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Estrous cycle phases on mare endometrium secretory function and fibrosis markers (TIMP 1 and TGFβ1) after neutrophil extrace traps and cytokines action

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Neutrophils extracellular traps (NETs) and cytokines may play a role against bacterial infection and may induce fibrosis. Estrous cycle of mares, effects on endometrial secretory function, tissue inhibitor of metalloproteinase 1 (TIMP) and transforming growth factor beta 1 (TGFβ) production after NETs and cytokines action were assessed. Endometrial explants (n = 5 mid-luteal- LP; n = 5 follicular phase-FP) were cultured for 24, 48 and 72 h with NETs components (elastase – ELA; cathepsin G – CAT; myeloperoxidase –MPO) or connective tissue growth factor (CTGF), tumor necrosis factor-alpha (TNF) and oxytocin (OT). Prostaglandin F2α (PGF), TIMP and TGFβ were analyzed by ELISA. In LP compared to FP (i) PGF decreased at 24 h (ELA, CAT, OT, TNF) and at 48 h (ELA; $p < 0.001$); (ii) TIMP increased at 24 h (MPO, CAT, CTGF) and 48 h (CAT, CTGF, OT) ($p < 0.05$); (iii) and TGFβ decreased at 24 h (CAT, CTGF) and increased at 72 h (CAT, CTGF, OT) ($p < 0.05$). Between 24 and 72 h in LP: (i) PGF increased (ELA, CAT, OT, TNF; $p < 0.05$); (ii) TIMP raised with CAT ($p < 0.05$); and (iii) TGFβ increased (CAT, CTGF, OT; $p < 0.05$). Between 24 and 72 h, in FP: (i) PGF did not differ; (ii) TIMP increased with time (ELA, MPO, CTGF; $p < 0.05$); (iii) no treatment effect for TGFβ. MPO raised TGFβ between 24 and 48 h ($p < 0.05$). Estrous cycle might influence mare endometrium luteolytic secretion and fibrosis establishment when stimulated by NETs and cytokines, as indicated by PGF, TIMP and TGFβ production in the luteal phase.

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Concentrations and expression of growth differentiation factor 9 as an indicator of follicular development and atresia in cattle

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During ovarian follicular development, granulosa cell (GC) and theca cell proliferation and differentiation are influenced by gonadotropins, growth factors, and numerous intra-ovarian factors secreted by both the oocyte and surrounding somatic cells. This includes members of the transforming growth factor-ß superfamily such as growth differentiation factor 9 (GDF9). The objective of this study was to characterize changes in intra-follicular GDF9 protein and mRNA levels during follicular growth in cattle. Follicular fluid (FFL) and GC were collected from cows selected (Twinner) and unselected (Control) for multiple ovulations and twin births, at day 3 or 4 (recruitment, D3) and day 5 or 6 (deviation, D5) of an estrous cycle. Follicles of Twinner vs. Control were classified as healthy (E2:P4 > 1) or atretic (E2: P4 < 1). FFL-GDF9 protein levels were determined by a newly developed double antibody radioimmunoassay and GC-GDF9 mRNA was quantified using quantitative real-time RT-PCR. Atretic follicles had 1.75-fold greater ($p < 0.05$) GC-GDF-9 mRNA abundance than healthy follicles, but FFL-GDF9 protein levels did not differ ($p > 0.10$). Twinner cows had 73% less ($p < 0.05$) GC-GDF9 mRNA than Control cows at D3, and this led to a lower FFL-GDF9 protein level observed at D5 (8 vs. 15 μg/ml; $p < 0.05$). We hypothesize that the lower levels of expression of GDF9 in Twinner cows may participate in the selection of multiple follicles.

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Treatment of anoestrus with GnRH or progesterone in dairy cattle

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The aim was to compare the effect of GnRH and progesterone treatments in anoestrous dairy cattle. The data consisted of 404 treatments of true anoestrus performed at fertility control visits on 15 dairy farms in years 2000–2010. The diagnosis was based on palpation per rectum (absence of CL and no signs of oestrus) by one specialised veterinarian, and ultrasound examination (stage of follicular wave, sign of regressed or growing CL) was used when necessary. Of the cases, 215 were treated with GnRH (194 with 12–20 μg of buserelin) and 189 with progesterone releasing device (154 CIDR, 35 PRID), which was in place for 7–12 days. Of the devices, 136 contained oestradiol whereas 53 did not. In 27 cases, the treatment was repeated, and depending on the outcome studied, only the first or last was included. The mean time of diagnosis post partum (pp) was 77.5 ± 42.1 (median 63) d. On average, GnRH treatment was given 42 days earlier than progesterone (59.9 ± 22.8 vs. 101.9 ± 47.4 days pp, respectively). Eventually 149 and 139 animals got pregnant and the mean time from treatment to conception was 76.9 ± 52.6 and 57.0 ± 52.8 days ($p = 0.001$, t-test) after the GnRH and progesterone treatment, respectively. Parity and time from calving to treatment did not seem to have any effect on the timing of the first AI and conception. Thus, the difference in the timing of conception cannot be explained by the fact that GnRH treatment was given earlier pp than progesterone. After progesterone treatment oestrus was evaluated to be synchronised in 78.4% of cases treated with device fitted with oestradiol while in 72.7% without oestradiol. Anoestrous animals conceived about 3 weeks earlier after having the progesterone device than those treated with GnRH.

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Pyovagina caused by pyometra and a persistent hymen in a 9-year old bitch

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A 9 year old intact bitch was presented at our clinic for a swelling in the perineal region, apathy, inappetance, vomiting, constipation and pollakiuria present since 3 days. The bitch had shown purulent vulvar discharge 1 month earlier. Normal heats had been observed twice a year since puberty. At palpation, the perineal mass had a soft consistence and could not be reduced. Vaginal palpation was impossible to perform. At rectal palpation, the mass could be palpated ventrally from the rectum. Blood analysis showed a leukocytosis and globulinemia. Ultrasound of the swelling showed a liquid with a cellular content, prolonging into the abdominal cavity. Abdominal ultrasound revealed a similar content within the entire uterus, concordant with a pyometra. The bitch received fluid therapy and antibiotics and an ovariohysterectomy was performed subsequently. As the vaginal swelling complicated the surgery, the content of the uterus and vagina was aspirated with a urinary catheter introduced at the level of the right uterine horn. After surgery, a speculum exam of the vagina revealed a persistent hymen that was then perforated with a swab. Normal bloody discharge during regular prooestrus and the recent purulent vulvar discharge reported indicate that the hymen was, at least partially, perforated. For unknown causes, the hymen must have re-sealed after uterine contamination and further development of the pyometra explaining the uncommon form of pyovagina visible as a mass in the perineal region of the patient.