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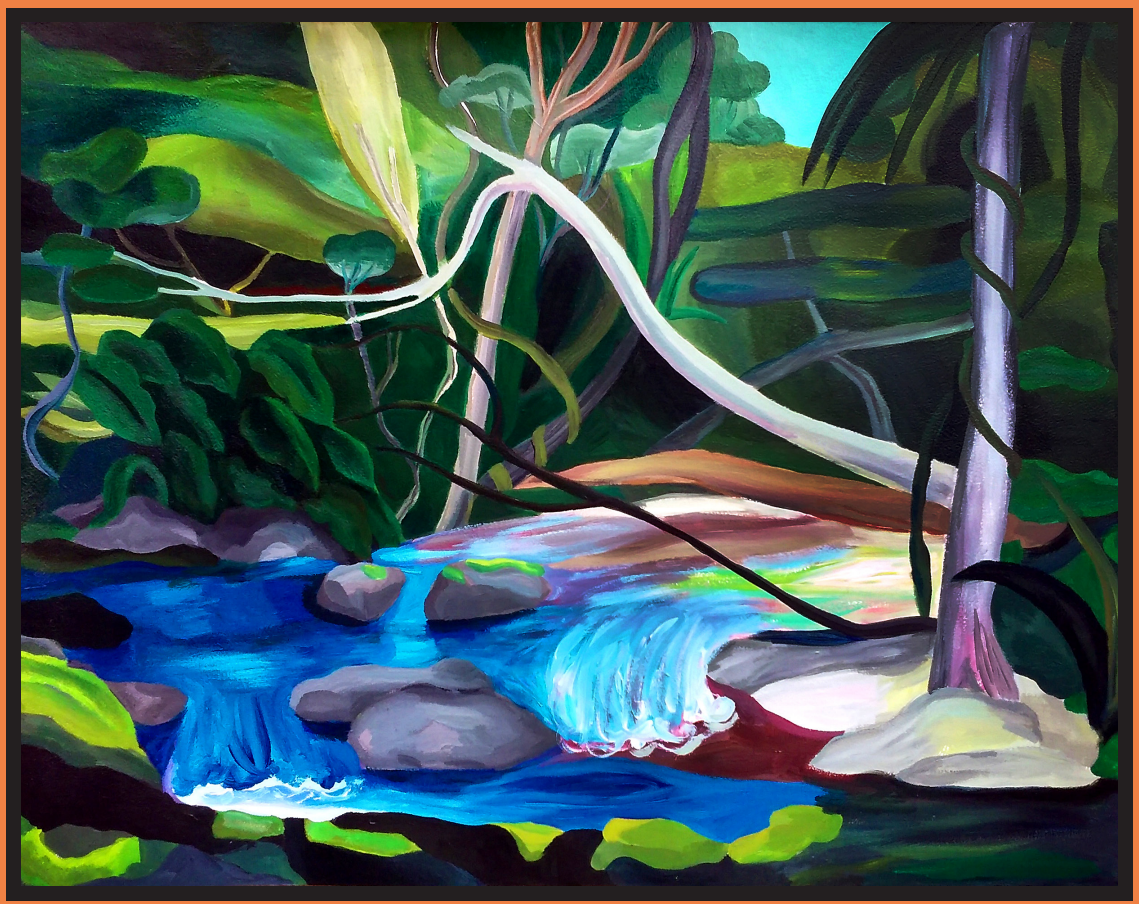

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(*3β-HSD* and *aromatase*), antral follicles were stored at -80°C for subsequent RNA extraction and quantitative RT-PCR (qRT-PCR). mRNA expression was normalized to the endogenous control, *β-Actin*. Culture media was stored at –80°C for further determination of progesterone (P4) and estradiol (E2) levels by electrochemiluminescence. Normalized mRNA results for the assessed apoptotic regulatory genes (*Bcl-2*, *Bax*, and *Bad*) showed no significant differences in the presence of HN ($p>0.05$). Concerning steroidogenesis, HN interestingly increased the P4 levels in the culture media ($p<0.05$) without modifying *3β-HSD* mRNA expression ($p>0.05$). Regarding E2, HN did not change either the E2 levels or *aromatase* mRNA expression ($p>0.05$). In conclusion, our results suggest that HN may probably modulate antral follicle development by increasing P4 levels.

521. 555. **DETRIMENTAL EFFECTS OF LINDANE ON THE BOVINE OVIDUCTAL EPITHELIAL CELLS**

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Lindane, the **γ-**isomer of the hexachlorocyclohexane, was a widely used organochlorine pesticide in agriculture and public health. Due to its high environmental persistence, bioaccumulation, and toxicity, in the present work we investigate its effects on primary cultures of the bovine oviductal epithelial cells (BOEC), focusing on cell viability, proliferation, migration, and genotoxicity. Different lindane concentrations (12.5 **μ**M, 25 **μ**M, 50 **μ**M, 100 **μ**M and 200 **μ**M) were tested on 80% confluent BOEC monolayers in DMEM 5% FSB at 38.5°C, 5% CO_2 and 100% humidity. Then cell viability was evaluated by the trypan blue exclusion test. Cell viability was above 86% at all the lindane concentrations assayed. However, concentrations starting from 25 μM exhibited a significant decrease in cell number/mL compared to the control ($p<0.05$). We selected 50 **μ**M to evaluate the lindane effect on the proliferative capacity of BOEC by a clonogenic assay. Cells at low density (500 cells/60 mm plates) were seeded and incubated overnight for cell attachment. The medium was then replaced with or without lindane (control) and cultured for 7-10 days. Colonies were then stained with Giemsa and scored; a substantial reduction in the colony formation was observed ($p<0.05$). Cell migration was evaluated by the wound healing assay; scratches were made in confluent BOEC cultures and incubated at 3 h, 6 h, and 12 h in DMEM 5% FBS with or without lindane (50 **μ**M). Cells in the presence of lindane displayed a tendency to delay cell migration. Lindane's genotoxicity was evaluated by a micronucleus assay after 48 h of exposure to 50 **μ**M of lindane. Cells fixed in 4% formaldehyde and stained with Hoëchst 33342 exhibited a 2,4-fold increase in micronucleus formation. Results suggest that lindane have detrimental effects on the oviductal epithelium altering essential cellular processes that potentially impair female fertility.

522. 589. **EVALUATION OF HISTOMORPHOMETRIC CHANGES IN THE UTERUS OF PRENATAL RESTRICTED LAMBS**

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The aim of the study was to describe the histomorphometric characteristics of the uterus of 45-days-old lambs belonging to mothers nutritionally restricted or controls from the second half of gestation. Merino pregnant ewes carrying single fetuses were randomly assigned to restricted (R) or control (C) groups and fed with 75% or 125% of metabolic energy requirements from ~80 days to ~140 days of gestation, respectively. Uterus samples from Merino female lambs of 45-days-old (n=7 for each group) were evaluated. For the histomorphometric evaluation, 3 cuts of 4 μm thickness of each uterine horns were obtained and stained with H-E in order to evaluate the following parameters: *height of the luminal epithelium (HLE) and the glandular epithelium (HGE)*, considering the distance between the basal membrane and the apical end of 30 cells of each of the epithelia with a magnification of 100x. *Glandular density (GDe)*, the number of glands in 10 fields at 40x magnification and *Glandular area* (GA) using the following formula: GA=π×(Axis 1)/2×(Axis 2)/2 were evaluated. The axes were the average of two right-angled diameters in a minimum of 30 glands per section at 100x magnification. Samples were analyzed by two observers. The results were analyzed by ANOVA. Data are expressed as mean ± SD. Results were considered significant with $P\leq0.05$. HLE and AG did not differ between groups (**R**: 11.73±0.18; 1347.43±36.74 and **C**: 11.81±0.18; 1277.54±36.74; respectively; $P\leq0.05$). The HGE and GDe were significantly higher for lambs from R than lambs from C group (**R:** 13.34±0.18; 25.19±0.59; **C:** 12.86±0.18; 22.32±0.64; respectively; $P\leq0.05$). The pre-natal maternal nutrition could affect the development of uterine glands in their lambs. In this study, a higher HE and GDe was observed in the uterus of lambs born from nutritional restricted mothers. Future studies are necessary to explain these results.

523. 602. **INNOVATIVE THERAPEUTIC STRATEGY FOR AUTOIMMUNE EPIDIDYMO-ORCHITIS IN RATS: LOCAL INJECTION OF MELATONIN WITHIN THERMOSENSITIVE PEOPPO COPOLYMERS**

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Experimental autoimmune epididymo-orchitis (EAO) is a well-established rodent model of organ-specific autoimmunity associated with infertility. In EAO oxidative stress negatively impacts on spermatogenesis. Our study aims to explore the potential of intratesticular injection of melatonin (MLT), an antioxidant hormone with poor solubility in aqueous medium, to ameliorate the effects of inflammation on epididymal sperm parameters. For this objective, we used Pluronic® F127, a biocompatible polymeric micelle (PMs) nanoplatform with *in vivo* thermosensitization seeking to optimize MLT delivery and availability within the testes. Two groups of adult male Wistar rats were evaluated: non-immunized (normal, N) and experimental rats which were immunized with testis homogenate and adjuvants to induce EAO. Rats received a single intratesticular injection of F127 25% (w/v) with MLT (2.5mg) or F127 25% (w/v) in saline (as control) and were sacrificed 10 days after. In EAO rats MLT prevents reduction in cauda sperm viability (% media±SEM, N+F127: 84,8±0,6; EAO+F127: 47,4±8,1; EAO+F127-MLT: 71,4±9,9 $p<0.05$ vs EAO+F127) and motility (% media±SEM, N+F127:78,5±2,4; EAO+F127: 37,8±6,1; EAO+F127-MLT: 61,9±11,1 $p<0.05$ vs EAO+F127). Head and tail sperm abnormalities were also reduced by MLT [% media±SEM, Head: N+F127: 7,90±0,5, EAO+F127: 37,6±1,6, EAO+F127-MLT: 10,4±0,8 $p<0.01$ vs EAO+F127; Tail: N+F127: 14,75±0,07, EAO+F127: 48,4±1,9, EAO+F127-MLT: 17,1±2,6 $p<0.01$ vs EAO+F127. Sperm count significantly decreased in all EAO groups vs. N rats. F127 25% (w/v) increased the maximum solubility of MLT by 14 times vs saline, potentially amplifying its effect. MLT injected within the testis might act on stored epididymal sperm mitigating the detrimental action of the inflammatory microenvironment possible through the strong antioxi-