION

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Our previous results indicate the presence of S100A9 protein in human oviduct secretion. The aim of the study was to evaluate the effect of S100A9 on sperm-ZP interaction and on sperm protein tyrosine phosphorylation (PTP). Human motile sperm were obtained from normozoospermic donors. Human oocytes were donated from patients from a fertility clinic. To evaluate sperm-ZP interaction, 3 to 5 oocytes were placed in a medium droplet with BSA and 0, 0.1, 1.0 and 10.0µg/ml of human recombinant S100A9 (hrS100A9) and inseminated with 10<sup>5</sup> motile sperm/ml. After 4 h incubation, oocytes were placed in 0.5% Y Eosine drops to stain dead sperm. Mean number of live sperm attached to ZP was calculated. To assess the effect of S100A9 on PTP, motile sperm were incubated in capacitating conditions and in the presence of hrS100A9 (0, 0.1, 1.0 and 10.0µg/ml) during 6 h at 37°C, 5% pCO<sub>2</sub>. Afterwards, sperm viability and motility were evaluated and sperm proteins were extracted. Samples were analyzed in 7.5% SDS-PAGE and transferred to nitrocellulose membranes. Bands were detected with anti-phosphotyrosine and anti-IgG-peroxidase antibodies, and then revealed with chemiluminescence. %PTP was calculated in each treatment, considering control values as 100%. Statistical analysis was performed with ANOVA. The presence of S100A9 did not have any significant effect on number of sperm bound to ZP ( $p > 0.05$ , n=8). The higher dose of S100A9 caused a significant augment on %PTP compared to control ( $p < 0.05$ , n=5). In conclusion, S100A9 modulates sperm signaling pathways involved in sperm capacitation, suggesting that it could be involved in the regulation of the reproductive process.

### A31

#### ERYTHROCYTE AGGREGATION IN OVERWEIGHT AND OBESE ADOLESCENTS: STUDY OF ERYTHROCYTE DEFORMABILITY (ED) AS MEAN CELL FACTOR

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Overweight and obesity during childhood and adolescence is a growing problem all over the world. Furthermore, knowing that in most cases this condition persists in adulthood. On the other hand, it is known that obesity is a cardiovascular risk factor, and the study of erythrocyte aggregation (EA) and its principal cellular determinant, erythrocyte deformability (ED), have been studied in obese groups, reporting contradictory results. We studied males and females adolescents, with 11 to 19 ages, without known metabolic diseases. They were classified according to BMI in: normal weight (N) (n=19):  $\geq 10 \leq 85$ ; overweight (OW) (n=6):  $\geq 85 \leq 95$  and obese (OB) (n=9):  $\geq 95$ . EA was determined by optical method in suspensions of red blood cells in dextrán 500 2% in saline. We determined: T (estimating the size of the aggregates) and V (estimating the initial rate). ED was determined by rigidity index (RI) by filtration with micropore membrane (5  $\mu\text{m}$ ). Results were analyzed with ANOVA test and expressed as media  $\pm$  standard deviation. Significance:  $p < 0.05$ . Results: EA: T in N:  $1.77 \pm 0.11\text{ns}$ ; OW:  $1.75 \pm 0.07\text{ns}$ ; OB:  $1.76 \pm 0.08\text{ns}$ . V in N:  $0.74 \pm 0.35\text{ns}$ ; OW:  $0.66 \pm 0.12\text{ns}$ ; OB:  $0.83 \pm 0.51\text{ns}$ . RI in N:  $18.2 \pm 8.2\text{ns}$ , OW:  $13.5 \pm 4.8\text{ns}$ , and OB  $15.4 \pm 7.8\text{ns}$ . According to our results, there are no differences in T and V between the three groups. On the other hand, we didn't find differences in RI, which is consistent with the no difference found in T and V. Even though our results are coincidence with obtained by other authors, we consider necessary to continue the study.

### A32

#### EFFECTS OF THE ADDITION OF DRIED BLUEBERRIES POWDER TO THE DIET ON THE OBESITY OF IIMb / $\beta$ RATS

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The IIMb /  $\beta$  rat line develops moderate non-hyperphagic obesity, defined both by overweight and by the volume of adipose depots. The objective of this study was to assess the effects of a diet containing powdered dried blueberries on the obesity of obese  $\beta$  rats. Male 70-days-old  $\beta$  rats were fed during 90 days: AIN 93 (n:6) (American Institute of Nutrition 1993, prepared with sunflower oil); JB (n:6): AIN 93 modified by replacing sunflower oil with first bovine juice (rich in saturated fatty acids) or JBArA (n:6): JB + dried blueberries powder (as source of anthocyanins, 0.5g per 100g of food). At the end of the experiment animals were euthanized, abdominal fat pads (AFP) and livers (L) were excised and relative weights were calculated as percentage of body weight; Biomass (B) and feed intake (FI) were also evaluated. Results are expressed as: mean  $\pm$  SD. ANOVA test was applied for comparisons, significant differences ( $p < 0.05$ ). B (g):  $238.5 \pm 30.6$ ;  $237.8 \pm 32.6$ ;  $239.9 \pm 17.7$  (p: 0.56) - FI (kg):  $3.54 \pm 0.22$ ;  $3.68 \pm 0.19$ ;  $3.64 \pm 0.27$  (p: 0.43) - AFP (g%):  $4.04 \pm 0.36$ ;  $4.18 \pm 0.46$ ;  $4.42 \pm 0.51$  (p: 0.26) - L (g%):  $3.82 \pm 0.28$ ;  $4.47 \pm 0.65$ ;  $3.77 \pm 0.26$  (p: 0.04). Liver weight was lower in JBArA group due to smaller hepatic fat depot (histologically tested). Supplementation with blueberries might be useful in diminishing hepatic steatosis, a comorbidity frequently associated to obesity.

### A33

#### EFFECTS OF A DIET WITH POWDERED DRIED BLUEBERRIES ON THE GLYCOLIPID PROFILE OF OBESE IIMb / $\beta$ LINE RATS

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Overweight and obesity are among the main risk factors for diabetes and dyslipemia. Increasing fruits consumption has been recommended to relieve non-communicable chronic diseases burden; beneficial effects of fruits might be due to their content of anthocyanins. IIMb/ $\beta$  rats develop peripubertal obesity, hypertriglycerolemia and glucose intolerance progressing to type 2 diabetes in adulthood. The objective of this study was to assess the effects of a diet containing powdered dried blueberries on the glycolipid profile of  $\beta$  rats. Eighteen male 70-days-old rats were fed: AIN 93 (American Institute of Nutrition, with sunflower oil); JB: AIN 93 modified, with first bovine juice or JBArA: JB + dried blueberries powder (0.5g/100g of food). After 90 days, plasma glucose (GLU), triacylglycerols (TAG), total cholesterol (COL) and HDL and LDL fractions were measured. Results are expressed as: mean (mg/dL)  $\pm$  SD. ANOVA test was applied for comparisons, significant differences ( $p < 0.05$ ). GLU:  $209.8 \pm 26.8$ ;  $198.0 \pm 4.9$ ;  $242.4 \pm 54.6$  (p: 0.30) - TAG:  $232.5 \pm 84.5$ ;  $306.8 \pm 118.7$ ;  $288.9 \pm 63.8$  (p: 0.66) - COL:  $174.8 \pm 17.9$ ;  $181.0 \pm 35.6$ ;  $167.9 \pm 24.3$  (p: 0.50) - HDL-COL:  $42.3 \pm 5.9$ ;  $52.7 \pm 10.2$ ;  $45.9 \pm 6.1$  (p: 0.17) - LDL-COL (mg / dL):  $86.0 \pm 15.4$ ;  $56.5 \pm 26.6$ ;  $64.1 \pm 8.8$  (p: 0.11). No statistically significant differences were found between groups. JBArA did not modify glycolipid profile of these rats

### A34

#### HISTOMORPHOLOGY OF ADIPOSE TISSUE AND ITS RELATION WITH MORPHOMETRIC AND SERUM VARIABLES IN eSS RATS OF DIFFERENT AGES

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The aim of this work was to analyze the evolution of histomorphological variables of retroperitoneal adipose tissue and its relation with morphometric and serum variables in eSS diabetic rats. At 70, 100, 200 and 340 day-old respectively (6 male rats per group): body weight W(g):  $279.2 \pm 7.4$ ,  $364.0 \pm 16.5$ ,  $422.8 \pm 11.9$ ,  $450.8 \pm 40.8$  ( $p < 0.0001$ ); body mass index BMI: ( $\text{g}/\text{cm}^2$ ):  $0.6182 \pm 0.0351$ ,  $0.7320 \pm 0.0338$ ,  $0.7498 \pm 0.1393$ ,  $0.8147 \pm 0.0240$  ( $p < 0.0003$ ); thoracic circumference TC (cm):  $13.20 \pm 0.60$ ,  $14.78 \pm 0.41$ ,  $15.32 \pm 0.42$ ,  $16.97 \pm 0.27$  ( $p < 0.0001$ ); abdominal circumference AC (cm):  $17.25 \pm 0.76$ ,  $19.15 \pm 0.93$ ,  $20.95 \pm 1.07$ ,  $21.70 \pm 1.28$  ( $p < 0.0001$ ); serum levels of glucose G (mg/dl):  $109.8 \pm 19.4$ ,  $138.0 \pm 11.0$ ,  $148.2 \pm 21.3$ ,  $175.5 \pm 13.1$  ( $p < 0.0001$ ), cholesterol C (mg/dl):  $83.5 \pm 4.7$ ,  $78.7 \pm 5.5$ ,  $120.8 \pm 30.4$ ,  $387.1 \pm 42.9$  ( $p < 0.0001$ ) and triglyceride TG (mg/dl):  $121.0 \pm 15.4$ ,  $144.8 \pm 33.6$ ,  $356.5 \pm 136.1$ ,  $1217.1 \pm 247.2$  ( $p < 0.0001$ ); adipocytes area ( $\mu\text{m}^2$ ):

3991±1399,5881±1663,7353±2891,8173±2243(p<0.0001);adipocytes perimeter(μm):244.4±36.7, 298.5±38.7, 339.4±53.9, 346.1±45.6 (p<0.0001).Tukey's test:W(70vs100:p<0.001,100vs200:p<0.01,200vs340:ns);BMI(70vs100:p<0.05,100vs200:ns,200vs340:p<0.05);TC(70vs100:p<0.001,100vs200:p<0.05,200vs340:p<0.001);AC(70vs100,100vs200,200vs340:p<0.05);G(70vs100:p<0.05,100vs200:ns,200vs340:p<0.05);C(70vs100,100vs200:ns,200vs340:p<0.001);TG(70vs100,100vs200:ns,200vs340:p<0.001);area:(70vs100,100vs200:p<0.001,200vs340:p<0.05);perimeter:(70vs100,100vs200:p<0.001,200vs340:ns). At the ages evaluated, adiposity indicators IMC, CT and CA increased significantly. Adipocyte hypertrophy was lowest among 200 and 340 days. This could be associated, in part, with the marked increase in triglycerides during the same period, because the buffer capacity of the adipocytes would be exceeded.

### A35

#### DIETARY HABITS OF MEDICAL STUDENTS AT ROSARIO NATIONAL UNIVERSITY (UNR)

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An observational, descriptive, cross-sectional study is being carried out in order to describe the eating habits of students at the first cycle at the School of Medicine, UNR. Up to now, 24-hour dietary recall questionnaire have been administered in 130 students, 45 men (34.6%) and 85 women (65.4%). The majority of the students (90%) reported having had breakfast, and 80% has consumed homemade foods. When analyzing the number of daily intakes, 56.2% of the sample has eaten four meals, 22.3% has eaten 3 meals or less, and the remaining 21.5% has eaten 5 or 6 meals. Regarding food group's intake, 70.85% has consumed milk or yogurt, 56% has ingested fruits, and 80% has eaten vegetables. As regards vegetable classification, 60% has consumed raw vegetables (salads), 45% cooked vegetables, and 36% potatoes. The most consumed type of meat was the vaccine (56.6%), followed by chicken (41.7%), pigs and fish (0.9% each of them). Refined grains were consumed by 60% of the students, and cold meat by 25.4%. Candy intake was reported by 38.5% of the sample. Sugar use was reported by 47.7%, and sweetener by 20.8% of students. 83% has eaten white bread, and only 15.4% whole wheat bread. Water and soft drinks were consumed by a similar proportion of participants (16.9 and 16.2%) and alcoholic beverages by 3.1%. 11.5% reported smoking and 66.6% of thosewhodid, reported smoking 3 orlesscigarettes per day.Contrary to the expectations, our medical students are having unhealthy dietary habits. Thus, it can be concluded that some of the Argentinean food recommendations to maintain health are not met.

### A36

#### IN VITRO STUDY OF THE ANTHELMINTIC ACTIVITY AGAINST *Trichinella spiralis*(Ts) ADULT PARASITES OF ALBENDAZOL (ABZ) MICROCRYSTALLINE FORMULATIONS BASED ON CELLULOSE DERIVATIVES

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ABZ, a benzimidazole carbamate used in oral chemotherapy against intestinal parasites, has low bioavailability due to its low solubility in water. The aim of this work was to evaluate the *in vitro*anthelmintic activity against adult Ts parasites, of four microcrystalline systems formulated using hydroxypropylmethylcellulose (HPMC) as stabilizing polymer (0.1 or 0.5% w/v) and poloxamer 338 (P338) as surfactant (0 or 0.02% w/v). Pure ABZ, used as a control, and the different formulations were dispersed in RPMI 1640 medium and incubated with adult Ts females at 37 ° C, in a 5% CO2 atmosphere, for 48 h. The viability of Ts females was determined at 2, 4, 7, 24, 29 and 48 h to classify them as "alive" or "dead"; each experiment was performed in duplicate. The effect of the formulations on the viability of the parasite was evaluated with survival curves calculated with the Kaplan-Meier method and compared with the Mantel-Cox test. The decrease in polymer concentration improved the parasitocidal activity of the systems without P338 (P = 0.0052). The addition of the surfactant P338 was effective at high levels of HPMC (P = 0.0032) but did not improve the activity of the formulation when the concentration of the polymer was low. The results suggest that the 0.5% w/v HPMC system releases ABZ more slowly retarding the dissolution, and the addition of the surfactant would favor solubilization of the active component. In contrast, at low polymer concentrations, P338 would decrease the performance of the system, so it would not be desirable to include it. Microcrystalline formulations as the described would be useful to establish effective therapies with low doses of the active principle to treat intestinal parasitosis.

### A37

#### FACTORS RELATED TO NASAL COLONIZATION BY *Staphylococcus aureus* IN MEDICAL STUDENTS

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*Staphylococcus aureus* (SA) has great ability to colonize skin and mucous membranes. Factors related to both the microorganism and the host can influence the colonization process. The aim of this study was to analyze the risk conditions in the population of medical students. Of 1157 students, 287 were colonized with SA (25%). The comparative analysis of the colonized and non-colonized students was expressed as odds ratio (95% Confidence Interval), no significant differences were obtained between both groups (p>0.05). Results obtained were: sex: 0.57(0.58-1.02); smokers: 0.68 (0.4-1.03); inflammation of the respiratory system such as asthma, sinusitis, rhinitis: 1.25 (0.9-1.25); use of decongestants such as nasal sprays or corticosteroids: 0.85 (0.63-1.17); skin infections: 0.84 (0.5-1.44); students exposure to hospital environment: 0.91(0.56-1.47). The risk factors analyzed did not show significant differences regarding the colonization by SA in the group of students studied.

### A38

#### NASAL CARRIAGE OF *Staphylococcus aureus* IN MEDICAL STUDENTS OF THE NATIONAL UNIVERSITY OF ROSARIO

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*Staphylococcus aureus* (SA) is a pathogenic microorganism associated with numerous communities and nosocomial infections. It could be part of the human's usual microbiota. Several factors influence this microorganism to cause disease in humans. Among them, the increasing ability of SA to generate resistance to beta-lactam antibiotics such as methicillin. The aim of this study was to determine the nasal carriers of SA in medical students and to know its methicillin resistance profile. We study 1157 students, 287 (24.8%) were colonized with SA, of which 271 (94.5%) were susceptible to methicillin (SAMS) and 16 (5.5%) were SA methicillin resistant (MRSA). 100% of the MRSA did not present accompanying resistance, therefore they were considered community-acquired MRSA (MRSA-AC). The determination of this microorganism in the nose of medical students is very useful to avoid horizontal transmission to other people and to prevent possible dissemination in the carrier causing infections with different degree of severity. In addition, it would contribute to the therapeutic decision making for its initial treatment.

### A39

#### *Staphylococcus aureus* NASAL COLONIZATION IN MEDICAL STUDENTS FROM ROSARIO CITY: THERAPEUTIC ALTERNATIVES

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Approximately 20-40% of adult people can be colonized by the bacterium *Staphylococcus aureus* (SA). It can produce from mild infections to potentially fatal systemic diseases in situations where the defense mechanisms of the skin are altered. Its treatment represents a therapeutic challenge because of the increasing acquisition of antibiotic resistance. The aim of this study was to determine the frequency of resistance to erythromycin, clindamycin, levofloxacin, and trimethoprim-sulfamethoxazole in SA isolates from nasal colonization in medical students from the National University of Rosario. 1157 samples of nasal swabs were processed, SA was isolated in 287 (24.4%), 100% were sensitive to quinolones and trimethoprim-sulfamethoxazole. Regarding erythromycin and clindamycin resistance, 208 (72.5%) of the positive cultures were resistant, showing 82 of them (39%) inducible resistance and 126 (61%) constitutive resistance. According to these results quinolones and trimethoprim-sulfamethoxazole would be a valid alternative for the empirical treatment of infections with SA strains resistant to methicillin, being limited the use of erythromycin and clindamycin.

### A40

#### PREVALENCE AND RISK FACTORS FOR NASOPHARYNGEAL CARRIAGE OF *Streptococcus pneumoniae* AMONG A PEDIATRIC POPULATION

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*Streptococcus pneumoniae* is a major cause of bacterial infections worldwide. Asymptomatic bacterial colonization of the nasopharynx is considered to be the first step in the pathogenesis of infections. Our aim was to study in a pediatric population the prevalence and risk factors for pneumococcal carriage. Of 550 children enrolled 203 (36.9%) carried *S. pneumoniae*. The potential risk factors were first examined by univariate analysis ( $p < 0.05$ ). Subsequently, multivariate analyses was performed for those variables with  $p < 0.25$ ; the association was expressed in odds ratio (OR) and 95% confidence intervals (CI). Pneumococcal carriage was not associated with sex and previous otitis. However, multivariate analysis identify four risk factors for nasopharyngeal carriage: aged, 0.85 (0.771-0.937), child-care center, 2.611 (1.522-4.476), child at home, 2.707 (1.793-4.086) and upper respiratory infection, 1.674 (1.080-2.596). We conclude the pneumococcal carriage significantly correlated with younger age, child-care center, child at home and upper respiratory infection.

### A41

#### STUDY OF FLUID BEHAVIOR OF BLOOD IN ADOLESCENTS WITH OVERWEIGHT

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In Argentina about 20.8% of adolescents aged 10 to 19 years old were overweight. The prevalence of overweight was significantly higher in the 10-to-12 age group than older than 16 years old. Overweight adolescents may develop obesity increasing the risk for cardiovascular diseases. In the last decades, the analysis of rheological aspects of blood has become important because they are involved in the pathogenesis of these conditions. There are few studies on the fluid behavior of blood in adolescents with overweight in order to clarify the presence or not of hemorheological disorders. In this work, we have studied the fluid behavior of the blood in adolescents of Rosario city. We have studied adolescents aged between 11 and 19 years old, of both sexes, without known metabolic disorders, classified according to the BMI percentile (Normal weight:  $\geq 10$  and  $< 85$  and Overweight:  $\geq 85$  and  $< 95$ ), who have been attended the "7 de Abril" Health Center or "Centro Rosarino de Estudios del adolescente". Blood (BV) and plasma viscosity (PV) measured by cone-plate viscometer at shear rate (SR) of 230 s<sup>-1</sup>, relative viscosity (RV) estimated from BV/PV, and hematocrit (Hto) were the studied variables. The results have been analyzed by IBM SPSS Statistics v20.0.0 and it was considered significant  $p \leq 0.05$ . The results obtained did not show significant difference in Hto or PV, both determinants of BV. The BV did not differ between the groups studied nor the RV. The RV estimated at high shear rate is related to the deformation capacity of the red blood cells. These results suggest that changes in the fluid behavior of blood at high flow rates are not observed in overweight adolescents. However, in the following work, we would consider necessary to increase the number of patients and to incorporate other variables that influence over the fluid behavior of blood.

#### A42

### DEVELOPMENT OF SARCOMA E-100 IN RATS FED WITH A SUCROSE-RICH DIET

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Insulin resistance (IR) is associated with an increased prevalence of different types of cancer; however, there is no data regarding the prevalence of sarcomatous tumors in subjects with IR. Our aim was to evaluate tumor growth in controls and in IR rats (fed with 30 % sucrose W/V) (SRD) after subcutaneous inoculation of SE-100 sarcoma cells. Study groups were: G1: control animals + vehicle; G2: control animals + SE-100, G3: IR animals + vehicle; G4: IR animals + SE-100. Results: Weight gain (expressed in g) was higher in animals maintained under SRD, G1 (104.4 ± 5.1), G2 (96.8 ± 10.3), G3 (145.7 ± 11.7) and G4 (142.7 ± 10.2) (G3 and G4 p <0.05 vs G1 and G2). The relative weight of epididymal fat tissue was significantly higher (p <0.05) in G3 than in all other groups. Serum triglyceride levels were significantly higher in the SRD-fed animals than in the control group (p <0.05): G1 235 ± 22; G2 226 ± 29; G3 379 ± 33; G4 339 ± 12 mg / dl. Plasma glucose levels (AUC) after the glucose tolerance test were higher in SRD-treated groups as compared to controls (p <0.05). From the nonlinear adjustment of the tumor volume during the time elapsed since the challenge, G4 animals showed a mean time of cellular duplication (4.32 ± 0.37) greater than those of G2 (2.40 ± 0.52) (p <0.05). Despite the higher rate of tumor growth in the animals of this group, no differences arose in tumor volume in SRD and control animals.

#### A43

### KNOWLEDGE, ATTITUDES AND PRACTICES REGARDING BREAST CANCER SCREENING

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We studied factors that might influence on the practice of breast cancer (BC) screening among women from Concordia, Entre Ríos. Methods: Observational, cross-sectional study. The survey was carried out over 90 mothers from three elementary schools (32 pertaining to low (G1), 29 to middle (G2), and 29 to high socioeconomic level (G3)). Statistical analysis: Means ± standard deviations; ANOVA; Chi-square and Fisher's exact tests. Results: Age = 39.6 ± 6.8 years. Work outside home 35.5% of the mothers from G1; 74.1% from G2 and 85.7% from G3 (p = 0.001). The proportion of women over 40 years who never performed a mammography was higher (p = 0.03) in G1 (35.7%) than in the rest of the sample (7.1%). 60.4% of women who completed high school, and 43.5% of those who did not finish it are not afraid of performing a mammography because of fearing the results. The whole sample recognize period breast exams and breast self-exams as early detection measures. Most of them consider breastfeeding (G1: 66.7%, G2: 76.2% and G3: 83.3%) and to have children (85.7%; 100% and 100%) as preventives factors; meanwhile the minority points pregnancy among 20-30 years (28.8%, 4.8% and 11.1%) as well. Family history of BC was recognized as a risk factor (93.1%; 100% and 100%). The results indicate that belonging to a certain group doesn't influence on knowledge, but it does in the performing of screening tests.

#### A44

### VARIABLES REGARDING PRACTICES RELATED TO BREAST AND CERVICAL CANCER PREVENTION AMONG ADULTS WOMEN IN CONCORDIA, ARGENTINA

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The aim is to assess whether there is relationship between socioeconomic/educational level, attitudes and practices regarding routine checks for the detection of these cancers. Methods: Observational, cross-sectional study. A survey was carried out among 90 mothers from three elementary schools pertaining to low, middle and high socioeconomic level (G1, G2, G3, respectively). Statistical analysis: Means ± standard deviations; ANOVA; Chi-square and Fisher's exact tests. Results: Mean age was 39.6 ± 6.8 years. Of women attending public health care providers, 28% had never performed a gynecological examination; whereas it was found in 6.7% of women accounting for private health care providers (p < 0.001). 72.9% of women with 1-3 children and 33.3% of those with 7 children or more had a papanicolaou during the last year. Of women with complete high school education, all those who perform a breast control disagree with the statement "A screening test has a high risk of leading to unnecessary surgery". Of those who agree, only 40% do as well (p < 0.001). None of them consider that there is no possibility of cure once the cancer is diagnosed. In conclusion, there are some factors associated with non-compliance with screening that can be modified with the aim of improving the adherence of women to them. It is necessary to continue studying about these limitations in order to implement appropriate interventions.

#### A45

### DIETARY HABITS OF MEDICAL STUDENTS AT ROSARIO NATIONAL UNIVERSITY (UNR)

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An observational, descriptive, cross-sectional study is being carried out in order to describe the eating habits of students at the first cycle at the School of Medicine, UNR. Up to now, 24-hour dietary recall questionnaire have been administered in 130 students, 45 men (34.6%) and 85 women (65.4%). The majority of the students (90%) reported having had breakfast, and 80% has consumed homemade foods. When analyzing the number of daily intakes, 56.2% of the sample has eaten four meals, 22.3% has eaten 3 meals or less, and the remaining 21.5% has eaten 5 or 6 meals. Regarding food group's intake, 70.85% has consumed milk or yogurt, 56% has ingested fruits, and 80% has eaten vegetables. As

regards vegetable classification, 60% has consumed raw vegetables (salads), 45% cooked vegetables, and 36% potatoes. The most consumed type of meat was the vaccine (56.6%), followed by chicken (41.7%), pigs and fish (0.9% each of them). Refined grains were consumed by 60% of the students, and cold meat by 25.4%. Candy intake was reported by 38.5% of the sample. Sugar use was reported by 47.7%, and sweetener by 20.8% of students. 83% has eaten white bread, and only 15.4% whole wheat bread. Water and soft drinks were consumed by a similar proportion of participants (16.9 and 16.2%) and alcoholic beverages by 3.1%. 11.5% reported smoking and 66.6% of thosewhodid, reported smoking 3 orlesscigarettes per day.Contrary to the expectations, our medical students are having unhealthy dietary habits. Thus, it can be concluded that some of the Argentinean food recommendations to maintain health are not met.

#### A46

### FOOD INTAKE PATTERNS IN GALLSTONE DISEASE (GD)

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The aim of the investigation was to determinate food intakes patters in subjects from a case-control study in Rosario, Argentina. Information was obtained from 51 cases and 69 controls. Food consumption patterns were identified by principal component analysis based on food frequency questionnaires responses. For the interpretation of the components, it was decided to use the correlations between the original variables and each main component. This eliminates the effect of the variability of each of the variables in the interpretation of each component. Logistic regression models were also used to estimate the Odds Ratios and their corresponding 95% confidence intervals (CI) to measure the risk of developing GD. Two different dietary patterns were identified. The first pattern presents high consumption of animal fats, entrails, soft drinks, sugar, potatoes, refined grains, processed meats, chicken with skin and fat beef and low intake of red vegetables and yellows, cabbages, other fruits and fish (Pattern 1). The other is characterized by high intake of skinless chicken, nuts, lean beef, vitamin A and C rich fruits, other fruits and whole dairy and low consumption of chicken with skin, green leaves vegetables and sprouts (Pattern 2). The intake of the cases is characterized mainly by Pattern 1, while the controls presented consumption according to Pattern 2. When risk analysis was performed, Pattern 1 shows an increased risk of developing GD (OR = 2.3,  $p < 0.0001$ , CI: 1.68-3.26), while Pattern 2 behaves as a protector factor (OR = 0.44,  $p = 0.0004$ , IC: 0.28-0.69).

#### A47

### ANALYSIS OF THE SYSTOLIC BEHAVIOR IN CENTRAL VASCULAR PULSE PRESSURE WAVES BY HIGUCHI'S ALGORITHM

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Pulse pressure waves allows to determinate the systolic volume and the integrity of the arterial walls. They behave like a complex, nonlinear and irregular system, which has fractal properties of self-similarity and fractal dimension (FD). The objective was to analyze the pulse pressure waves behavior through the fractal determination of systolic pressure by Higuchi's algorithm. Seven non-treated hypertensive male patients between 50 and 60 years ( $55 \pm 5$ ) were analyzed. The waves were obtained by Mobil-O-Graph equipment in three different time zones per day. The FD and the determination coefficient ( $R^2$ ) with their mean (M) and standard deviation (SD) were determined every 10, 30, 60 and 200 seconds. Results: [10 sec]: FD (M 0.08; SD 0.02)  $R^2$  (M 0.5; SD 0.07); [30 sec]: FD (M 0.01; SD 0.004)  $R^2$  (M 0.46; SD 0.09); [60 sec]: FD (M 0.03; SD 0.02)  $R^2$  (M 0.46; SD 0.001); [200 sec]: FD (M 0.02; SD 0.001)  $R^2$  (M 0.5; SD 0.09). We concluded that the analyzed cases showed poor adaptation of the system and the hemodynamic behavior could adapt in brief periods. The FD obtained through Higuchi's algorithm would allow us to probe the non-adaptability of systems accurately. This leads to a deepening of the study for predictive purposes.

#### A48

### ANTITUMOR EFFECT OF METRONOMIC CHEMOTHERAPY (MCT) with cyclophosphamide (cy) and losartan (los) on m-234p MURINE MAMMARY ADENOCARCINOMA

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MCT consist of the chronic administration of low drugs doses, at regular intervals, without extended rest periods. Cy is an alkylating drug with toxic action on proliferating cells. Los is an antagonist of angiotensin II receptor, used to treat hypertension. We had shown that Cy can inhibit M-234p growth. Our present aim was to study the combined effect of Cy+Los administered metronomically. Female BALB/c mice were challenged s.c. with M-234p (day 0) and divided into 4 groups ( $n = 5-6/\text{group}$ ). GI: Control, with no further treatment; GII: Treated with Cy 25mg/kg/day in the drinking water (day 5); GIII: Treated with Los 600mg/kg/day in the drinking water (day 5); GIV: Treated as GII + GIII. Mice were weighted and tumor volume measured, 3 times/week. The combined treatment was more effective than those with each individual drug ( $P < 0.0001$ ). In 1/6 mice of GIV group there was a total tumor regression from day 47 on, with no recurrences until day 120. Survival in mice of GIV was higher than that of the other groups ( $P < 0.001$ ). No weight losses were observed. It is concluded that: 1) MCT with Cy+Los inhibited M-234p growth, resulting in one complete regression, and increased mice survival. 2) Combined treatment was more effective than the individual drugs. 3) The treatment was devoid of general toxic effects. 4) These results show the convenience of the use of the repositioned drug Losartan in antitumor treatments

**A49**

**INTERFERON AND TUMOR ESCAPE IN AN IMMUNOEDITION MURINE MODEL: CAUSE OR CONSEQUENCE?**

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Cancer immunoediting is a process with 3 phases: elimination (EL), equilibrium (EQ), escape (ES). When CBI/L mice are s.c. challenged with adenocarcinoma M-406, they show the 3 phases: 51.6% (ES), 18.5% (EQ), 29.8% (EL). Our aim was to evaluate in M-406 bearing CBI/L mice, 4 serum cytokines in the 3 phases. We observed high levels of IFN- $\gamma$  in ES, IL-4 in EQ, IL-10 in EL with no modifications of IL-2. The heterogeneity of variances in the 3 phases was, IL-10 (P=0.085); IFN- $\gamma$  (P=0.002); IL-4 (P<0.0001); IL-2 (P=0.532) and was greater in EQ than in ES and EL. The analysis of the 4 ILs with the multivariate technique of principal components (PC) showed 4 components: 1<sup>st</sup>(PC1) explained almost all the variance (95.46%), presented a negative correlation with IFN- $\gamma$  (r=-1) and with IL-4 (r=-0.699) and was not associated with IL-10 (r=-0.035). The 2<sup>nd</sup> component (PC2) explained 3.82% of the variance and was positively and significantly associated with IL-10 (r=0.692) and negatively and significantly with IL-4 (r=-0.688), without association with IFN- $\gamma$  (r=0.028). We conclude: 1) the high variances heterogeneity in EQ could indicate that in this stage coexist mice with levels of ILs compatible with ES and EL; 2) IFN- $\gamma$  would be involved in the ES phase. This behavior opens questions about its role in M-406 growth. The high level of IFN- $\gamma$  is the cause of tumor growth or the consequence of an exacerbated immune response trying to destroy, without success, tumor cells in escape? The answer to the question requires new experimental approaches to test the corresponding hypotheses.

**A50**

**COMPARISON OF THE GERMICIDAL EFFICACY BETWEEN AN ALCOHOL-BASED HAND RUB, ANTIMICROBIAL SOAP AND PLAIN SOAP IN HAND HYGIENE OF MEDICAL STUDENTS**

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Health care associated infections (HCAI) affect millions of patients annually, can be difficult to treat and potentially fatal. Hand hygiene (HH) is essential for the prevention of HCAI and is the most effective way to reduce the propagation of healthcare associated pathogens. The objective of this study was to determine which agent used in HH provided the most germicidal efficacy. A descriptive nonrandomized, prospective, experimental study was conducted on 105 medical students between 4<sup>th</sup> and 6<sup>th</sup> year. The samples were taken from the dominant hand of each student, before and after, through a sterile swab and later cultured on a blood agar (BA) plate. The alcohol-based hand rub and the two liquid soaps (antimicrobial and plain) were compared, utilizing the WHO guidelines of HH. Each BA plate was divided into 2 sections: 1) prior to, and 2) after HH. Germicidal efficacy was determined by the quantification of colony-forming units (CFU) in BA. The median values of the CFU were compared, before and after; by applying the nonparametric Wilcoxon test for related samples. Those students who used plain soap (PS) had a 21% reduction in CFU, those who used antimicrobial soap (AS) 84%, and those who used an alcohol-based hand rub (AB) 94%. There was no significant statistical difference between AS and AB (p = 0.233), but there was a difference between AS and PS (p <0.001), as well as between AB and PS (p <0.001). In conclusion, all three agents decreased CFU after HH. The median value of CFU with AB was lower in comparison to the other two agents. There was no significant difference between AB and AS; however, both agents proved to be superior to PS in their germicidal efficacy.

**A51**

**MONOSODIUM GLUTAMATE DETECTION THRESHOLDS AND ANTHROPOMETRIC NUTRITIONAL DIAGNOSTIC**

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The objective was to describe the monosodium glutamate detection thresholds and the students' anthropometric nutritional diagnostic. The participants were 152 students from the Facultad de Bioquímica y Cs. Biológicas, UNL (121 women and 31 men; aged between 18-30 years old). Subjects were normal-weight (n=105); overweight (n=33) and 7 were obese. Only 6 students were underweight. Detection thresholds tests were done with seven GMS concentrations according to the method of Elman et al (2013) and the nutritional diagnostic was obtained with the weight and height. A possible association among them was assessed by using Chi-square (X<sup>2</sup>). The results showed that the two lowest GMS concentrations (0.254 g/l and 0.49 g/l) were the most detected by the 30% and 39% of the people, respectively. After that, detection thresholds progressively decreased while GMS concentrations increased. The concentration of GMS for the 50% of the population was 0.3068 g/l. There was no association between GMS detection thresholds and the IMC (p=0.843). We concluded that the majority of the students were normal weight and that the two first GMS concentrations were the most detected, independently of nutritional status.

**A52**

**COMPARISON OF KNOWLEDGE OF STUDENTS, BEFORE AND AFTER RECEIVING A TRAINING INTERVENTION**

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Our objective was to determine if, through intervention with clinical cases, the academic performance of the students after the class is modified and if they acquire fundamental knowledge about the subject matter. 56 students participated in the intervention. The comparison between the note of a previous evaluation and a subsequent one is made. The evaluation consisted of 10 multiple choice questions about the contents that were addressed in the class through clinical cases. To determine the effectiveness of an experimental procedure whose observations are obtained from non-independent samples, the Pareto Comparisons Test is used. It concluded that through the intervention with clinical cases a remarkable

improvement in the students' grades is achieved, interpreting this as a greater understanding of the topics addressed in the class and inferring that the main contents were apprehended correctly by the same ones. In addition, this methodology requires a greater commitment on the part of the students, since they must participate actively in the intervention to achieve a better performance in the post-class evaluation.

### A53

#### APPLICATION OF CLINICAL CASES AS A TEACHING METHODOLOGY

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Biochemistry is an annual subject in second year of medical school. Auxiliaries are advanced students who while studying other assignments are also acting as tutors in educational interventions carried out by students of Biochemistry and supervised by a teacher. The application of clinical cases in the development of the theoretical classes of the Biochemistry course in Medicine, allows students to understand the basic contents and apply them to medical practice in the near future. Two theoretical classes were given: the first class (without clinical case) and a second class (with clinical case). A survey, previously validated, was applied during the class and it addressed the different methodologies used in each class. The results shown that methodological approach through the application of clinical cases generated more participation and motivation in the students during the classes, since the most of them agreed that learning by using clinical cases was very helpful and they could apply their knowledge to the daily medical practice.

### A54

#### ASSESSMENT OF SECOND YEAR MEDICAL STUDENTS ON DIAGNOSTIC EVALUATION IN BIOCHEMISTRY

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Biochemistry is an assignment for the second year of medical school. The course begins with a diagnostic evaluation with questions over previous knowledge that students must have before the assignment. Objectives: Utilize the second-year learned knowledge. Material and methods: To apply previous knowledge that pertains to evaluation of diagnostics. In 2015 and 2016 the "Chi-Cuadrado" Test was given. In 2015, 38 students were asked to participate and gave consent to the evaluation, while in 2016, 59 students did so. Results: 2015- 18 opposed to 20 positive. 2016- 20 opposed to 46 positive. The grade of association to determine of what importance given in the results can be calculated by the Odds Ratio (OR)= 3.18; strong grade of association. Interval of confidence of 95% of the OR= 2.27, 4.45 respectively. Conclusion: It is implied that the diagnostic evaluation was of great importance for the students in 2016 as a strategy of self-evaluation at the beginning of the course and they could confront their weaknesses and work towards the achievement of the assignment.

### A55

#### IN VITRO STUDY OF RELATIONSHIP BETWEEN FORCE AND HARDNESS IN PIECES OF HUMAN DENTAL ENAMEL WITH AND WITHOUT FLUOROSIS

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Hardness is considered an essential physiological property of human dental enamel. Much more serious is the fluorosis, teeth pieces are subject to extensive mechanical failures of the surface. Objective: To compare relationship between force and hardness (F-H) in samples of human dental enamel with and without fluorosis. Material and methods: *In vitro* experimental study. Dental pieces of adult humans derived from extractions due to periodontal problems or orthodontic treatment were used. N=11 teeth with fluorosis and n=11 without fluorosis were included (beta=10% y alpha=5%, maximum error admitted at simple size calculation). From each tooth, 4 blocks were cut, each of one catted parallel to respective tooth surface (vestibular, mesial, palatine-lingual, distal); those little pieces were included into acrylic resin and polished. A Vickers microhardness test was used, consisting in measuring the diagonal lengths of an indent left by indentation of a diamond pyramid with a given load into the sample material. The F-H relationship was measured with Spearman coefficient (Sp), adjusting by group (fluorosis: yes-no) and by dental surface (palatine/lingual, mesial, distal, vestibular); statistical significance level: P<0.05. Results: F-H relationship showed statistically associated in the group of teeth with fluorosis (Sp= 0.774 P=0.009) but not in the group without fluorosis (Sp= 0.249 P=0.586). Adjusting by dental surface, F-H relationship was statistically significant in fluorosis group (Sp= 0.699 P=0.024 in mesial side; Sp=0.964 P= 0.001 in distal side; Sp=0.701 P=0.036 in palatine-lingual side and Sp=0.617 P=0.077 in vestibular side), but not in the group without fluorosis. Conclusion: Evidence showed that F-H relationship was statistically significant only in dental pieces with fluorosis, suggesting that dental fluorosis involving a weakening of complete dental structure.

### A56

#### COMPARATIVE STUDY OF PULP CALCIFICATIONS INCIDENCE IN RATS MOLARS: DIABETICS VS CONTROLS

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Our objective was to compare calcification incidence in control and diabetic rat molar. Male rats line IIM / Fm sub-line "m" of 3 weeks of age were randomly divided into 2 groups: diabetics and controls. Diabetes was induced according to the Prahl and Steenrod method. The treated animals' glycemia was measured by the enzymatic method (Trinder). Animals were killed from the 6th to the 11th week of life (n = 30). The jaws were dissected. Hemimandibles and a variable-thickness aluminum scale were x-rayed on. The films were digitized. In both groups was evaluated: the presence of pulp calcifications in each of the 3 molars, calcifications number in each molar, localization within the piece and mineral density of chamber and root canal (D.M.O). Digital image analysis software (Image 3) permitted absorbance comparison.

Diabetic animals had higher percentage of calcifications than controls. Calcifications were significantly higher in 2nd and 3rd molar of diabetic rats, not noting statistical significance in 1st molar. In both group were observed greater amount of calcifications in camera than in root canals. Diabetic rats showed higher number of calcifications in camera and root canal respect to controls. In diabetic rats the D.M.O of 2nd and 3rd molar were significantly higher than controls, not observed difference when comparing the 1st molar. Rats have high rate of calcifications, these most important being on camera than in root canals. Diabetic state increased the incidence of calcifications mainly 2nd and 3rd molar, probably as a result of an increase in the concentration of osteopontin because chronic hyperglycemia.

#### A57

### FISHERMEN AND FAMILY IN THE ROSARIO CITY COAST AND NEIGHBORING ISLANDS: DMF INDEX, CONSUMPTION OF SWEETS, ITS FREQUENCY AND DENTAL BRUSH

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Residents on the coast access urban dental centers, those living on the Charigue and Wintering islands must relocate. They need to apply preventive measures: choose the consistency of sweet foods, reduce frequent intake and brush daily. Objective: to relate buccal condition, type of sweets, frequency and adherence to brushing. Subjects aged 15-45 years, both sexes by residence; the average DMF index was obtained; Sweet foods: adhesive and adhesives little, frequency and brushing on every day, sometimes and never. Chi<sup>2</sup>; Fishers Test. Of the 50 patients, 40% live on the coast, 30 years average; 38% in Charigue, 32 years and 22% in Wintering, 31 years old. No difference in DMF index (p=0.706); Decayed (p=0.342) and Missing (p=0.456); difference in Filled, greater in Wintering (p=0.0015). **Adhesives:** sweet of milk was not consumed in the coast, in Charigue it consumed 37% and 9% in Wintering (p=0.002). No difference in: soft candies, alfajores, jams, masitas and quince and sweet potatoes. **Little adhesives:** chocolate was ingested for 20% of the coast, none of Charigue and 45% of Wintering (p=0.005). No difference in: lollipops, hard candies and cakes. They sometimes consume: 85%; 95%; 91% in order (p=0.315). Brush daily: 25%, 74% and 55% respectively (p=0.033). In Wintering, they restored more than in the coast and Charigue. Of the less adhesive, the chocolate, was consumed in the coast and Wintering and did not differentiate in the other sweets. Sporadic consumption is favorable but daily brushing, which is greater in islanders, should increase in coastal residents.

#### A58

### PATIENTS OF CITY OF ROSARIO: AGE, INDEX OF CARIOSOS TEETH, MISSING AND FILLED, DENTAL SIZE AND BRUSH FREQUENCY

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Caries and periodontal disease are produced by bacterial plaque. It is called dental plaque to the one related to the development of caries and periodontopathological to which the periodontium of insertion sickens. One begins demineralizing to the enamel, advances with an ulcer and without treatment destroys the tooth, the encias inflamed causing mobility and loss. The teeth, with shape and size according to the face, should be brush at least three times a day. Objective: to determine DMF index; Dental size and brushing frequency in patients aged 15-45 years attending public and private dental services in Rosario City. A standardized anamnesis addressed to people of the provincial, municipal, and private service, we investigated age and average DMF index; We classified dental size into large, medium and small; Frequency of brushing in one, two or three times a day and correct occasions: after breakfast, lunch and dinner. Of 333 people, 52% attended the provincial, 26% to the municipal and 22% to the private (p<0.001); Average age 26 years, 24 and 30 respectively. DMF index average order 25 ± 4.10; 25 ± 4.26 and 23 ± 4.34. Dental size: provincial large 62%, medium 27% and small 11%; municipal: 34%, 49% and 17%; private: 49%, 41% and 10% respectively (p=0.002). Three times a day they brush 26%, 24% and 21% respectively (p=0.108). After breakfast: provincial 14%, municipal and private 18% (p=0.5); after lunch: 31%, 43% and 27% in each service (p=0.14) and after dinner: 64%, 55% and 56% respectively (p=0.4). The highest number of consultations was in the public sector at an early age, with people brushing three times a day more than in the private service, the first having a higher DMF index, there was no relation to the size of the teeth.

#### A59

### RETZIUS STRETCH MARKS AND AMELO-DENTINAL LIMIT IN PRIMARY AND SECONDARY TEETH: A DESCRIPTIVE STUDY

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The crown of primary teeth is small and little mineralized; in secondary teeth, the crown is large and more mineralized. Is formed by enamel and dentine, they have different embryological origin and they originate an amelo-dentinal limit net crossed by dentinal structures (dentinal canaliculi, adamantine spindles and Linderer plumes) that end in the enamel. Optical microscope in primary and secondary enamel show brown or yellow stretch marks called of Retzius, which represent stages of lower mineralization during coronal formation. Objectives: Objectives: compare amelodentinal limit and stretch marks. With 7 primary teeth (incisors, canines and molars) and 7 secondary homologues (incisors, canines and premolars), we mark half of crown, we cut them with metal disc at low speed and split them in approximately equal halves. One was prepared by wearing, rubbed on wet stones until a translucent sheet was obtained and then adhered to the glass. At lower magnification, the limit was classified, visible in its entirety in complete and incomplete if it showed visible and not visible parts; Amount of stretch marks in one or several and its thickness in thin, wide or both. Using a higher magnification, not visible part, the cause was analyzed. Primary: incomplete limit 86%; Secondary: 100% complete. Primary: one stretch marks 43% and several 28%; Secondary, several 86%. Primary: thin 57% and both 14%; Secondary, thin 86%. In primaries, clustered dentinal structures caused the incomplete limit. In secondary these were occasional. In both dentitions the limit differs. In secondary, by volume, there was more than one stretch marks, most of these were thin on both primary and secondary teeth. We should go deeper into this topic to corroborate our findings.

### A60

#### ANALYSIS OF STUDY METHODOLOGIES AND ACADEMIC PERFORMANCE IN THE 1ST BIOCHEMICAL TEST IN 2ND YEARS OF MEDICINE STUDENTS

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The analysis of the methodologies used by the students allows the adaptation of didactic resources and instances of learning, to improve the teaching and learning process. The purpose of this study was to analyze the methodologies and study strategies of students of Biochemistry, and their performance in the first test of the subject. Material and methods: Surveys were provided to 33 students after the partial examination, investigating the study techniques used to prepare the test; the time spent; the total of subjects covered, and the final result of the test. Results: 13 students studied less than 60% of the subjects, of which only 1 was approved and 12 were disapproved. 21 students studied more than 60% of subjects, of which 10 passed the exam and 11 did not pass. ( $X^2 = 5.84$ ,  $p < 0.05$ ). Of the total number of approved students (11), the methodology that gives the highest probability of passing the test is the completion of abstracts (Proportion = 0.46, calculated 5/11). Of the total students evaluated: 5 out of 10 left subjects without studying, and 5 out of 10 did not. According to the degree of priority that the students gave to the topics of the partial, 6 out of 10 students prioritize one topic over another. At the time of the test only 2 out of 10 students saw, studied and assimilated between 80 and 100% of the subjects corresponding to the test. Conclusion: The realization of abstracts as a study technique that results in a high probability of passing the test, means that it can be taken as a methodology to be promoted and improved in the field of classes and consultations of the subject.

### A61

#### DETECTION OF VECTOR-BORNE PATHOGENS IN DOGS FROM CORDOBA AND SANTA FE, ARGENTINA

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The diagnosis of pathogen agents responsible of canine vector-borne diseases (CVBD) presents a lot of difficulties, so constitutes a challenge for researchers. The aim of this study was to investigate the frequency and distribution of these pathogens in some locations of Santa Fe and Córdoba. Blood samples from 70 healthy dogs were taken. The detection of CVBD agents was realized by: 1-observation of colored blood smears; 2-DNA isolation and specific PCR amplification; 3-species identification by DNA sequencing. Hemotropic mycoplasma (HM) (*Mycoplasmahaemocanis*, *M. haematoparvum* and *M. suis*) were found in 77.1%, *Anaplasma platys* 15.7% and *Babesia canis vogeli* in 7% of the dogs; two *Bartonella* species were detected too. Only few previous reports involved *Anaplasma*, *Babesia* and *Ehrlichia* in Argentina; prevalence of *A. platys* in Córdoba (12%) and Santa Fe (17.8%) were similar meanwhile prevalence of *Babesia canis vogeli* in Córdoba (10%) and Santa Fe (6.8%) were higher to those reported for these two agents. HM species were the most prevalent pathogens in the studied dogs: *M. haemocanis* was detected in 66.7% and 39.1%, and *M. haematoparvum* was detected in 16.7% and 39.1% of dogs from Córdoba and Santa Fe respectively. *M. suis* was detected in two dogs; this is the first report about the presence of *M. suis* DNA in dog blood. Most of these CVBD agents found in healthy dogs are zoonotic.

### A62

#### STUDY OF A CADAVERS CONSERVATION TECHNIQUE WITHOUT THE USE OF FORMALDEHYDE

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The knowledge of domestic animal anatomy is fundamental in the training of the future Veterinarians. This science requires that the conservation of the specimens retain the original characteristics. Formaldehyde has been used for biological specimen preservation; however, its toxicity and its carcinogenic effect led to investigate an alternative for its replacement. We evaluated a technique that did not use formaldehyde - modified Thiel- for the preservation of anatomical pieces in veterinary anatomy. Modified Thiel technique was proved in ten New Zealander rabbit breed. Conservative solution "A" was injected. All the corpses were immersed in "B" solution during 30 days for impregnation and fixation, and the dissection was performed. Pelvic limbs were chosen as study pieces and were preserved for 7 months. Fungal contamination and putrefaction were evaluated and macroscopic morphology was observed every 15 days. During dissection, anatomical pieces had fresh features and provided a good alternative for manipulation at any time of conservation. It was verified that the adapted technique can be used in domestic animals, conserving the volume, color and tissue elasticity without putrefaction or fungal contamination.

### A63

#### EVALUATION OF ANTIBODY KINETICS AFTER VACCINATION AGAINST LEPTOSPIROSIS IN DOGS

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The technique of microscopic agglutination test (MAT) detects IgM and IgG agglutinins. The aim of this work was to determine through MAT if agglutinin titers after vaccination are the same or surpass the test cut point in a period of 90 days after vaccination against *Leptospira interrogans* in dogs. Blood sera from 16 dogs of different ages and sex from Carcaraña and Soldini Cities, Santa Fe, were analyzed. Samples were serologically analyzed prior to vaccination, and two doses of a bivalent inactivated bacterin were administered at an interval of 21 days between doses. Sera samples were obtained at 15, 30 and 90 days after vaccination. Blood sera were analyzed through MAT and Icterohaemorrhagiae and Canicola, (serovars included in the commercial vaccine); Pomona, Grippotyphosa, Pyrogenes and Castellonis were tested. The cut point was 1:100. Out of the 16 dogs tested prior to vaccination, one was positive and was ruled out. The analysis after 15

days of vaccination showed 4 (4/15) seropositive animals, 1 reaction to Icterohaemorrhagiae with a 1:200 titer and 2 to Canicola with 1:100, and the last showed coagglutination and the highest titer was 1:200 to Canicola. On day 30 postvaccination 14 (14/15) dogs resulted seropositive, 2 to Icterohaemorrhagiae with 1:200 and 1:400 titers, and 4 with 1:100 to 1:200 titers to Canicola; the remaining dogs showed coagglutination with titers ranging from 1:100 to 1:200. The serum from the dog showing coagglutination at day 15, coagglutination was observed and also a 1:1600 titer to Canicola. After 90 days postvaccination, 1 (1/15) dog remained reactive to Icterohaemorrhagiae with a 1:100 titer. Results showed that after 90 days of vaccination most dogs did not show detectable MAT antibody titers.

#### A64

### REPRODUCTIVE CHARACTERISTICS OF MALTESE DOG BREED

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A study of 86 parturitions of Maltese bitches was carried out between 1st January 2014 and 1st January 2016 with the aim of determining the average number of puppies per litter, percentage of labor dystocia over the total labors, percentage of males and females born and causes of dystocia, as well as the incidence of clinically detectable congenital pathologies. Over 86 labors, 59 were normal and 27 were dystocia. In the dystocia labors, the most frequent cause was abnormally positioned fetus (39%); followed by primary atony (30%), secondary atony (23%) and lack of dilation (8%). The total number of puppies born was 195, an average of 2.3 puppies per litter. Comparing puppies born in normal labors and those born in dystocia labors, the average is 2.2 and 2.4 puppies per litter respectively. This difference is not statistically significant; therefore, there is no relationship between puppies born in dystocia or in normal labors. When evaluating the percentage of males and females born, the percentages are 52% and 48% respectively. When evaluating viability and vitality of puppies born, it was observed that there was a 5% of stillborn puppies in normal labors and an 18% of stillborn puppies in dystocia labors. A chi-square test was performed and the conclusion was that there is a relationship between the perinatal stillbirths and the type of labor, which allows to state that dystocia labors increase the percentage of perinatal mortality. When studying the puppies born with congenital alterations clinically detected, it could be concluded that the percentages (3.01% in normal labors and 3.2% in dystocia labors) were not statistically significant. Among the congenital alterations clinically detectable, the most frequent was the cleft palate, followed by Amelia, hydrocephaly and gastroschisis. It can be concluded that the results observed in this breed do not differ significantly from those reported for the canine species in general.

#### A65

### EFFECT OF POLYPHENOLS IN LAYING HENS DIET ON FLY POPULATION AND EGG WEIGHT

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In this work, polyphenols from red quebracho (*Schinopsis lorentzii*) were added to the diet of laying hens as an alternative to the Integrated Pest Management (IPM) with the aim of evaluating the impact on fly population in laying hen barns and their incidence on egg weight (EW). Two 4x100 mts barns, A (Treated group) and B (Control group), with 5,000 24-week Hy-line (colour) laying hens each were studied for a year. Both barns had identical environmental and handling conditions. Both groups were fed balanced feed save for group A which was also given polyphenols from the cortex of red quebracho at 70.9% soluble tannin per 1,000g/tn feed. The variable used as response to treatment was fly larvae density per grid (0.20 m<sup>2</sup> grid side) in guano: high density (>50 larvae), moderate density (between 10-50 larvae) and low or null density (<10 larvae). Ten monthly samples were taken from each barn for 12 months placing the grids in a zigzag pattern every 10 metres under each cage line and collecting the sample at 5cm depth. The comparative analysis showed that in group A larvae density was lower or null

and samples with high larvae density were significantly lower ( $\chi^2 = 8,12$ ,  $p < 0,05$ ). EW was studied after weighing 30 eggs monthly, at random and individually from each lot (200g  $\pm$  0.01g). Compared with the SENASA standards, group A had an average EW of 63.2-76.8g with a 95% reliability placing it in the extra-large category while EW in group B was 64.8-74.9g. No significant differences were observed in the average EW between groups. The low larvae density in group A may be due to the substrate modification, making it unsuitable for larvae reproduction and feeding, improving the environmental conditions and the IPM efficiency, and showing a positive impact on the economic, social and environmental sustainability.

#### A66

### INFLUENCE OF HEAT STRESS ON WHITE BLOOD CELL TOTAL COUNT AND NEUTROPHIL/LYMPHOCYTE RATE IN HOLANDO ARGENTINO DAIRY COWS

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The aim of this study was to evaluate the effects of heat stress on WBC total count and neutrophil/lymphocyte rate in response to cortisol release in dairy bovines. It was carried out in the Milking Parlor Module of the Facultad de Ciencias Agrarias (Zavalla). Thirty cows were used. Samples were taken under thermoneutrality and heat stress conditions at 7 am and at 7 pm milkings. ITH was calculated by Thom adapted formula. Blood from coccygeal vein was used to determine cortisol by radioimmunoassay, a second sample from the same vein was taken to determine WBC total count using a haemocytometer and neutrophil/lymphocyte rate was made from blood smears (May-Grünwald-Giemsa). A statistical program was used. There was a significant increase in cortisol levels under heat stress conditions. A significant decrease in WBC count was observed in 7pm sampling under heat stress conditions. Neutrophil relative ratio showed higher values under heat stress conditions. Lymphocyte percentage showed lower significant values in those samplings made under heat stress conditions. Thereby, a significant increase on neutrophil/lymphocyte rate was observed under heat stress conditions. A slight to moderate correlation ( $r=0.328$ ) between cortisol and neutrophil/lymphocyte rate under thermoneutrality conditions was showed. There was no correlation ( $r=0.085$ ) between these variables under heat stress conditions. It could not be concluded that serious stress impacts on the WBC total count and neutrophil/lymphocyte rate.

**A67**

**AN ESSAY ON AN ALTERNATIVE DIET ADDING GREEN FODDER HYDROPONICS FOR MEAT PRODUCING RABBITS**

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Forty eight white New Zealand rabbits with an initial weight of 1,721.77grs (55.24gr standard deviation) were studied in two groups equally divided at random. The experimental group was distributed in two rabbits per cage totaling 12 cages. The assay lasted 28 days (from November - December) during which GFH was given in equal quantities. The GFH was obtained from the produce of 0.21m<sup>2</sup> trays sown with 1kg of wheat seeds and 80gr of commercial balanced. The control group, distributed in 12 cages was fed the same commercial balanced food without supplementation and ad libitum. An average 5,206g of GFH were fed daily to the experimental group. It was observed that there were leftovers of an average 1,300grs the first 9 days, from day 10 to 17, the leftovers were 423gr a day and from day 24 to the end of the experiment, the rabbits ate all the GFH given. No diarrhea or meteorism attributable to GFH was observed. Comparing body weight gain in both groups, the experimental group showed a lower average gain than the control group, and an apt slaughter piece was obtained at an age prior to heat signs in the experimental group.

**A68**

**ANIMAL BITE ACCIDENTS IN CHILDREN LESS THAN 15 YEARS FROM ROSARIO, SANTA FE**

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Previously we showed that in Rosario city the risk of animal bite accidents (ABA) is different in the children according to the districts and they are majority at the expense of dog bites. The aim of this study is to evaluate the risk of ABA caused by dogs in children according to age, sex and characterize the site of injury. It was conducted a retrospective cohort study, in children less than 15 years of age was carried out during the years 2012-2013, based on information obtained Rabies-prevention office in Carrasco Hospital, Rosario. Means and its standard deviation (SD) were used as descriptive measures. Relative Risk (RR) calculations and adjusted rates were made using population projections based on data from the 2010 Census (Municipality of Rosario). Correlation analysis was performed using Rho de Spearman. It was considered significant  $p < 0.05$ . In the period studied 717 ABA were recorded in children younger than 15 years. It was found that boys (B) have RR significantly higher than girls (G) to suffer these accidents.  $RR_{B/G} = 1.769416 (1.518717-2.061498)$ . This RR is maintained in the different age strata studied. Three strata were considered: children between 0-4 years; children aged 5-9 years and children aged 10-14 years. Adjusted rates (percent) for the risk of ABA, according sex (95% CI) and based on the risk in both sexes in the oldest group of children were: 0-4: 158.8 (141.5 -176.2); 5-9: 135.2 (117.6 - 152.8) and 10-14 years: 99.9 (84.9 - 115.1). 97.1% of the lesions were in the areas classified as head and neck (1), upper limbs (2) and lower limbs (3). Younger children were more prone to (1) injuries, while in older strata were more prone to lesions to (3) ( $p = 0.0001$ ). It was found that male children are at higher risk for the bites of dogs and that risk of these is higher as the age stratum considered decreases. In addition, according to the site of injury the smaller ones were more prone to potentially more serious injuries.

**A69**

**PARASITISM BY ENTOMOPATHOGENIC NEMATODES (STEINERNEMATIDAE, HETERORHABDITIDAE) IN *Botanochara octoplagiata* (Spaeth, 1909) (COLEOPTERA: CHRYSOMELIDAE)**

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*Heterorhabditis bacteriophora* (HB) and *Steinernemamarum* (SR) have a lethal effect on the insects. *Botanochara octoplagiata* feed on sweet potato leaves, causing damage. The infectivity of HB N54, HB N82, and SR (OLI) was evaluated in adults of *B. octoplagiata*. Individual infections were performed. Five hundred infective juveniles (JIs)/host and one control group were used. Ten individuals were considered. Life cycle aspects of the nematodes were determined: number of generations, development of parasitic stages, and duration of the parasitic phase. HB N82 caused higher mortality than HB N54: 60% vs. 30%, whereas SR (OLI) caused 50% mortality. With the isolates of HB the insect was killed, but the nematodes could not complete its development since it was annulled by the insect immune system. In insects infected with SR (OLI), emergence of JIs was recorded, obtaining an average value of 197 JIs/host. It has been pointed out that heterorhabditids are more infective, due to the presence of a tooth that allows them to enter the host not only by the natural openings, as do the steinernematids, but also by penetration of the cuticle. It is again verified that SR (OLI) causes a similar mortality to the heterorhabditids. Its infectivity is superior since it was the only one that was able to complete its life cycle in *B. octoplagiata*.

**A70**

**INFECTIVITY OF *Heterorhabditis bacteriophora* (NEMATODA: HETERORHABDITIDAE) IN PHYTOPHAGOUS INSECTS**

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Aphids and bedbugs fed on ornamental plants and can transmit pathogenic microorganisms, whereas cutter ants carry seeds, pieces of flowers or leaves. *Arabidopsis thaliana* (Brassicaceae) is used as an experimental model for the study of plant-insect interactions and may be affected by larvae of dipterans, which feed on their roots. *Heterorhabditis bacteriophora* has a lethal effect on its hosts, and it is used as a biological control agent. It was evaluated in laboratory the infectivity of the isolate N54 in *Aphis craccivora*, *Pachylisargentinus*, *Acromyrmex* sp., and *Bradysia*

sp. Individual infections were performed. Fifty infective juveniles (IJs)/insect and one control group were used. It was considered 14 individuals of each species. Mortality was higher than 80% in flies, and less than 50% in the rest of the insects, with aphids being the least susceptible (13.3%). The higher mortality of larvae of *Bradysia* sp. shows that the larval stages are the most susceptible to parasitism by this nematode. The susceptibility is lower in adult stages, as was observed for *P. argentinus*, *Acromyrmex* sp. and *A. craccivora*. This could be due to the more chitinized integument of adults that would hinder the penetration of IJs and, to the insect defence mechanisms, which could interfere with the nematode symbiont bacterium. There are differences in the susceptibility of the four species tested. It indicates the need to know which is the most efficient isolate for each insect to be controlled.

#### A71

### **ABUNDANCE OF *Aedes aegypti* AND RELATIONSHIP AMONG OFFER-CRIDERS FOR THE VARIABLE MATERIAL OF THE RECIPIENT IN THE CITY OF DIAMANTE, ENTRE RÍOS**

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The abundance of *Aedes aegypti* in urban areas is a sanitary problem of importance in our country because of the diseases it transmits. The objective of this work was to estimate abundance through indices, to study the relationship between abundance-offer of hatchery (container with water), and to analyze the relationship between the presence of immature-material of the recipient. The survey was carried out in April 2015 in the city of Diamante. For the study were selected 64 blocks of the city, and of these, 89 dwelling, all at random. In each dwelling the open spaces were inspected and recorded all possible recipient of water, the type of material and if they possessed immature mosquitoes. Three indexes were calculated for the city: housing (IV), containers (IR) and Breteau (IB). Of the total number of dwelling surveyed, 81 dwelling had recipients, of which 473 recipients were found in total containers of water and 178 contained water, of which 15 had hatcheries. 65% of the recipients containing water were made of plastic, 22% glass, 7% metal and 6% other materials, of these recipients, 8 plastic, 3 glass and 4 other materials were breeding. The indices gave: 12.4 for IV, 8.99 for IR and 17.97 for IB. This study shows that the greater proportion of breeding sites in plastic and glass recipients would be contributing to the level of infestation found, responding to this marked offer of breeding sites for this mosquito. These containers were commonly used by the owners, but were in the open and had standing water. Greater control is essential in every home

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