# **ROSARIO SOCIETY OF BIOLOGY**

# XLI ANNUAL MEETING (Online modality)

# The impact of emergent issues in Biology

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Abstract Book



ABSTRACTS A01 / A32

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#### ABSTRACTS

#### A1

# IMPACT OF THE BODY MASS INDEX ON THE SPERM NUCLEUS AND SEMINAL PARAMETERS IN INFERTILE MEN

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Obesity and overweight are the results of an imbalance between caloric intake and energy expenditure. The body mass index (BMI) (kg/m<sup>2</sup>) is frequently used to identify and classify weight gain in individuals. The hypothalamicpituitary-gonadal axis is dysregulated by excess weight, affecting the normal development of spermatogenesis. The WHO defines overweight as a BMI greater than or equal to  $25 \text{ kg/m}^2$  and obesity as a BMI greater than or equal to 30 kg/m<sup>2</sup>. The aim of the work was to analyze the impact of BMI on the sperm nucleus and seminal parameters in a selected population of infertile men. A retrospective observational study of 332 men aged between 22 and 48 years who attended URHMA with their partners from October 2017 to February 2021 was carried out. Three groups were formed: G1 (N = 98) samples of men with BMI  $\ge$  18 and < 25; G2 (N = 126) samples from men with BMI  $\ge$  25 and < 30; G3 (N = 108) samples from patients with a BMI  $\ge 30$ . Sperm motility was analyzed with the ISAS-LAB computerized system (Proiser). Sperm concentration was determined in Makler Chamber and sperm morphology with hematoxylin. The state of nuclear chromatin condensation was evaluated with aniline blue (AA) and the integrity of nuclear DNA with acridine orange (NA). The Student's t-test was applied to compare the averages of the analyzed variables. MP (% progressive mobile spermatozoids) G1:  $60.58 \pm 25.98$  vs. G2:  $57.66 \pm 26.10$  (P = 0.210); G1: 60.58 $\pm$  25.98 vs. G3: 52.67  $\pm$  26.14 (P = 0.022); G2: 57.66  $\pm$  26.10 vs. G3: 52.67  $\pm$  26.14 (P = 0.102). C (million spermatozoids/mL semen) G1:  $51.31 \pm 48.26$  vs. G2:  $57.87 \pm 48.47$  (P = 0.186); G1:  $51.31 \pm 48.26$  vs. G3:  $46.98 \pm 100$ 40.42 (P = 0.257); G2: 57.87 ± 48.47 vs. G3: 46.98 ± 40.42 (P = 0.063). M (% gametes with normal morphology) G1:  $4.67 \pm 2.10$  vs. G2:  $4.72 \pm 2.11$  (P = 0.427); G1:  $4.67 \pm 2.10$  vs. G3:  $3.89 \pm 2.11$  (P = 0.006); G2:  $4.72 \pm 2.11$  vs. G3:  $3.89 \pm 2.11$  (*P* = 0.004). AA (% spermatozoids with mature nucleus): G1:  $73.45 \pm 16.12$  vs. G2:  $68.38 \pm 16.59$ (P = 0.012); G1: 73.45 ± 16.12 vs. G3: 68.87 ± 16.21 (P = 0.022); G2: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G2: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G2: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G2: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G2: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G2: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G2: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G2: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G2: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G2: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G2: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G3: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G3: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G3: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G3: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G3: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G3: 68.38 ± 16.59 vs. G3: 68.87 ± 16.21 (P = 0.022); G3: 68.87 ± 16.21 0.405). NA (% spermatozoids with native DNA): G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  vs. G2:  $57.97 \pm 23.48$  (P = 0.059); G1:  $63.03 \pm 23.88$  (P = 0.059); G1:  $63.03 \pm 23.88$ 23.88 vs. G3:  $55.66 \pm 24.91$  (P = 0.017); G2:  $57.97 \pm 23.48$  vs. G3:  $55.66 \pm 24.91$  (P = 0.236). Significant differences were found in the group of obese men compared to the group with normal BMI in the variables MP and M. In both groups with abnormal BMI, alterations in nuclear maturity and in the integrity of sperm DNA were obtained. It is important to evaluate the BMI in the study of the male factor since obesity and overweight represent risk factors for sexual and reproductive dysfunctions in men that negatively affect sperm function influencing reproductive processes.

#### A2

## SPERM DNA INTEGRITY AND TRANSFERRIN IN HUMAN SEMINAL PLASMA

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Sperm DNA integrity is crucial for successful fertilization. On the other hand, iron has an important role in spermatogenesis. Iron transport involves transferrin protein (Tf). In seminal plasma (SP), an isoform of transferrin, testicular transferrin (TTf), is secreted by Sertoli cells. The aim of this work was to statistically evaluate the relationship between levels of TTf in SP and the degree of denaturation of sperm DNA. 49 semen samples belonging to 18 healthy volunteers and 31 patients who consulted for infertility were studied; semen analysis was performed according to the WHO (2010) standards. The acridine orange (AOT) technique was used to evaluate the degree of denaturation of sperm DNA. TTf concentrations were determined by radial immunodiffusion (RID). Statistical analysis was performed by the Spearman ordered rank correlation coefficient (r) for TTf in PS versus degree of denaturation of sperm DNA. The results obtained were: r = -0.4398; *P*-value = 0.0055; *P* < 0.01. According to these results, it is concluded that the percentage of spermatozoa with DNA in the native state (intact) presents a negative association with the concentration of Tf in PS. Then, the values obtained by the AOT were categorized into two groups: native DNA and denatured DNA, each of which was compared with the concentration of TTf in SP, and the non-parametric Mann-Whitney test was applied. It is concluded that there are significant differences in the median value of TTf in PS in the samples with native DNA and denatured DNA (P = 0.045). In future works, we intend to analyze the relationship between the studied samples, whether they are healthy controls and/or patients who consult for infertility, and the variables evaluated in this work, native DNA, denatured DNA, and concentration of TTf in PS.

We believe it is necessary to increase the number of samples in future studies, to get closer to more accurate conclusions about the relationship between TfT, a promising biomarker of male infertility, and the integrity of sperm DNA, a factor that is becoming increasingly important in the diagnosis of human infertility.

A3

## TREATMENT FOR 10 DAYS WITH THE EXTRACT ENRICHED IN PROANTOCIANIDINS FROM *Ligaria cuneifolia (PLc)* ON THE PATHWAY OF METABOLIZATION AND HEPATIC EXCRETION OF CHOLESTEROL AND GLUCOSE METABOLIZATION IN WISTAR RATS FED A HYPERLIPEMIC DIET

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In folk medicine, Ligaria cuneifolia is used to increase blood fluidity by lowering plasma cholesterol (Cho). Previous results showed that a proanthocyanidin-enriched fraction (PLc) led to decreased levels of plasmatic Cho in rats fed a diet enriched in Cho. Treatment for 3, 7, and 10 days with PLc produced a decrease in Cho levels associated with an increase in biliary excretion of bile salts and bile flow. This work aimed to analyze the effect during 10 days of the PLc-enriched fractions on the pathway of metabolization and hepatic excretion of Cho and glucose (Glu) metabolization, in Wistar rats fed a hyperlipidemic diet. We used Wistar rats fed with HFD (standard diet added with 40% of the first bovine juice) for 28 days, then injected i.p. every 24 h for 10 days with physiologican solution (HFD; N = 6) or with PLc 3 mg/100 g body weight (T; N = 6). On the eleventh day, rats were anesthetized with ketamine/xylazine (100 mg/kg/3 mg/kg, i.p.); blood was obtained by cardiac puncture. In plasma, Cho (total, HDL, and LDL) and triglycerides (TG) were determined by enzymatic methods and Glu with the Accu-Check® Glucometer measuring equipment (Roche). In bile, bile flow (BF) was determined by gravimetry, and bile salts biliary excretion (EBS) was calculated (Concentration of bile salts x BF). The Student's t-test was applied for unpaired data. Results are expressed as mean ± SE. Plasmatic Cho (% mg): C: 168.00 ± 6.63, T: 100.11 ± 4.91 \*; ChoHDL: C: 32.20 ± 1.46, T:  $28.00 \pm 2.39$  NS; ChoLDL: C:  $22.60 \pm 1.07$ , T:  $20.00 \pm 1.38$  NS; TG: C:  $191.80 \pm 21.45$ , T:  $133.00 \pm 9.68$ ; Glu (mg/dL) C: 197.5 ± 4.04 T: 159.2 ± 12.09 \*; BF ( $\mu$ L/min.g of liver): C: 2.31 ± 0.05, T: 2.91 ± 0.06 \*; EBS (nmol/min. g liver): C: 34.15 ± 3.66, T: 54.50 ± 6.50 \* (\*P < 0.05 and NS: not significant compared to C). The results showed a lipid-lowering and hypoglycemic effect of PLc in rats fed HFD. The decrease in plasma Cho could be due to increased biliary excretion of bile salts (the main compound of hepatic Cho metabolism). It is proposed to characterize the mechanism of action that produces a decrease in glucose.

A4

## TREATMENT FOR 7 DAYS WITH THE EXTRACT ENRICHED IN PROANTOCIANIDINS FROM *Ligaria cuneifolia (PLc)* ON THE CELLULAR FACTORS THAT INTERACT WITH THE KINETICS OF ERYTHROCITARY AGGREGATION IN BLOOD OF HIGH-FAT DIET WISTAR RATS

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In folk medicine, *Ligaria cuneifolia* is used to increase blood fluidity by lowering plasma cholesterol (Cho). Previous results showed that a proanthocyanidin-enriched fraction (PLc) led to decreased levels of plasmatic Cho and triglycerides (TG) in rats fed with a high-fat diet (HFD). Our objective was to analyze the effect of administration of the PLc for 7 days to hyperlipidemic rats, the blood flow at low speeds, estimated by the kinetics of the erythrocyte aggregation, characterizing cellular factors. We used Wistar rats fed with HFD (standard diet added with 40% of the first bovine juice) for 28 days, then injected i.p. every 24 h for 7 days with: physiologican solution (HFD; N = 6) or with PLc 3 mg/100 g body weight (T7; N = 6). On the eighth day, rats were anesthetized with ketamine/xylazine (100 mg/kg/3mg/kg, i.p.); blood was obtained by cardiac puncture. In serum, we determined total Cho and TG by using enzymatic methods. In blood, we evaluated the kinetics of erythrocyte aggregation (EA) using an optic method obtaining two parameters that estimate: size of aggregates (T) and speed of aggregation (V). Cellular factors: it was calculated the distinction of Bessis. The number of cells observed was 150 for every aliquot. Then the MI was calculated with the formula:  $\Sigma$  (shape index "x" number of cells / total number of cells); the Rigidity Index: (RI),

which is the inverse of erythrocyte deformability, was determined with a filtration method, by nucleopore membranes. The results are expressed as mean  $\pm$  SD. Cho (% mg): HFD: 145.3  $\pm$  8.5 T7: 62.9  $\pm$  3.2\*\*; CoHDL: HFD: 25.0  $\pm$  1.3 T7: 29.5  $\pm$  2.3(NS); CoLDL: HFD: 28.9  $\pm$  2.4 T7: 21.4  $\pm$  2.2\*; TG: HFD: 360.1  $\pm$  27.5 T7: 71.4  $\pm$  11.0\*\*. Blood: MI: HFD: -1.450  $\pm$  0.5679 T: -1.267  $\pm$  0.2204 (NS); RI: HFD: 6.37  $\pm$  0.47 T7: 6.52  $\pm$  0.23 (NS); EA: HFD: 2.042  $\pm$  0.02 T7: 1.163  $\pm$  0.21\*; V: HFD: 0.110  $\pm$  0.003 T7: 0.005  $\pm$  0.002\*\* (\**P* < 0.05; \*\**P* < 0.001 and NS: non-significant *vs.* HFD; Student's *t*-test for unpaired data). Treatment with PLc shows a lipid-lowering effect, with no changes either in shape (MI) or stiffness (RI), lowering the speed of erythrocyte aggregation. We have obtained a fraction of Lc that decreases Cho and TG in serum, improving blood flow at low-speed flow estimated by EA.

A5

## COMPREHENSIVE ANALYSIS OF THE INSERTION DIFFICULTIES OF STUDENTS IN HISTOLOGY AND EMBRYOLOGY AT THE UNR FACULTY OF DENTISTRY, IN TIMES OF PANDEMIC

<u>Obelli J</u>, Ruiz A, Davini A, Paoleti Z, Gastaldo C, García Montero M, Iglesias M, Donzelli M, Vicente Galán G, Manzano V, Esquivel L, Alegre A

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With the appearance of the COVID 19 pandemic, a suspension of face-to-face activities in the university educational field was established in Argentina in March 2020. The teaching-learning process (EA) in the basic area of dental careers became virtual. In this context of adaptation to the virtual EA process, a diversity of problems arose that conditioned its normal development. In addition to these difficulties for the students, there is scarce academic training for the new entrants of this year who have completed their secondary-level studies in 2020. The objective of this work was to identify, interpret, analyze and diagnose the integral situation of the student population in the context of a pandemic, to know the socio-demographic characteristics, academic backgrounds, different technological applications that they use, computer, technological and socio-affective problems; willingness to maintain virtuality; teacher performance. A descriptive, cross-sectional, observational, retrospective, and qualitative-quantitative study was developed. A mixed semi-structured survey using a Google form, 245 students studying Histology and Embryology, the first year of the Dentistry career of the UNR years 2020 and 2021, voluntary and anonymous, with multiple single answer options, with an expected non-response rate of less than 25%. It was available for 10 calendar days. The modality adopted was multiple choice of a single answer. An analytical comparison was made with a study carried out with similar characteristics for students in the 2020 cycle. The most obvious difficulties manifested in this analytical study are based on socio-demographic problems, with the majority of the students residing outside the city of Rosario. Socio-economically, their academic performance has seriously worsened, because in many situations they are family support. In the previous training of the middle school, it was evidenced that the virtuality of the closing of the cycle was deficient in the pre-university preparation. In relation to the socio-affective, a feeling of dissatisfaction and anguish prevails. Furthermore, technological inaccessibility in many situations acted unfavorably. In comparison, the problems maintain a similarity, but in 2021 there is a marked increase in the academic dropout rate, as well as a decrease in academic performance.

#### A6

## NEUROTOXICITY OF GEMCITABINE / DCA / LOSARTAN TREATMENT IN PATIENTS WITH PANCREATIC CANCER

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Pancreatic cancer (PANC) is one of the deadliest cancers. 'Repositioning of drugs in oncology (ReDO)' refers to the identification and development of new uses of existing drugs for the treatment of cancer. Gemcitabine (GEM) is a cytotoxic agent used in PANC. Dichloroacetic acid (DCA) proved effective in many solid tumors, by modifying cell metabolism, affecting the glycolysis pathway. Since the metabolism of solid tumors is based on aerobic glycolysis, DCA increases oxygen consumption through the expression of HIF-1 $\alpha$ . Losartan (LOS) is an angiotensin II receptor 1 antagonist, showing efficacy in the treatment of cancer by reducing the collagen of the tumor stroma, improving the arrival of drugs to the tumor; also, it inhibits metastasis and angiogenesis. A single-arm, non-randomized, openlabel, phase II clinical trial was developed as a 1st-line treatment for patients with locally advanced, inoperable, or metastatic pancreatic cancer with GEM, DCA, and LOS. The objective of this work was to analyze the adverse effects of the treatment (safety, toxicity, and tolerability) and efficacy by RECIST criteria. Eligibility criteria: patients 21–80 years old, locally advanced, inoperable, or metastatic adenocarcinoma of the pancreas without previous treatment, at least one lesion according to RECIST criteria, ECOG 0–2 scale, adequate renal and hematological function, normal calcium, informed consent. Treatment: GEM 1000 mg/m<sup>2</sup> IV weekly for 7 weeks, then on days 1, 8, 15 every 28 days

+ DCA 5 mg/kg for 21 days, then 7.5 mg/kg bod with adjuvant thiamine + LOS: 50 mg bod Treatment was administered until toxicity or progression. The study was closed early because the primary safety and efficacy endpoint was not met with the study schedule. The 4 patients included in the study presented neurological toxicity. One of them is irreversible, 3 reversible. The main neurological adverse events were ataxia, hypersomnolence, grade 3 peripheral neuropathy, grade 2 tremor in the upper limbs. As another frequent adverse event, patients showed grade 3 asthenia. Three patients presented disease progression, 1 continued in the study without receiving DCA. Neurological toxicity deepened after increasing the dose of DCA. Three of the 4 patients developed rapid disease progression. In our experience, the combination of drugs administered did not allow us to meet the primary safety and efficacy objectives, leading to an early closure of the study Although the short number of patients avoid obtaining definitive conclusions, these results suggest the hypothesis of an increase in the neurological toxicity of DCA when is associated with Gemcitabine and Losartan, something to be investigated.

## A7

## PROCESS OF ACCOMPANYING A GROUP OF RURAL AND PEASANT WOMEN IN CAMPO HARDY

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This work is the result of an Extension Project of the National University of Rosario executed between the years 2018/2021. The social actors involved are a group of "Rural and Peasant Women" from the town of Campo Hardy at the north of Santa Fe state. They fly their flag and in an associative way look for productive results. They are aware of the socio-political situation and try to make visible the difficult situation in which they find themselves. They do not give up but also beyond the productive (poultry-garden) they carry out ventures such as social lunchroom for children of the area, in order to reinforce school feeding. The group is made up of ten women. The purpose of this work is to make visible the support provided through analyzing the problems and establishing lines of action in the short and long term. The characterization of the group and the diagnosis of the situation were carried out in previous projects. The analysis of the problems was raised with a mixed approach approached as a case study. Both quantitative and qualitative information were collected through data collection instruments. The data were collected in informal spaces by direct observations and/or dialogue with social actors. The main productive problems that emerged from the analysis were: mortality in breeding, inadequate preparation of facilities, difficulties in water access, and difficulties in egg marketing. The main problem is the cost of balanced nutrition. The result analysis was based on the problems covered in the workshops, not only in the territory before Covid 19, but also virtually nowadays. Each farmer built her own chicken coop with recycled elements, which adds value to elements that do not have it. The raw material with which they can formulate their own balanced food was analyzed. Advantages are taken from vegetable crops, and they dabbled in larval rearing. The INTA Agency processed a drilling to improve access to water. They also worked in the egg marketing chain. Production is not their only goal. They also go for a vegetable garden, social lunchroom, and so on. The analysis of these types of complex realities often exceeds the timeframe of a project and the issue that was proposed to approach in the first instance. This happens because other problems of a different nature arise such as social, health, or gender issues.

## **A8**

## KNOWLEDGE BUILDING ON PEEK DENTAL IMPLANTS. YOUR RELATIONSHIP WITH STUDY HABITS

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During the construction of knowledge, the practice of correct study habits is able to be a path that leads to meaningful learning, through which new knowledge is related in a non-arbitrary and substantive way to the cognitive structure of the student apprentices. In the university environment, the training of professionals requires constant monitoring of mechanisms that promote the correct process of teaching and learning, with good study routines being one of the axes to take into account to guarantee academic success. In the field of health, as in Dentistry, the knowledge incorporated in undergraduate training constitutes a fundamental instrument that allows the professional to obtain skills and abilities, whose application can find a solution to the requirements of health problems in the community. This work proposes to carry out a survey of study practices and their relationship with academic success, in apprentices who are studying subjects whose contents include aspects of professional practice such as advances in implantology. In this scenario, permanent research attempts to improve the physical and chemical properties of dental implants by analyzing the behavior of materials that can improve the success of treatment, such as polyetheretherketone (PEEK). We consider of relevant importance the transfer of said research to students. It is proposed to explore the degree of knowledge construction according to study habits (HE) in dental degree students,

in a learning instance on the subject, plausible materials to be used in the development of dental implants, emphasizing PEEK. A descriptive, cross-sectional study was carried out on a sample represented by 64 students studying Dentistry at the UNLP, who were invited to answer the items corresponding to scale II of Pozar's Study Habits Form. Subsequently, a learning instance was developed with the same students, with an evaluation whose qualifications were expressed in three categories: Disapproved (D), regular (R), and promoted (P). The topic addressed consisted of "implications of new technologies and their impact on the physical and chemical properties of the structures made of dental implants with PEEK". At the end of the meeting, the results of study planning habits were linked with the grades obtained. The analysis of the construction of knowledge according to the study habits of the participants during the learning instance yielded the following data: HE Excellent (6): R 2 and P 4; HE Good (23) R 16 and P 7; Normal HE (31), R 26 and P 5; HE Mal (4), R 6 and P 2. It can be concluded that among the participants, the learners with better study planning habits have shown a better learning performance on implantology.

## A9

## CHARACTERIZATION OF HOUSES WITH THREE OR MORE DOGS IN THE CITY OF CASILDA

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The growth of the population of companion animals continues to be a matter of study and observation worldwide due to the social, economic, and health impact it entails. To characterize the socio-environmental conditions of people who live with three or more dogs in the city of Casilda, in 2018, the chairs of Epidemiology, Public Health and Ethics and Veterinary Legislation of the Faculty of Veterinary Sciences (FCV), National University of Rosario (UNR), conducted a canine census in the city. The census explicitly focused on households with 3 or more dogs. The houses were characterized according to the quality of their construction materials (CALMAT). CALMAT I and II were identified as V1 (safer), whereas CALMAT III and IV were deemed as V2 (more precarious). The census also enabled probing of the link with access to the drinking water network, the number of residents, and the free exit to the public highway for dogs. Data analysis was carried out using relative frequencies and 95% confidence intervals (95% CI). Inferences are the result of bivariate analysis using the Chi-square test or Fisher's test, as appropriate (P < 0.05 for significance). The total of households with three or more dogs was 98. It was identified that 58.2% (47.8-68.1%) of the dwellings are V1 and 41.8% (31.9-52.2 %) are V2. Access to the drinking water network showed a significant association with V1 (P < 0.01). In turn, 39% (24.2–55.5 %) of the V2 did not have a connection to the drinking water network. Moreover, the V2s were associated with several households of 5 or more inhabitants (P < 0.01). From the analysis of the results, we found that in those households with three or more dogs and a precarious housing condition, an association is observed between greater numbers of residents and a high proportion of households without drinking water service. Finally, the dogs belonging to V2 showed a strong association with free access to public roads (P <0.01). Also, there is a strong association with dogs that reside in these homes that freely roam public roads. Reflection on health should include the material living conditions of people and dogs.

### A10

## EFFECT OF THE ADDITION OF CITRIC ACID IN THE FEED OF LAYING HENS ON EGG WHITE FOAMS

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In poultry production, as an alternative to the use of antibiotics, organic acids are usually used as additives in the feed of laying hens to obtain a lower incidence of diseases and better-productive yields. Despite these possible benefits, its effect on the techno-functional properties of eggs is not yet known. The aim of this work was to evaluate the effect of the addition of citric acid in the feed of laying hens on the capacity of formation and stabilization of foams of egg whites. Two groups of hens of 250 each were studied: treated (GT) and control (GC), both with identical sanitary, handling, and feeding conditions, except for the addition of 0.035% citric acid in the diet of the birds of the GT, for 3 months. Random samplings were carried out and 15 eggs were removed from each group to analyze their properties. The thickness of the eggshell, the pH, and the conductivity of the whites were determined by direct measurement. The apparent viscosity was determined at 200 rpm. The foams were obtained by placing 10 mL of white in a test tube and shaking it with a mini-vat for 1 min. Foaming capacity was determined as the quotient between the volume of the newly formed foam and the volume of the initial white. The stability of the foams was analyzed by measuring the amount of liquid drained as a function of time. The size of the bubbles was analyzed by digital analysis of images of two-dimensional foam micrographs. Both the pH and the conductivity were higher for the whites of the GT, indicating

that the ionic force is greater in these samples and suggesting a greater permeability of the eggshell, which coincided with an average decrease of 2% in its thickness. No significant differences were found in the viscosity of both types of white. The foaming capacity varied between 5.0 and 7.5, being higher for the whites of the GT. The foams formed by the whites of the GT were significantly less stable since no drained liquid was detected for the foams of the GC until 45 min, while for the same time the percentage of drained liquid represented 70% for the whites of the GT. The bubble size distribution was similar for both foams at the beginning, but the sizes were higher for GT after 1 h of incubation at a room temperature of 24°C. It is concluded that the stability of the foams obtained by beating the egg whites with the addition of 0.035% citric acid in the diets of laying hens is lower compared to those that do not have this additive. Since the incorporation of acidifiers in laying hens is beneficial, alternatives will be evaluated in the future to compensate for the undesirable effects. Among these, the use of potassium bitartrate at the time of foam formation is considered.

#### A11

## HEMORHEOLOGICAL EFFECT OF B-SITOSTEROL IN A MODEL OF *IN VITRO* HYPERGLYCEMIA

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Diabetes mellitus represents a syndrome of multiple etiology, characterized by chronic hyperglycemia, due to an alteration in the metabolism of carbohydrates, lipids, and proteins. Hemorheological abnormalities have been found in diabetic patients aggravated by hypercholesterolemia involving cardiovascular risk. The objective of the present work was to determine the action of  $\beta$ -Sitosterol (BS) on human (RBC) and glycated (gRBC) red blood cells using an in vitro hyperglycemia model. For this, a solution of BS 40 mM (β-Sitosterol Sigma Aldrich S1270, 95% purity) was prepared, previously dissolved in benzyl alcohol/ $H_{2\Omega}$  (71 mg/8 mL), and then diluted in phosphate-buffered saline (pH 7.4 and 300 mOsm). RBC obtained by venipuncture of 5 healthy donors were used, which were washed and then incubated at 37°C for 2 h in equal volumes with 0.4 g/dL glucose solution, which corresponds to a blood glucose level of 200 mg/dL measured with the Accu-Chek® device. The gRBC were incubated with the BS solution and the controls at 37°C for 1 h. The samples were suspended in autologous plasma at 0.3% to get digital microscopic images by quintuplicate, obtaining the percentage of isolated cells (CA) and the coefficient of isolated cells (CcA). Values of deformability index (ID), elastic modulus ( $\mu$ ), and membrane surface viscosity ( $\eta_m$ ) from treated erythrocytes and controls were obtained with an Erythrocyte Rheometer. The aggregation index (IA) and the time to reach half of the total aggregation (t1/2) were determined using the Optical Chip Aggregometer. No significant alterations were observed in ID and  $\mu$ . However, significant alterations were observed in  $\eta_m$  (P < 0.05) and the aggregation parameters (P < 0.005). These results suggest that BS interacts with the lipid bilayer and the glycocalyx of the erythrocyte membrane, both in RBC and gRBC. Furthermore, the increase in CA and the negative values of C<sub>CA</sub> from treated samples suggest an antiplatelet activity of BS. This preliminary work presents an alternative for future research for the possible use of BS as an adjunct in diabetes treatment in injectable pharmaceutical form.

#### A12

## HEMORHEOLOGICAL STUDY OF IN VITRO ACTIVITY OF AQUEOUS EXTRACTS OF Phyllanthus sellowianus IN GLYCATED RED BLOOD CELLS Mascaro Grosso H<sup>1</sup>, Riquelme B<sup>1,2,3</sup>

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The 'white sarandi' (*Phyllanthus sellowianus*) is a shrub blooming in spring and fructifying in summer, which is traditionally used for diabetes treatment, and it is considered to have antiviral properties. This work presents the study of the action of 'white sarandi' extracts on the erythrocyte adhesion under hyperglycemic conditions. The extracts were obtained by different extraction methods (infusion and cooking) from bark and leaves. Fresh red blood cells (RBC) were separated by centrifugation from the anticoagulated blood of healthy donors. Then, an aliquot of RBCs was incubated in equal volumes with 0.4 g/dL glucose solution in PBS (pH 7.4, 300 mOsm) to *in vitro* simulate a hyperglycemia corresponding to the blood glucose level in a diabetic patient of 200 mg/dL. The obtained glycated RBC (gRBC) and control (non-glycated RBC) were incubated with infusion (PI) and cooking (PC) of *P. sellowianus* for two hours with controlled agitation at 37°C. The following kinetics aggregation parameters of erythrocytes were obtained using the Optical Chip Aggregometer: amplitude to 100 s (Amp 100), half aggregation time (t<sub>1/2</sub>), and aggregation index (AI). Erythrocytes treated with both *P. sellowianus* extracts (RBC+PC, RBC+PI) show t<sub>1/2</sub> values significantly lower value than the control sample. However, glycated erythrocytes treated with both extracts

(gRBC+PC, gRBC+PI) show a decrease in amplitude and an increase in  $t_{1/2}$ . These results show an aggregation decrease, and the values were close to the control sample. This work suggests that the phytochemicals present in *P*. *sellowianus* extracts influence the aggregation of human red blood cells. Moreover, these extracts in RBC incubated with glucose would reverse the effect of glycation, but further studies are required to determine the optimal concentrations for this to occur. These results will contribute to a better understanding of the mechanisms of action of *P. sellowianus* extracts for diabetes treatment.

A13

## ANALYSIS OF THE RELATIONSHIP BETWEEN LIPID PROFILE, CHOLESTEROL OF THE ERYTHROCYTE MEMBRANE AND CELL FORM OF THE ERYTHROCYTE IN STUDENTS OF MEDICINE WITH NORMAL WEIGHT, OVERWEIGHT OR OBESITY

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Current environmental and socioeconomic changes have been promoting unhealthy lifestyles, which favor the genesis of chronic non-communicable diseases. The objective of this study was to analyze the relationship between lipid profile (LP), erythrocyte membrane cholesterol content (EMCc) and erythrocyte cell morphology in students of the Faculty of Medical Sciences, with normal weight (NW), overweight (OW), or obesity (OB). Those who did not sign the informed consent, had pre-existing diseases, pregnancy or suspected it, were under drug treatment, or were underweight for their age were excluded. Total cholesterol (TCol), HDL cholesterol (HDLc), LDL (LDLc), and triglycerides (TG) were determined. The erythrocyte morphology was evaluated by optical microscopy, according to the Bessis technique an average morphological index (MI) was calculated. EMCc was determined by extracting membrane lipids from lysed red blood cells with 2-propanol and chloroform. The results obtained for the different determinations were statistically analyzed using one-way ANOVA with MedCalc Software. Difference was considered significant at P < 0.05. The results obtained are expressed as mean  $\pm$  standard deviation. TCol (mg/dL) NW 158  $\pm$  23; OW 159  $\pm$  25 and OB 159  $\pm$  32. HDLc (mg/dL) NW 58  $\pm$  17, OW 52  $\pm$  13 and OB 42  $\pm$  10. LDLc (mg/dL) NW 93-23; OW 94  $\pm$  17 and OB 99  $\pm$  35. TG (mg/dL) NW 92  $\pm$  31, OW 95  $\pm$  24 and OB 103  $\pm$  46. A significant difference was found only between the HDLc levels of NW and OB. EMCc (g/L) NW 0.56  $\pm$  0.15; OW  $0.62 \pm 0.12$  and OB  $0.83 \pm 0.34$ ; with significant difference between OB and the OW and NW groups, but not between OW and NW. MI NW  $-0.20 \pm 0.68$ ; OW  $-0.60 \pm 0.48$  and OB  $-0.76 \pm 0.31$ . No significant differences between the groups. We can conclude that an increase in EMCc is observed in the OB group, which could be attributed despite all normal LP, to the lower concentration of HDLc in the OB group. This could also explain the greater tendency the MI to stomatocyte shapes in this group. We intend to continue studying what has been observed in association with new hemorheological variables (for example, erythrocyte deformability) and to increase the number of participants.

A14

## STUDY OF ERYTHROCYTE DEFORMABILITY AND RELATIVE BLOOD VISCOSITY IN MEDICAL STUDENTS OF THE NATIONAL UNIVERSITY OF ROSARIO WITH NORMAL WEIGHT, OVERWEIGHT AND OBESITY IN RELATION TO THE DEGREE OF PHYSICAL ACTIVITY

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Overweight (OW) and obesity (OB) are considered states responsible for the development of chronic noncommunicable diseases. The objective of the study was to evaluate the erythrocyte deformability (ED) and the relative viscosity (RV) in students of both sexes between 18 and 25 years old, categorized by body mass indexes (BMI) according to the WHO in: normal weight (NW), OW and OB, and by the degree of physical activity according to the METs scale in: <600 low (LI), 600–1500 = medium (MI) and >1500 = high intensity (HI). Those who did not sign the informed consent, had pre-existing diseases, pregnancy or low weight, were excluded. Hematological parameters were measured in a SYSMEX KX-21N counter. The red blood cells (RBC) were suspended in saline solution at 10% hematocrit (HCT) and then filtered through polycarbonate membrane with 5-µm pores. The filtration Index (FI) =  $(t_s-t_0/t_0) \times (100 / \text{HCT})$ : inverse of ED.  $t_s$  is the passage time of the suspension.  $t_0$  is the passage time of the buffer without RBC. We measured blood viscosity (BV) and plasma viscosity (PV) with a Wells-Brookfield viscometer at 230 s<sup>-1</sup>, at 37°C. relative viscosity was calculated as RV = BV / PV and corrected to 40% HCT. The level of physical activity was established according to the World Confederation for Physical Therapy. Data were analyzed with Medcalc Software applying one-way ANOVA test, with P < 0.05. The results are expressed as mean  $\pm$  standard deviation. 49 students participated: 29NW, 12OW and 8OB. The FI was  $10.64 \pm 1.58$  (NW);  $16.14 \pm 4.76$  (OW) and 19.49  $\pm$  5.08 (OB); with significant difference between all groups. The RV was  $3.13 \pm 0.41$  (NW);  $3.51 \pm 0.43$  (OW) and  $3.75 \pm 0.63$  (OB); with significant difference of the groups OB and OW with respect to NW. According to the level of physical activity, the FI was  $16.26 \pm 5.81$  (LI);  $13.34 \pm 3.21$  (MI) and  $11.61 \pm 3.21$  (HI) with significant difference of the groups HI and MI with respect to LI. The RV was  $3.67 \pm 0.55$  (LI);  $3.14 \pm 0.40$  (MI) and  $3.15 \pm 0.38$  (HI) with significant difference of the groups HI and MI with respect to LI. Spearman's correlation coefficient showed a weak negative correlation between BMI and METs. Based on the results obtained, we can conclude that students with OW or OB and LI physical activity have a higher RV and a lower EF than the NW. These findings allow us to infer that in the states of OW and OB, changes would occur at the level of the cellular component that would be responsible for the hemorheological alterations in these groups, and also that LI would have an importance in this difference. The low correlation between BMI and METs suggests that these variables could influence independently, so we will be going to consider the incorporation of new intervening variables for the continuation of this study.

#### A15

## KINETICS OF RUMINAL DEGRADATION OF LEAVES

AND PODS OF Gleditsia triacanthos L.

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Trees provide a greater quantity and diversity of goods and services to the ecosystem compared to herbaceous plant species. Gleditsia triacanthos L., a tree of the Fabaceae family, is a species native to central western North America, naturalized in regions of Argentina. Its leaves and pods are very palatable and consumed by sheep, goats and cattle, they also have high levels of protein and are free of potentially toxic compounds. Although there is much information on the chemical composition and dynamics of the digestion of herbaceous forages consumed by ruminants, it is scarce for arboreal and shrubs, which motivates their study. The objective was to describe and compare the ruminal degradation kinetics in sacco of leaves and pods of Gleditsia triacanthos L (GT) taken in a period of annual growth. The samples were taken from 15 trees located in the Agrotechnical School-Faculty of Veterinarians Science Complex of the UNR, Casilda, Santa Fe, Argentina. Were obtained 8 samples composed of subsamples of each tree, 6 corresponding to leaves collected once a month from November to April and two to mature pods harvested in June and July. The immediately extracted samples were dried at 60°C, 48 h, ground and sieved (2 mm) and the concentration of dry matter (DM) was determined. Kinetics of ruminal degradation in sacco of DM (DMRD) was determined. ASTM 230 nylon bags (pore size: 62 µm) were incubated with 3 g of DM (17 mg DM/cm<sup>2</sup>) of each sample for 0, 3, 6, 12, 24, and 48 h, for two periods, in the rumen of three sheep of the Pampinta breed provided with a ruminal cannula kept stabled with access to water and alfalfa hay. Immediately after rumen removals, were washed with fresh water, dried at 60°C for 48 h and weighed to obtain the percentage of DM degradation. The DMRD data were fitted to Orskov & McDonald (1979): DMRD  $\% = a + b (1 - e^{-ct})$ , where a is the soluble or rapidly degradable fraction, b the slowly degradable fraction, c the degradation rate and a + b the potentially degradable fraction in rumen. The data were studied by ANDEVA and Tukey's Test (P > 0.05). The average DM (SD) of leaves was 38.7 (6.16) and of pods 98.8 (0.04) %. In the leaves and pods, fraction a was 20.3 and 40.1; fraction b, 55.7 and 32.9; a + b fraction, 76 and 73%; and the degradation rate (c) 0.0384 and 0.0959% / h, respectively. DM concentration of the leaves was higher than that of cultivated herbaceous forage species. The high level of DM of the pods is similar to that described for concentrated foods. The leaves presented lower soluble fraction and degradation rate and higher slowly and potentially degradable fractions than pods. Both the leaves and the pods of GT had a potential degradability that was greater than 70%, therefore they can be considered food resources with high ruminal degradability.

#### A16

## DEGRADABILITY AND GAS PRODUCTION IN RUMEN OF TREES AND BUSHES OF ISLANDS OF THE DELTA OF PARANÁ IN FRONT OF ROSARIO CITY

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The objective was to describe and compare the degradability and gas production in the rumen of arboreal and shrub leaves consumed by ruminants in the islands of the Upper Delta of the Paraná River in front of the City of Rosario. The plant species used were: (AC) *Vachellia caven*, (GT) *Gleditsia triacanthos*, (SH) *Salix humboldtiana*, (TI) *Tessaria integrifolia*, (BS) *Baccharis salicifolia*, and (LA) *Lippia alba*. The leaf samples were obtained on the islands at km 430 of the Paraná River, in front of Rosario city, dried at 60°C, ground and sieved (2 mm). With the samples of each plant species, a composite sample was made. The ruminal dry matter degradability (RDMD) was determined

for each plant species at 0, 3, 6, 12, 24, and 48 h of incubation in rumen in sacco, in two female Pampinta breed sheep provided with a ruminal cannula, fed with alfalfa hay, for three periods (Mehrez and Orskov, 1977). To determine the gas production, in vitro incubation systems were used, consisting of 50 cc glass bottles, closed with a rubber stopper, connected to 20-mL glass syringes, loaded with ruminal fluid (RF) from the aforementioned sheep. (Smacchia, 1995). The RF was obtained 2 h after the start of ingestion, filtered with a 2 mm mesh. The systems were loaded with 30 mL of the RF and 300 mg of samples of the plant species (AC, GT, SH, TI, BS and LA), kept at 39°C for 24 h in a water bath and manually shaken at regular intervals. Systems loaded with LR alone were used as a control. Three incubations were made, and each treatment was done in triplicate. The mL of gas produced (1, 3, 6, 12, and 24 h) were measured by displacement of the syringe plunger. The RDMD and gas production data were adjusted to the Orskov and McDonald (1979)  $Y = a + b (1 - e^{-ct})$ . The adjustment to the proposed model was adequate, with R<sup>2</sup> between 92 and 99 in RDMD and between 94 and 99% in gas production. The ruminal degradability presented average values of 19.6, 56.4, and 76 %, respectively for the rapidly (a), slowly (b) and potentially (a + b) degradable fractions, and 0.05337% / h for the degradation rate (c). The average values in the gas production were 0.67 mL in the soluble fraction (a), 114.24 mL in the slowly fermentable fraction (b), and 0.0256% / h in the gas production rate (c). The %RDMD at the end of the incubation was higher in TI (96.4), followed by BS (81.4), SH (79.5) and LA (79.4), then by GT (70) and lower in AC (49.1). The highest gas production (mL) at 24 h was presented by LA (36.9), then with similar values BS (33.6), SH (32.1), GT (27.7), followed by AC (23.5) and lower gas production in IT (17.6). In general, the samples had a high level of ruminal degradability, with the exception of CA, which was intermediate. Gas production in LA, BS, SH and GT exceeded that of good quality alfalfa hay. These samples showed a behavior similar to that observed in herbaceous plant species with forage potential in the same islands of the upper Delta of the Paraná River, which we studied previously.

#### A17

## PREVALENCE AND RESISTANCE OF Staphylococcus aureus IN NASAL SAMPLES IN A POPULATION OF PREGNANT WOMEN BETWEEN 35–37 WEEKS OF GESTATION

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Staphylococcus aureus (SA) is a Gram-positive coccus that may be part of skin and mucous membranes microbiota in approximately 25% of healthy people (carriers). From these locations, it can spread to other sites of the human body, producing pathologies of varying severity (skin and soft tissues infections, osteomyelitis, genitourinary system, septicemia, etc.). This microorganism can be transmitted through person-to-person contact or by exposure to contaminated fomites. This, together with the existence of multidrug-resistant strains to commonly used antimicrobials, makes it necessary to know both its prevalence in the community and its antimicrobial sensitivity pattern as widely as possible. Transmission from a carrier mother to her non-colonized child can occur vertically (during passage through the birth canal) and horizontally (through nasal carriage) and can cause perinatal infections. The purpose of our study was to know the prevalence of nasal colonization by SA in pregnant women between 35-37 weeks of gestation and its sensitivity pattern to antimicrobials. We obtained 31 samples from nostrils of pregnant women using cotton's swabs after they signed an informed consent. These samples were seeded into Salted Mannitol Agar (Britania®) and incubated at 35°C for 24-48 h. Suspicious SA colonies were identified by conventional biochemical tests (catalase, coagulase, and DNase test). Antibacterial sensitivity to various firth line antibiotics (erythromycin, clindamycin, gentamicin, levofloxacin, cefoxitin, and trimethoprim-sulfamethoxazole) was assessed by the Kirby-Bauer diffusion method, according to Clinical and Laboratory Standards Institute (CLSI) protocols. Of the 31 pregnant women studied, 23 (74.19%) were found to be colonized with SA. Of the 23 isolates, 20 (85.71%) turned out to be sensitive to methicillin (SAMS), and 3 (14.28%) turned out to be resistant (MRSA). Two of these 3 isolates showed resistance to other antimicrobials. A high percentage of isolates with resistance to erythromycin (34.8%) and clindamycin (21.7%), so their empirical use is not recommended, as we have shown in previous studies. Levofloxacin and cotrimoxazole are still active against SA. These results are a continuation of a 4 years' investigation, and they continue showing isolates of SA in pregnant women.

## ANOVAGINAL COPORTATION OF Staphylococcus aureus AND GROUP B BETA HEMOLYTIC Streptococcus (S. agalactiae) AND ANTIMICROBIAL SENSITIVITY OF Staphylococcus aureus, IN A POPULATION OF PREGNANT WOMEN BETWEEN 35– 37 WEEKS OF GESTATION

<u>Catalano F</u>, Tavella D, Massonna C, Morello B, Bordon M, Brandolisio N, Guzman P, Fogliato S, Revelli L, Sáez B, Córdoba L, Zafra M, Hails I, Bulfoni M, Ombrella A, Ponessa A, Gambandé T

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Staphylococcus aureus (SA) has a great importance as an infectious agent, both in communitarian and nosocomial environments. It can cause from mild infections (food poisoning, superficial skin infections) to life-threatening pathologies (sepsis, endocarditis, others). Due to the uprising of antibiotic resistant isolates is important to know local epidemiology and sensibility pattern to optimize treatments. SA may be part of the normal skin and mucous membranes microbiota in approximately 25% of healthy people, without causing infection, making them "asymptomatic carriers". The possibility to transmit this microorganism from a carrier to a non-colonized subject due to close contact, as can happen from a mother to their child during the passage through the birth canal, has been reported in literature. Group B beta-hemolytic Streptococcus (EGB) usually colonizes the gastrointestinal tract; however, it can also be present in the genitourinary tract and oropharynx in about 20% of the population, according to local literature. This percentage can increase with the number of previous deliveries and sexual activity. It has been reported an association between SA and EGB colonization. The purpose of this project was to determine the prevalence of vaginal and anal SA and EGB colonization in a population of pregnant women between 35 and 37 weeks of gestation. We obtained 280 anovaginal samples by swabbing, after signing an informed consent. These were seeded in Salted Mannitol Agar (Britania®) and Todd-Hewitt broth supplemented with antibiotics, then the colonies were picked after 24 h and seeded in sheep-blood Columbia Agar. Suspicious SA colonies were identified by conventional biochemical tests (mannitol fermentation test, DNase test and coagulase test for SA; and bile-esculin test, CAMP test and/or antigen detection for EGB). Antibacterial sensitivity was assessed by the Kirby-Bauer diffusion method, according to Clinical and Laboratory Standards Institute (CLSI) protocols. Of the 280 original samples SA grew in 35 (12.5%), EGB in 50 (17.85%), and both microorganisms grew in 9 of them (3.21%). Of the 35 SA isolates, 10 (28.5%) were methicillin-resistant (SAMR) and did not present any additional resistance, so this was interpreted as community acquired methicillin-resistant SA (SAMR-CA). Even if these are preliminary results in the context of an ongoing study, we found a considerable number of SA isolations in pregnant women.

#### A19

## CENSUS OF DOGS AND CATS IN CORONEL BOGADO FOR THE PLANNING OF PUBLIC HEALTH ACTIONS

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Knowing the characteristics of the population of dogs and cats in a community facilitates planning for actions that could lead towards a better coexistence between humans and their companion animals, to better serve these animals, and protect community health. Outlining needed actions is a prominent concern among communities where this coexistence is precarious or there is an intent for improvement. This requires accurate and up-to-date information that can inform future directives. With this in mind, the inhabitants of Coronel Bogado, with the support of their commune and a veterinary medical professional who graduated from UNR, requested scientific assistance for the planning, implementation, and analysis of a census of dogs and cats. This intervention was made possible by establishing a collaboration agreement with the deliberate purpose of improving the health of populations, as an indivisible concept that integrates the species and their environment as proposed in the concept of One Health. Work began in March 2021 to identify the information that is relevant to the community. A planning group was formed with teacherresearchers from the chairs of the Epidemiology, Public Health, and Research Methodology Departments, together with a veterinary doctor, a social worker, and a group of volunteers from Coronel Bogado, to design the questionnaire. Once the object of study and associated variables were identified, the team prepared a survey using the Google form application. This instrument, inquiring on information pertaining to the sociocultural context, dogs, cats, and coexistence behaviors, was validated and pilot tested. Sixteen (16) community resident pollsters participated. A total of 271 survey responses were collected, out of a total of 328 visits made from May 4 to May 24, 2021. The responses collected (with prior informed consent) depict households in 50 blocks, which accommodate 822 adults and 180 minors. Survey responses revealed a total of 341 dogs and 96 cats, distributed in 186 and 66 households, respectively. Contacting the community house-to-house (as done for this survey) increases the perception of the relevance of the issue, as an instance of raising awareness that the individual decisions regarding keeping companion animals indeed impact the social group. The construction of a database with the collected responses will make it possible to establish

different relationships, disaggregating the level of information and linking the different variables surveyed in order to design policies based on reliable information.

#### A20

## PRELIMINARY STUDY OF HEMATOLOGICAL PARAMETERS IN THREE SPECIES OF BIRDS OF PREY FROM THE PROVINCE OF MENDOZA (ARGENTINA)

#### Zerpa $C^1$ , Ciminari $J^2$ , Pidone $CL^3$

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In the rehabilitation of birds of prey, hematological values are related to their general condition. The objective of this work was to determine the hematological parameters of black-chested buzzard-eagles (Geranoaetus melanoleucus). crested caracaras (Caracara plancus), and condors (Vultur gryphus) from Mendoza, and generate a database for the province. The study was carried out at the facilities of the Mendoza Ecopark and the S.O.S Acción Salvaje Rescue Center. Blood sampling was carried out on clinically healthy black-chested buzzard-eagles (N = 24), crested caracaras (N = 17) and condors (N = 4) from the institutions and those who entered quarantine. Blood was taken from the ulnar vein, with a tuberculin syringe and 25/8 needle, and placed in tubes with EDTA. Blood glucose values were also recorded using the Accu-Check®Performa glucometer. For hematology, the hematozimetric method was used, performed in a Neubauer chamber. 20 µL of blood was placed in a tube with 4 mL of Natt Herrick's solution and with a capillary tube it was placed in the Neubauer chamber for counting. The differential count of leukocytes was performed by observing a smear stained with staining 15 under a light microscope. For hematocrit (hto), a blood sample was placed in a microhematocrit capillary and centrifuged at 12000 rpm for 5 min. Total plasma proteins were measured with a refractometer, where a drop of plasma was placed. The values obtained from the mean  $(\bar{x})$  and the standard deviation (SD) for the different variables studied was: black-chested buzzard-eagles: hto (%):  $31.8 \pm$ 13.4; total proteins:  $4.4 \pm 1.9$ ; blood glucose (mg/dL):  $295 \pm 46.3$ ; red blood cell count (millions/mm<sup>3</sup>):  $2.38 \pm 1.05$ ; white blood cell count (number/mm<sup>3</sup>):  $7338 \pm 4410$ ; heterophiles (%):  $57 \pm 21.1$ ; heterophiles (quantity/mm<sup>3</sup>): 4320  $\pm$  3274; lymphocytes (%):28  $\pm$  20.1; lymphocytes (amount/mm<sup>3</sup>): 2437  $\pm$  2709; monocytes (%): 5  $\pm$  3.8; monocytes  $(\text{amount/mm}^3)$ :  $402 \pm 511.4$ ; eosinophils (%):  $10 \pm 8.2$ ; eosinophils (amount/mm<sup>3</sup>):  $1212 \pm 2943$ . Crested caracaras: hto (%):  $39 \pm 18.5$ ; total proteins:  $2.8 \pm 1.6$ ; blood glucosa (mg/dL):  $249 \pm 31.4$ ; red blood cell count (millions/mm<sup>3</sup>):  $2.06 \pm 0.84$ ; white blood cell count (number/mm<sup>3</sup>): 3497 \pm 2074; heterophiles (%): 51 \pm 19.6; heterophiles (quantity/mm<sup>3</sup>):  $1738 \pm 1215$ ; lymphocytes (%):  $45 \pm 19.7$ ; lymphocytes (amount/mm<sup>3</sup>):  $1607 \pm 1194$ ; monocytes (%):  $2 \pm 2.1$ ; monocytes (amount/mm<sup>3</sup>):  $67 \pm 71.6$ ; eosinophils (%):  $4 \pm 8.5$ ; eosinophils (amount/mm<sup>3</sup>):  $114 \pm 266$ . Condors: hto (%):  $39 \pm 19.5$ ; total proteins:  $4 \pm 2$ ; blood glucosa (mg/dL):  $193.5 \pm 16.2$ ; red blood cell count (millions/ mm<sup>3</sup>):  $1.89 \pm 0.80$ ; white blood cell count (number/mm<sup>3</sup>):  $4364 \pm 759$ ; heterophiles (%): $48.75 \pm 16.5$ ; heterophiles  $(quantity/mm^3)$ : 2213 ± 1103; lymphocytes (%):47 ± 15.9; lymphocytes (amount/mm^3): 1998 ± 482; monocytes (%):  $2 \pm 3.3$ ; monocytes (amount/mm<sup>3</sup>):  $84 \pm 123.1$ ; eosinophils (%):  $2 \pm 2.8$ ; eosinophils (amount/mm<sup>3</sup>):  $68 \pm 16$ . The calculations were made through the Excel program. Finally, the importance of obtaining these data is highlighted, in order to conserve these species.

## A21

## PREVALENCE OF Staphylococcus aureus IN ANOVAGINAL SAMPLES OF A GROUP OF PREGNANT WOMEN BETWEEN 35 TO 37 WEEKS OF THEIR PREGNANCY

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*Staphylococcus aureus* (SA) is a Gram-positive coccus arranged in the shape of a cluster of grapes. It could be part of the skin and mucous membranes microbiota in approximately 25% of healthy people (carrier state), from where it can spread to other parts of the organism. Although modern antibiotics have drastically reduced the severity of bacterial infections, these continue to be a relevant cause of morbidity and mortality. This fact, in addition to the existance of SA strains that resist to most commonly used antibiotics, makes it necessary to determine both its prevalence in the community and its antimicrobial sensitivity profile. Transmission from the carrier to another individual by direct contact has also been reported, this can happen from mother to child during birth and puerperium. The importance of genital colonization by SA is increasing, rising the risk of neonatal infection by vertical and horizontal transmission, potentially causing diverse infections. The aim of this study was to stablish the prevalence of vaginal and anal colonization by SA in pregnant women between 35 to 37 weeks of gestation, and to evaluate antimicrobial sensitivity. A total of 280 anovaginal samples were obtained by swabbing, after signing an informed

consent form. These were seeded on Salted Manitol Agar (Britania) and incubated at 35°C for 48 h. Suspected SA colonies were identified by conventional biochemical tests, and sensitivity to the following antimicrobials (erythromycin, clindamycin, gentamicin, levonofloxacin, and trimethoprim sulfamethoxazole) was assessed by the Kirby-Bauer diffusion method, according to Clinical and Laboratory Standards Institute (CLSI) standards. Of the 280 samples obtained, 35 (12.5%) were found to be colonized by SA. Of the latter, 25 (71.43%) were considered as SAMS (methicillin-sensitive *Staphylococcus aureus*) and 10 (28.57%) as SAMR-AC (community-acquired methicillin-resistant *Staphylococcus aureus*). We also obtained resistance to gentamicin in 6 isolates (17.4%), to erythromycin in 20 isolates (57.14%) and to clindamycin in 13 (37.14%). Given these results, the use of erythromycin and clindamycin as empirical treatment in puerperal and neonatal SA infections would not be recommended, as we have shown in our previous studies. Levofloxacin and TMS maintain their activity against SA, meaning that we can still recommend their empirical use in combination with ampicillin for the treatment of perinatal infections in our location. These results denote the increasing development of resistance to antibiotics that this microorganism has and, therefore, it motivates us to continue with this research project, considering that these are preliminary results of an ongoing research Project.

## A22

## CHARACTERIZATION OF THE SEROLOGICAL RESPONSE TO INFECTION BY Leptospira spp. IN DOMESTIC CATS IN THE SOUTH OF SANTA FE

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Leptospirosis is an infectious disease, caused by pathogenic serovars of Leptospira spp. Infection in cats would occur through contact with infected urine or preys. Prevalence studies show seropositivity rates of between 4% and 33%. The objectives were (1) to determine the seropositivity rate to Leptospira spp. and to serogroups in cats, and (2) to characterize the immune response of the population studied. Samples of 589 blood serum from domestic cats (Felis catus), of different breeds, ages, and sex, from southern Santa Fe were analyzed. The cats had indoor/outdoor habits and were hunters, some showed signs of kidney failure. Blood samples were obtained by venipuncture and sera were refrigerated at -20°C. Reference strains of Leptospira spp. were used: L. interrogans: Pomona Pomona; Icterohaemorrhagiae Copenhageni M 20, Canicola Canicola Hond Utrech IV, Australis Bratislava Jez bratislava, Pyrogenes Salinem, Sejroe Hardjo type Prajitno Hardoprajitno, Autumnalis Autumnalis Akiyami A, Bataviae Bataviae Swart; L. kirschneri: Grippotyphosa Moskva V, Cynopteri Cynopteri 3522 C; and L. borgpetersenii: Ballum Castellonis Castellón 3. The cut-off point was 1:25. A seropositivity rate of 22.75% was detected in 134 seroreactive animals. Within this group, 69 (51.49%) sera presented cross-reactions, 45 (65.21%) between Castellonis and Autumnalis, the only serovars with titers from 1:25 in cats without clinical signs. Among the remaining 24 (34.78%), the only clinical case diagnosed by analysis of paired samples stood out, with titers of 1:6400 for Pomona, 1:400 for Copenhageni, 1:50 for Bratislava. High titers for Pomona and Copenhageni were observed in paired samples. The other 65 (48.50%) reacted to a single serovar: Bratislava 19 (29.23%), Autumnalis 18 (27.69%), Castellonis 17 (26.15%), Pomona, jn 5 (7.69%), Grippotyphosa, 3 (4.61), and Copenhageni 2 (3.07%). The highest titer of 1: 200 was found for Pomona and Bratislava in cats with clinical signs. The seropositivity rate was within the expected range for the study region (endemic disease). It was observed that cats were infected mainly by serovars of the serogroups: Australis, Autumnalis, Castellonis, Pomona, Grippotyphosa, and Icterohaemorrhagiae. The high detection frequency and low titers to the Australis, Autumnalis, and Ballum serogroups, observed mainly in cats without clinical signs, suggest frequent exposure to serovars of these serogroups, perhaps due to hunting habits. The low detection frequency and high titers to Pomona and Icterohaemorrhagiae in the clinical case, suggest that these serogroups could be incidentally related to the domestic cat.

#### A23

## SUFFERING OR AGGRESSIVENESS OF DOGS FROM THE PERSPECTIVE OF NEIGHBORS OF CASILDA CITY

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Previously, we showed the association between wandering of dogs and their greater chance to traumatic injuries. Additionally, it has been reported the highest exposure to dog bites in individuals from most vulnerable urban areas. Responsible pet ownership entails, among others, the commitment to minimize potential risk of injuries or preventable pathologies, as well as those that this can generate to the community through aggressive behaviors. The general objective of this work was to describe the frequency of indicators of suffering or aggressiveness of dogs referred by inhabitants of the Casilda city and to contextualize these indicators with the housing infrastructure. Crosssectional study based on data from census of dogs in Casilda city in 2018, directed by the Chair of Epidemiology and Chair of Public Health-FCV. Information on dwelling's type was collected: they were identified as V1 -safer- and V2 -more precarious-; according to the requirements for CALMAT I and II in the first case and CALMAT III and IV in the second case; free access to public roads by animals (AVP) and the perception of the responders of indicators of animal suffering or aggressiveness in the last year, specifically: dog involved in a traffic accident (AT), malnutrition (MN), and dog biting to person (MP). Descriptive data analysis was carried out using relative frequencies and 95% confidence intervals (95% CI). Inferences were made from bivariate analysis using the Chi-square test. A P < 0.05 was considered significant. The respondents that had dogs were 419. The 72.8% (68.2–76.9%) of them had live in V1 dwellings and the 27.2% (23.1–31.8%) in V2. Dogs belonging to V2 were associated with free AVP (P <0.0001). The 52.7% (47.8–57.6%) of the respondents reported having witnessed in the year prior to the consultation at least one of the indicators of suffering or aggressiveness studied. The 24.1% (20.1–28.5%) witnessed dogs suffering from AT; 30.8% (26.4-35.5%) reported having seen dogs that impressed with MN and 28.9% (24.6-33.5%) witnessed MP dogs. Reference to dogs suffering from AT was significantly associated with V2 responders (P = 0.05); in the same way the perception of animals MN (P < 0.001) and the testimony of dog MP (P < 0.01). More than a quarter of the respondents who had dogs in their homes belonged to V2 homes, characterized by greater precariousness compared to V1 homes. This infrastructure is more conducive to free AVP and is likely to at least partially justify this situation. Responders belonging to V2 reported more perception of situations of suffering or aggressiveness of dogs than those of V1. Interdisciplinary efforts should continue to join together in order to address the problem of animal suffering or aggressiveness, trying to integrate the aspects that make up its complexity.

A24

## INTRODUCTION TO THE STUDY OF THE CONVOLVULACEAE FAMILY IN THE PROVINCE OF SANTA FE (ARGENTINA) II

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Historically, the family Convolvulaceae has been classified in the order Tubiflorales, according to its morphological features. According to the APG-IV classification, it is now placed within the order Solanales based on molecular criteria. It comprises prostrate, climber or erect herbaceous plants with alternate and simple leaves, entire or palmately-lobed. Flowers are actinomorphic, gamopetalous, pentamerous, grouped in cymose inflorescences. The fruit is a capsule with glabrous or hairy endospermic seeds. The aim of the present contribution is to continue with the taxonomic study of the family in Santa Fe. We carried out a bibliographical review and consulted national herbaria with important collections of the province (SF, SI, UNR); the fieldwork experience of the authors and lab work were used to validate the specimens' identity. The preliminary results show that the Convolvulaceae family is represented in the province by seven genera. These are: *Ipomoea L.* (treated in a previous contribution); *Aniseia* Choisy, with *A. argentina* (N.E. Br.) O'Donell; *Cressa L.*, with *C. truxillensis* Kunth; *Distimake* Raf., with *D. dissectus* (Jacq.) Simões & Staples var. *dissectus*; *Dichondra* J.R. Forst. & G. Forst. with *D. microcalyx* (Hallier f.) Fabris and *D. sericea* Sw. var. *sericea*; *Evolvulus* L. with *E. arizonicus* A. Gray and *E. sericeus* Sw. var. *sericeus*; and *Convolvulus* L., with *C. crenatifolius* Ruiz & Pav. subsp. *crenatifolius*, *C. hermanniae* L'Hér. subsp. *hermanniae*, *C. laciniatus* Desr., and the adventitious and crop weed *C. arvensis* L. A species distribution map, botanical descriptions, and illustrations for each taxon and a key for genera field recognition are provided.

#### A25

## INTRODUCTION TO THE STUDY OF THE RUBIACEAE FAMILY IN THE PROVINCE OF SANTA FE (ARGENTINA) I: TRIBE SPERMACOCEAE

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The family Rubiaceae was historically placed in the order Rubiales, but according to APG IV it is currently considered part of Gentianales. It includes trees, shrubs or herbs, generally unarmed with opposite, simple and entire leaves with stipules. Flowers, solitary or grouped in cymose inflorescences, actinomorphic, pentamerous or tetramerous; petals, with indument in adaxial face; stamens, 4–5 with filaments adnate to corolla; carpels, usually 2, ovary inferior with 1 to numerous ovules. Dry or fleshy fruits. According to the classification system, the family comprises three subfamilies: Rubioideae, Cinchonoideae, and Guettardoideae. The first one presents some tribes, among them

Spermacoceae, made up of herbaceous or shrubby plants, with stipules in the form of a fimbriated sheath, flowers tetramerous, ovary with two uniovulate locules, and fruit a schizocarp, capsule or pyxidium. The objective of this work is to initiate the taxonomic study and analyze the geographical distribution of the Rubiaceae family in Santa Fe, beginning with the Tribe Spermacoceae. The methodology consisted of bibliographic review, consultation of herbaria with important collections of the province (SF, SI, UNR), observation of most of the species in their habitat, and lab work to corroborate determinations. The preliminary results show that the tribe Spermacoceae is represented by five genera and 12 species, all herbs or suffruices. These are: *Borreria* G. Mey., with *B. dasycephala* (Cham. & Schltdl.) Bacigalupo & E.L. Cabral; *Mitracarpus* Zucc., with *M. megapotamicus* (Spreng.) Kuntze; *Galianthe* Griseb., with *G. centranthoides* (Cham. & Schltdl.) E.L. Cabral, *G. eupatorioides* (Cham. & Schltdl.) E.L. Cabral; *Richardia* L. with *R. brasiliensis* Gomes, a perennial herb and crop weed, *R. humistrata* (Cham. & Schltdl.) Steud., and *R. stellaris* (Cham. & Schltdl.) Steud.; and *Spermacoce* L. with *S. eryngioides* (Cham. & Schltdl.) Kuntze, *S. glabra* Michx., *S. hassleri* E.L. Cabral & J. Florentin, both typical of humid environments, and *S. verticillata* L., rhizomatous suffrutex with markedly tetragonous stems. Taxonomic information, illustrations, a map of geographical distribution of the species and a dichotomous key based on morphological features are presented.

#### A26

## COMORBIDITIES AND STRESSFUL LIFE EVENTS (CVA) IN ADULT WOMEN WITH HASHIMOTO'S THYROIDITIS (HT) LIVING IN SAN NICOLÁS, BUENOS AIRES

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Hashimoto's thyroiditis (HT) is considered the main cause of hypothyroidism in populations without iodine deficiency. There are few studies about this autoimmune thyroid disease in Argentina. The aim of our study was to describe socio-demographic characteristics, co-morbidities, and stressful life events (SLE) in adult women with HT. Ninety consecutive women who attended an endocrinology consultation in a health care center in San Nicolás, Argentina, during the years 2019 and 2020, were invited to participate in this cross-sectional descriptive study. For all patients, three basic techniques were used to collect the information: physical examination, review of medical records, and implementation of a standardized questionnaire. Patients stress level perception was measured by applying a visual analog scale. The average age of the studied patients was  $49.4 \pm 14.7$  (median = 52.5; range: 19– 79). At the time of conducting this study, the median time elapsed since diagnosis was 6 years. Thirty-four patients (37.8%) had a family history of thyroid disease: 93.3% of patients were hypothyroid and 6.7% were euthyroid; 62.5% have goiter. Associated autoimmune comorbidities were found in 11 patients (12.2%): rheumatoid arthritis (N = 4), celiac disease (N = 3), vitiligo (N = 2), Addison's disease (N = 1), and thrombocytopenic purpura autoimmune (N = 3)1). These 11 patients were younger than the median age. The association between a family history of thyroid disease and the presence of other autoimmune diseases was not significant (P = 0.402). The most frequent non-autoimmune comorbidities were overweight (42.7%), obesity (24.7%), skin and appendage weakness (22.2%), hypertension (22.2%), dyslipidemia (16.7%), osteopenia (11.1%), osteoporosis (5.6%), and type 2 diabetes (4.4%). In three patients, thyroid cancer was diagnosed after the date of HT diagnosis, the ages of these patients were 26, 33, and 49 years. Stress level  $\geq$  7 was reported by 84.3% of patients. The predominant SLE were the death of a close relative (37.6%), a family member with chronic illness or disability (17.6%), economic discomfort (16, 8%), and domestic violence (4.7%). No association was found between the presence of other autoimmune diseases and the two most frequent SLE (Fisher's P-value = 0.16 and 0.27, respectively). Stress and family history of thyroid disease were not determining factors in the development of other autoimmune diseases in these patients. It would be necessary to carry out analytical epidemiological studies on our population in order to evaluate this and other risk factors that could be involved.

#### A27

## BIOMARKERS OF DISEASE RELAPSE IN HIGH-RISK PEDIATRIC PATIENTS WITH SOLID TUMORS TREATED WITH METRONOMIC CHEMOTHERAPY

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Metronomic chemotherapy (MCT) consists in the chronic administration of low doses of chemotherapeutic drugs, without prolonged drug-free periods. It has demonstrated therapeutic efficacy with good tolerability and low to null toxicity. Among other effects, it can modulate the antitumor immune response. Our objective was to identify

biomarkers of the antitumor immune response as prognostic indicators of tumor relapse or progression. Nineteen pediatric cancer patients (2-18 years old) (Ewing's Sarcoma 7, Rhabdomyosarcoma 7, Osteosarcoma 3, Other 2) receiving maintenance MCT after reaching complete remission with standard chemotherapy, surgery and/or radiotherapy, were analyzed. CD4, CD8, and Tregs lymphocytes were studied by flow cytometry; soluble sPD-1 and sPD-L1 by ELISA. Patients were distributed into three groups: Good evolution (GE, N = 10): those which ended MCT without distant recurrence or progression; Bad evolution (BE, N = 5): MCT was discontinued due to relapse or progression; and *Distant Relapse* (DR, N = 4): patients that relapsed in their diseases after, at least, 6 months of successfully completing MCT. BE group was not considered for the analysis because patients did not finalize treatment. The ratio between end-of-treatment/start-of treatment values of CD4, CD8, Tregs, sPD-1, and sPD-L1 was determined for each patient and the median (range) of each group was calculated: Tregs [GE: 1.37 (0.17–7.47), DR: 5.69 (5.5–21.29)]; CD4: [GE: 1.37 (0.64–1.89), DR: 1.37 (0.19–2.16)]; CD8: [GE:0.98 (0.44–1.74), DR: 0.32 (0.23– 0.53)]; sPD-1: [GE: 1.24 (0.43-3.1), DR: 2.11 (1.52-3.08)]; sPD-L1: [GE: 0.55 (0-1.26), DR: 0.72 (0.66-0.91)]. **Tregs**: GE vs. DR, P = 0.032; **CD4**: GE vs. DR, NS; **CD8**: GE vs. DR, P = 0.007; **sPD-1**: GE vs. DR, P = 0.038; sPD-L1: GE vs. DR, NS; Mann–Whitney's non-parametric t-test. We concluded that (1) high end-of-treatment values of Tregs and sPD-1, along with low values of CD8, with respect to start-of-treatment values, after successful completion of MCT would indicate a high probability of distant relapse, and that (2) confirmation of these putative biomarkers with an analysis of a higher number of patients could support the necessary basis for its further use in patients' treatment.

## A28 HORMONAL AND OSMOTIC CONTROL OF SOYBEAN EMBRYONIC AXES GERMINATION

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Under suitable incubation conditions, quiescent seeds present a typical triphasic imbibition dynamic. The onset of phase III is determined by the expansion of cells at the elongation zone (EZ) of the embryonic axis (A) that leads to germination. Expansin proteins are considered primary promoters of cell expansion. Abscisic acid is the main phytohormone that inhibits soybean germination (Glycine max. L. Merr) by restraining cell walls loosening necessary for expansion. Whereas, solutions of low water potential avoid germination limiting water uptake and consequently, the expansion ability by A. In previous works, we identified the main expansin (EXP1) responsible for the cell wall loosening of soybean A during germination. Furthermore, EXP1 was up- and down- regulated under germination inhibitory conditions in -1 MPa of polyethylene glycol 8000 (PEG) and 50 µM of abscisic acid (ABA), respectively. The present work aimed to evaluate the control by ABA and PEG on soybean A germination. Three replicates of 10 A each were incubated in distilled water (control) and 50 µM ABA, and six replicates in -1 MPa PEG, for 24 h at 27  $\pm$  1 °C and in the dark. All the replicates in ABA and three of the replicates in PEG were then transferred to distilled water (ABA-WATER, PEG-WATER) to induce germination during the next 24 h. The last three replicates of A in PEG were transferred to ABA (PEG-ABA) for the next 24 h to evaluate the hormonal control in the presence of high EXP1 levels expressed during PEG preincubation. The A were periodically weighted to measure water uptake (mg. E-1) during incubations. The start of phase III after transferring to the second medium for A preincubated in PEG or ABA and the rate of water gain (slope in phase III) were compared. The A preincubated in PEG restarted water uptake 3 h after its transfer to both water and ABA, whereas A preincubated in ABA did it 12 h after transferring them to water. The rates of water gain in phase III were 6.7; 3.1; 2.7 and 2.2 mg. E-1. h-1 for PEG–WATER, ABA– WATER, PEG-ABA and Control treatments, respectively. It is concluded that the EXP1 accumulation during PEG incubation could contribute to the faster onset of phase III after transfer to water or ABA, whereas A preincubated in ABA required a longer water incubation time to revert its repressive effect on EXP1. Additionally, a possible control by ABA decreasing the rate of water gain in phase III was identified for ABA-WATER and PEG-ABA treatments with respect to PEG-WATER.

#### A29

### PHYSIOLOGICAL QUALITY IN SOYBEAN SEEDS WITH CHLOROPHYLL RETENTION PRODUCED UNDER HEAT-DROUGHT STRESS Martínez MA<sup>1,2</sup>, Montechiarini NH<sup>2</sup>, Gosparini CO<sup>2,3</sup>

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Chlorophyll degradation in soybean seeds [(*Glycine max* L.) Merr.] can be affected by heat and drought stress (ETH) during the late maturation phase, producing green seeds with low physiological quality. In order to evaluate the effect of ETH on the percentages of green seeds, chlorophyll retention and physiological quality, seeds of SRM 3410, SPS

4x4 and DM 4214 STS were produced under ETH conditions during the R5-R7 period in a greenhouse. Non-stressed seeds of those cultivars were used as control (C). Stressed seeds were classified as green (G, with chlorophyll retention) and yellow (Y, without chlorophyll retention). The percentage of green seeds (% GS) and the total chlorophyll (Chl) content were analyzed. Physiological quality was evaluated by the Viability (% Vi), Vigour (% Vg), Germination (% Ge), and times to reach 50% germination (% tG50) in seeds (S), embryos (E), and embryonic axes (Ax). The combination of ETH impacted significantly on % GS in comparison with the CS on the different cultivars. In SRM 3410, SPS 4x4 and DM 4214 STS, % GS were 29.8, 70.3, and 52.5%, while in CS were 1.8, 4.0, and 0.8 %. The respective Chl contents in GS were 9.7, 12.7, and 13 ppm. In GS, the % Vi and % Vg of SRM 3410 (82 and 59%), SPS 4x4 (85 and 63%) and DM 4214 STS (83 and 62%) were lower than the YS (91 and 83%; 99 and 93; 96 and 88%) and CS (90 and 81%; 99 and 92%; 99 and 89%). Significant differences were observed in % Ge of GS with respect to the YS and CS, which were 27, 87, and 87% in SRM 3410; 33, 97, and 100% in SPS 4x4; and 40, 97, and 97% in DM 4214 STS. In GE, YE, and CE were 53, 100, and 100% Ge in SRM 3410; 73, 100, and 100% Ge in SPS 4x4; and 73, 100, and 100% Ge in DM 4214 STS. Meanwhile, in % Ge of Ax were 67, 70, and 73% in G; 100, 100, and 100 % in Y and C, respectively. The average tG50 were significantly longer for G in relation to Y and C in S, E, and Ax. Our results showed that heat and drought stress conditions during R5-R7 resulting in a high incidence of green seeds with low viability, vigour, and germination.

A30

## PLATELET-DERIVED MICROPARTICLES IN PATIENTS WITH SEVERE HEMOPHILIA

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Platelet microparticles (PM) are small phosphatidyl serine rich platelet-derived vesicles (100 nm–1  $\mu$ m), involved in the maintenance of hemostasis. It was proposed that, in hemophiliac patients, they could improve the bleeding profile after factor replacement therapy. The aim of this study was determined if the percentage (%) of plasma MPs in patients with severe hemophilia A before and after factor VIII infusion is different from the % found in normal controls. We studied platelet-free citrated plasma samples obtained from 20 normal individuals and 19 patients with severe hemophilia A pre-infusion, 30 min after factor VIII infusion. MPs were quantified by flow cytometry using a CD 41 (P2) IOT PE monoclonal antibody. The samples were studied with a BD model FACSAria II cytometer and data were analyzed with the FACSDiva v.6.1.3 software. The medians of MPs % were: Pre-treatment patients, 0.50%; post-infusion, 0.46%; normal controls, 0.04%. Data analysis showed a difference between the % of MPs found in hemophiliacs compared to controls, but we did not observe a difference between pre- and post-treatment samples. Unlike other studies, our results show a higher % of MPs in hemophiliacs compared to controls; this could explain their role in a compensatory mechanism of thrombin activation in hemophilia. However, replacement therapy does not modify MPs %, therefore in this condition MPs could not be involved in ameliorating the hemorrhagic phenotype of the patients.

A31

## IMMUNOFLUORESCENCE (IF) STAINING FOR DETECTION OF PML-RARα ONCOPROTEIN AS A RAPID DIAGNOSTIC TEST FOR ACUTE PROMYELOCYTIC LEUKEMIA (APL)

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APL is a highly aggressive subtype of acute myeloid leukemia (AML) that, however, can be cured with targeted therapies. APL is characterized by fusion of the *PML* (promyelocytic leukemia) and *RAR* $\alpha$  (retinoic acid receptor- $\alpha$ ) genes [t(15;17)], leading to expression of PML-RAR $\alpha$  oncoprotein. Clinically, APL represents a hematological emergency since it registers high early mortality due to the risk of hemorrhage, disseminated intravascular coagulation or primary fibrinolysis if the diagnosis is delayed, so the rapid detection of PML-RAR $\alpha$  is of crucial importance for the diagnosis and prognosis of the disease. Detection methods include conventional cytogenetic analysis (CTG), FISH, nested polymerase chain reaction (PCR), and IF staining for PML protein. CTG can detect the t(15;17)(q24;q21) and other chromosomal abnormalities, although the process time may take several days. Nested PCR allows the qualitative detection of the fusion transcript and the isoform (bcr 1, 2 or 3), which is essential for the monitoring of minimal residual disease (MRD). IF allows the detection of the oncoprotein in a period of 2 to 4 h. We performed IF and nested PCR to analyze bone marrow or peripheral blood samples from 20 patients with morphological suspicion of APL. The smears were fixed with methanol, and a primary mouse monoclonal antibody (PG-M3) and a secondary antibody (anti-mouse) conjugated to FITC were used. The samples were analyzed on a

Zeizz® Axio A.1 microscope. 46.7% of the patients were positive with both methods. We can conclude that IF is a simple, rapid, and sensitive technique for the detection of the PML-RAR $\alpha$  oncoprotein. Although it could not replace the nested PCR, IF is a rapid diagnostic test for patients with morphological and clinical suspicion of APL.

#### A32

## EVALUATION OF FUNGAL PATHOGENS IN VITRO CULTURED Zephyranthes candida (LINDL.) BULBS AND THEIR RELATIONSHIP WITH EXPLANT GERMINATION

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The genus Zephyranthes Herb. is represented by herbaceous, bulbous, and perennial plants, with great ornamental value due to the fact that its flowers present a variety of sizes and colors. Zephyranthes candida is propagated by seeds or bulbs. Plant tissue culture techniques reduce the time required for the vegetative cycle, since the bulb reaches the minimum size in a shorter time to complete its reproductive cycle and can also increase the production of seedlings by dividing the bulbs contamination is high, which makes it difficult to multiply in vitro. The objective of this work was to evaluate the presence of pathogens and their effect on the germination of bulbs used as explants for the multiplication of Z. candida. The bulbs were washed with running water under a tap, the first cataphiles were extracted, and work was carried out with whole bulbs, halved, and quartered bulbs. They were disinfected by immersing them for 1 min in 90% alcohol and 20 min in 3% sodium hypochlorite with the addition of 2 drops of Tween 20. Subsequently, they were rinsed with sterilized distilled water. 26 whole bulbs (E), 26 cut transversely in half (E/2), and 18 cut in half (E/4), were seeded in a culture medium by Murashige and Skog (1962) with 30 g/L of sucrose. 8 g/L agar solidified, and the pH was adjusted to 5.8. The explants were grown in a growth chamber at  $24 \pm$ 2 °C. The pathogens were identified with a magnifying glass, microscope, and the use of fungal codes. The following were evaluated: (1) percentage of contamination at sowing, (2) percentage of germination, and (3) type of pathogens present. Disinfection was not efficient, of the total of explants only 10 were not contaminated, of which 6 correspond to E/4. Germination was highly affected when cutting the explants, being 76.9% for (E), 15.38% (E/2), and none for (E/4). The pathogens present were Aspergillus niger, Aspergillus flavus, Penicillium sp., Fusarium sp., and Saccharomyces sp. The pathogens with the greatest presence were Fusarium sp. and Aspergillus flavus, in the cut explants A. niger was eliminated. The presence of fungal pathogens did not affect the germination of the bulbs, being able to generate whole plants in whole explants and dissected in half. This study is an advance in the identification of fungal pathogens in vitro culture for this plant species, as the basis for future disinfection tests and obtaining healthy explants, an essential condition for in vitro multiplication.

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Paoleti Z <u>Paparella C</u> Pazos F Pedrero E Perfumo P Perotti E <u>Perrotta CH</u> Perroud H Peter M <u>Pezzotto SM</u> Pidone CL <u>Pilla T</u> Poli G Ponessa A Prado D Pratti A Pugno A <b>Q</b> Quaglia N	A5 A1 A8 A24 A1 A16 A10 A6 A28 A26 A20 A3 A22 A17 / A18 / A21 A24 / A25 A30 / A31 A28 A9 / A23	Urli L V Vallone C Vallone R Vázquez I Vicente Galán G Viola N W Wagner M Williams GM Y Yaafar N Yordán E Z	A7 A7 A25 A5 A10 A3 / A4 A30 / A31 A22 A28 A17 / A18 /
Paoleti Z <u>Paparella C</u> Pazos F Pedrero E Perfumo P Perotti E <u>Perrotta CH</u> Perroud H Peter M <u>Pezzotto SM</u> Pidone CL <u>Pilla T</u> Poli G Ponessa A Prado D Pratti A Pugno A <b>Q</b> Quaglia N Quiroz M	A5 A1 A8 A24 A1 A16 A10 A6 A28 A26 A20 A3 A22 A17 / A18 / A21 A24 / A25 A30 / A31 A28 A9 / A23	Urli L V Vallone C Vallone R Vázquez I Vicente Galán G Viola N W Wagner M Williams GM Y Yaafar N Yordán E Z	A7 A25 A5 A10 A3 / A4 A30 / A31 A22 A28