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COMPARISON OF TWO METHODS OF FREQUENT USE TO DETERMINE THE ORGANIC CARBON CONTENT IN FORAGES OF THE UPPER DELTA OF PARANÁ

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Given the diversity and quantity of forage plants that this region presents, it was proposed to determine the energy potential (organic carbon content) of the same through two usual analytical techniques: Antrona method (non-structural carbohydrates, CNES) and oxidation with dichromate (Cr₂O₇²⁻) (Oxidizable carbon, C_{oxid}). The plant species used in this work were the following: *Vigna luteola* (VL), *Polygonum acuminatum* (PA), *Panicum pernambuscense* (C1), *P. elephantipes* (C2), *Echinochloa polystachya* (C3), *Eichhornia azurea* (EA), *E. crassipes* (EC), *Baccharis salicifolia* (BS), *Lippia alba* (LA), *Salix humboldtiana* (SH), *Tessaria integrifolia* (TI), *Acacia caven* (AC), *Gleditsia triacanthos* (GT), and two *Medicago sativa* hays (HA16 and HA18). The samples were obtained monthly during the summer spring growth period, in a pre-flowering state by cutting with mechanical scissors, in the Islands that are in front of the city of Rosario. They were then dried at 60°C, ground and sieved with a 2 mm-screen. With the samples of each plant species, a composite sample was prepared. The determinations were made in duplicate. The average values, obtained with both techniques, presented a positive correlation of 0.596 ($P \leq 0.05$). C_{oxid} values (%) ranged from 7.5 to 15.4; being its general mean and standard deviation of 12.53 ± 1.97. CNES values (%) varied between 3.5 and 9.8; being its general mean and standard deviation of 6.94 ± 1.86. Regarding C_{oxid}, the CNES represented only 55%, a predictable difference that indicates the proportion of saccharide derivatives over the total of reduced carbon compounds in the samples studied. The C_{oxid} allowed to show and/or highlight existing differences in the carbon composition, which, without being associated with carbohydrates, influence the reducing power of forage (phenolic derivatives, for example). The results indicate that C_{oxid} allows differentiating samples from the same plant species that presented similar CNES values both in the case of legumes (HA16, HA18 and AC) and in the case of grasses, which also presented similarities with aquatic or shrub species (EC, EA, C1, C2, C3, TI, or PA, LA). It is also concluded that between the studied species differences may occur at the level of the concentration of saccharide derivatives and that these are not reflected in the amount of C_{oxid} that the forage presents (EA, PA, SH or C3, VL, LA), indicating the presence of saccharides with different reducing power. The study suggests the convenience of the complementary use of the two analytical techniques studied when it is required to characterize the energy potential of this type of forage as substrates in microbial processes such as methanogenesis and CO₂ production in the rumen of animals that are confined to the island for productive purposes.

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INCREASE OF ANTIOXIDANT COMPONENTS IN PLASMA OF HEIFERS FEED WITH SOYBEAN EXPELLER DURING NATURAL SERVICE

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Currently, there are numerous balanced diets formulated for beef cattle that contains soybean as an ingredient that supply a high-quality protein. Isoflavones belong to a group of flavonoids that are present in these type of food with an important protective role against cellular oxidative damage. The objective of this work was to evaluate the effect of the soybean expeller diet on the antioxidant state of heifer plasmas during the mating season, and its relationship with pregnancy and calving rates. For that, two groups of *Braford* heifers from the IIACS-INTA were fed for 3 months during the mating period differentially with a diet that contains soybean expeller at 0.6% of the body weight, and the other group was used as a control (diet without soybean). The research was carried out in 2 replicates in consecutive years, with 32 heifers per repetition, divided into two experimental groups. Blood samples from both groups were collected by puncture of jugular vein. Subsequently, hematocrits were determined for each sample. For the determinations of bioactive compounds and antioxidant activity, 10 plasma samples were selected and analyzed in duplicate from each group studied. The total content of phenolic compounds and flavonoid concentrations were determined by spectrophotometry. Animals that consumed soybean expeller during service had the highest levels of these bioactive components. Antioxidant activity was evaluated in plasma by measuring antiradical activity and the protective effect against enzymatic lipid peroxidation. An increase in antioxidant activity was determined for both measurements in the group that consumed soybean expeller. In addition, the level of malondialdehyde (MDA) was evaluated as a biomarker of oxidative stress by TBARS method. Plasma samples showed significantly lower MDA concentrations than in the control group. Finally, the hematocrits, the pregnancy and calving rates were not significantly different between groups. These results demonstrate, for the first time, that the inclusion of soy expeller in the diet of heifers during the mating season increased the antioxidant capacity of plasma maintaining adequate physiological conditions without affecting reproductive parameters.

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NEW TOOLS FOR *IN VITRO* HANDLING AND SELECTION OF CRYOPRESERVED SPERM SAMPLES IN EQUINES

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Cryopreserved sperm (SPZ) are used widely in domestic animals for assisted reproduction techniques (ART). However, cryopreservation procedures negatively affect SPZ quality, causing changes at the structural and molecular levels. Therefore, using cryopreserved SPZ for ART can decrease the efficiency of these techniques as well as the quality of the embryos obtained, being necessary to optimize the handling of this type of sample. Particularly in equines, ICSI is the most used technique for low-quality equine semen samples where SPZ is selected based on their motility and morphology. Physiologically, the oviduct epithelial cells (OEC) are involved in the selection of a population of SPZ suitable for oocyte fertilization,

which is released from the oviduct due to the capacitation process. This work aimed to evaluate different conditions for the *in vitro* manipulation and incubation of equine cryopreserved SPZ and to establish an *in vitro* OEC culture to select an SPZ population with fertilizing capacity suitable for its use in ART in this species. Regarding the handling and the effect on the motility of post-thawed equine cryopreserved SPZ samples, we used non-capacitating Whitten's medium, less effect on motility was observed through the time (120 min) when SPZ were incubated at concentrations of 30 mill/mL compared to lower concentrated samples (CASA, $P < 0.05$). Also, the motility decreased by half with centrifugations at 200g longer than 1 min ($P < 0.05$). On the other hand, we have established a culture model of OECs *in vitro*, in which we demonstrated the expression of epithelial markers (E-cadherin and cytokeratin) by both RT-PCR and immunofluorescence (IF). When we performed SPZ-OEC cocultures, we observed that the attached SPZs were motile and presented intact acrosome (PSA-FITC, IF), suggesting a selection by the oviductal model. Then, the co-cultures were incubated in capacitating conditions (capacitating Whitten's medium) and the released SPZ population was recovered. Under these conditions, a greater number of live sperm (Hoechst258 assessed by IF), capacitated (activation of PKA and phosphorylation on tyrosine residues by IF), with progressive motility (CASA) and with the intact acrosome (PSA-FITC, IF) compared to the control was observed ($P < 0.05$). Moreover, the decrease in motility through the time was less in these SPZ than in the cryopreserved SPZ incubated in the absence of OECs. This sperm population could be recovered for its use in different ART. Improvements in handling and selection of cryopreserved SPZ not only generate tools to solve the problem that IVF presents in equines but would also improve efficiency in other ART such as ICSI, allowing the use of a population of higher quality gametes which it would positively impact the quality of the embryos obtained.

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TRACE MINERALS IN SERUM OF LACTATING SOWS

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Oligominerals such as iron (Fe), copper (Cu) and zinc (Zn) are essential for pig nutrition. Objective was to analyze the profile of oligominerals (iron, copper and zinc) in serum samples of sows lactation in two commercial farms of Argentina. The blood samples were extracted without anticoagulants, from 68 lactating sows of commercial genetics constituted by crosses of Yorkshine, Landrace and Pietrain breeds; randomly selected from establishments located in Santa Fe (A) and Entre Ríos (B). The diet was based on corn, soy expeler and enriched with mineral vitamin nucleus of inorganic origin for lactation category. Between both farms there is difference in management of the site of calving and lactation, in A they are carried out to field and in establishment B they are carried out in confinement. Samples for the determination of Fe, Cu and Zn in serum were analyzed by atomic absorption spectrophotometry (FAAS) method. Infostat program was applied, outliers were identified and eliminated prior to analysis, then Shapiro-Wilk test and Levene test were applied. All samples were found with normal distribution and showed homogeneity in their variances. T-test was performed assuming a significance of 0.05. Standard deviations and test values t were: Fe ($\mu\text{g/dL}$) 6.07, 19.19, t 0.00 – Cu (ppm) 0.35, 0.32, t 0.00 – Zn (ppm) 0.50, 0.42, t 0.01 for farms A and B, respectively. During investigation period were observed within normal, values average values of iron in both farms and copper of farm A. Establishment B, zinc was found slightly above the upper limit of reference range and same oligomineral, at farm A, showed averages above normal value. Means of three minerals were found to be significantly different ($P < 0.05$) for the lactation condition between the two farms. This behavior can be explained, if we consider that oligominerals Fe, Cu and Zn are normally added as correctors by commercial food companies at levels higher than recommended by most research centers.

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EVALUATION OF THE PROTEIN CONTENT IN SORGHUM FORAGE

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The use of marginal areas for livestock production and their edaphoclimatic limitations led to search adapted food for livestock, adapted to these conditions. Facing the development of sustainable systems, sorghum presents a very good potential for livestock feed. Sorghum cultivation resists conditions of extreme temperatures and drought, giving grain and forage in arid, sub-humid and even hot areas. It shows a great versatility for livestock feeding since it can be used as greening, deferred, silo, as grain etc. Geneticists, over the years, have sought to improve their biomass yields. Genetics improvement has achieved interesting increases in dry matter productivity per hectare. To be used as food for livestock, we are not only interested in its performance but also its nutritional value. The protein content is one of the parameters to consider when quantifying forage quality, due to the functions they perform in the animal's body. The objective of this work was to evaluate the crude protein content (CP) of commercial varieties of forage sorghum used as livestock food and to compare with data from varieties commercial sorghum used ten years ago. We worked with 20 samples of forage sorghum of 4 commercial varieties. The samples were dried in a drying oven at a temperature of 60°C until obtaining dry matter (DM). Then they were ground in a brand laboratory mill (Wiley®) with a 1 mm sieve and the flour obtained was used to determine the CP by the Kjeldahl method, according to the AOAC (1990), multiplying the value of N_2 obtained by the factor 6.25 to estimate the % of CP. The results obtained were the following MN°1: % CP = 9.32 ± 0.22 ; MN° 2: % CP = 8.97 ± 0.48 ; MN° 3% CP = 9.06 ± 0.29 ; MN° 4% CP = 9.32 ± 0.48 . The data of commercial sorghum varieties used ten years ago, was obtained from the database of the Animal Nutrition Laboratory, FAZ, UNT. Whose medium value of % CP = 6.71 ± 0.51 vs. the mean value of the new varieties of % CP = 9.19 ± 0.15 . Observing an improvement in the % of CP of 2.48. Concluding that in the new varieties, there was not only an improvement in the yields, but also in a nutritional parameter, as sensitive as the protein values.