

66 CUMULUS-OOCYTE-COMPLEX SECRETIONS FROM THE FIRST 8 HOURS OF *IN VITRO* MATURATION AFFECT OOCYTE DEVELOPMENTAL COMPETENCE

K. Uhde, H. T. A. van Tol, T. A. E. Stout, and B. A. J. Roelen

Department of Farm Animal Health, Utrecht University, Faculty of Veterinary Medicine, Utrecht, the Netherlands

Mammalian oocytes are surrounded by cumulus cells, forming a structure known as the cumulus-oocyte complex (COC). Cumulus cells play important protective functions during oocyte maturation, for example, protecting the oocyte against reactive oxygen species. However, it is not yet fully understood how the cumulus complex modulates the developmental competence of the enclosed oocyte. It was investigated whether direct contact between an oocyte and its cumulus cells is essential throughout the maturation process. To this end, bovine oocytes aspirated from ovarian follicles were matured *in vitro*. Eight hours after the onset of maturation the cumulus cells were removed, and the oocytes either placed back in the original medium or cultured further in fresh maturation medium. In all experiments, COCs/oocytes were matured for 23 h in M199 supplemented with 0.05 IU FSH and penicillin/streptomycin. All experiments were performed in triplicate, with 35 to 45 COCs per group. Student's *t*-test was used for a paired comparison. Denudation after 8 h and return to the same maturation medium had no effect on the cleavage rate (93%) compared with culture without denudation (90.7%). Only if the oocytes were transferred to fresh medium did the cleavage rate decrease slightly (75.4%; $P = 0.038$). By contrast, blastocyst formation was reduced nearly four times if COCs were denuded before being returned to the medium, compared with controls (14% v. 50.8%; $P < 0.001$). If the oocytes were transferred to fresh medium after denudation, very few blastocysts resulted (0.9%; $P < 0.001$). In a second study, oocytes denuded immediately after removal from the follicle were matured in the absence or presence of cumulus cells in a Corning® Transwell® system. Culturing denuded oocytes in the presence of cumulus cells resulted in similar cleavage rates (83.5%) to control conditions (84.8%). However, blastocyst formation was markedly lower (4.3%) than for controls (29.6%; $P = 0.003$). We conclude that COCs secrete substances during the first 8 h of maturation that are beneficial for oocyte acquisition of developmental competence. Moreover, intimate contact between the cumulus cells and oocyte is essential.

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67 INHIBITION OF 5 α -REDUCTASE DURING LATE GESTATION IN THE MARE

*M. Wynn^A, E. Legacki^B, A. Conley^B, S. Loux^A, A. Esteller-Vico^A, S. Stanley^B, E. Squires^A,
M. Troedsson^A, and B. Ball^A*

^AUniversity of Kentucky, Lexington, KY, USA;

^BUniversity of California, Davis, Davis, CA, USA

During the latter half of pregnancy in the mare, circulating concentrations of progesterone (P4) are low and a major bioactive progestogen, 5 α -dihydroprogesterone (DHP), is present in high concentrations. DHP is formed through the activity of 5 α -reductase, which converts P4 to DHP. Further metabolites of DHP have been attributed to fetal and myometrial quiescence. The purpose of this study was to examine the effects of a 5 α -reductase inhibitor (dutasteride) on P4 metabolism and onset of parturition. Pregnant mares ($n = 5$) were treated with dutasteride (0.01 mg kg⁻¹; IM), and control mares ($n = 4$) received vehicle alone from 300 to 320 days of gestation or until foaling. Serum concentrations of P4 and DHP were determined with liquid chromatography/tandem mass spectrometry (LC/MS-MS) daily for 9 days preceding parturition. Endocrine data were analysed with a random effects mixed model with time, treatment (TRT), and time \times TRT interaction. Gestational data were analysed with a Wilcoxon test. Although there was a significant effect of time on P4 and DHP, there was no effect of TRT or time \times TRT on these progestogens. For the ratio of DHP/P4, there were significant effects of time, TRT, and time \times TRT interaction such that the ratio of DHP/P4 was greater in the control than dutasteride-treated mares. Birth weight and gestational length were not different ($P > 0.2$) between the dutasteride-treated and control mares, although placental weights were greater ($P < 0.05$) in dutasteride-treated mares. Because the ratio of DHP/P4 was suppressed in dutasteride-treated mares, it appears that dutasteride was active in late gestational mares. Although gestational length and neonatal weights were not different between groups, placentas from dutasteride-treated mares were heavier than those from control mares. The reason for the increase in placental weights was not determined.

68 STEROIDOGENIC CONTROL OF INTRAUTERINE SEXUAL DIFFERENTIATION IN SPIX'S YELLOW-TOOTHED CAVY, *GALEA SPIXII*

A. C. Santos^A, D. C. Viana^A, F. D. Oliveira^A, M. F. Oliveira^B, and A. C. Assis-Neto^A

^AUniversity of São Paulo, São Paulo, SP, Brazil;

^BFederal Rural University of Semiárid, Mossoró, RN, Brazil

The Spix's yellow toothed (SYT) cavy is a species of rodent that lives in Caatinga vegetation of northeast Brazil. The SYT is utilised as a protein source by inhabitants of that region and has zootechnic potential. SYT cavies have been bred in captivity for studies related to reproductive biology. The presence of a penile clitoris trespassed by the urethra was described in the adult female. The aim of this research is describe sexual differentiation events in the male and female conceptus and relate molecular mechanisms of androgen and oestrogen metabolism of the placenta with the differentiation of external genitalia. Conceptuses after 15, 22, 30, 40, >50 (full term) days of gestation (DG; $n = 9$ males and 9 females at each gestational period) from 30 pregnant females, which were paired with 10 males, were used. After detection of copulation, the above described DG