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HYDROMETRA IN A SHEEP AFTER OESTRUS SYNCHRONIZATION AND INSEMINATION IN THE ANOESTRAL SEASON

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Summary: A case of hydrometra in a Blackhead Pleven sheep following oestrus synchronization and artificial insemination in the anoestral season is detected.

The causes of the disease are discussed, the clinical signs and the echographic findings are described. The haematological, blood biochemical parameters and progesterone concentration were assayed. The gross anatomical and histological changes in the genital tract are reported.

The presented case showed that in sheep hydrometra was not accompanied by deviations in the general health condition, haematological and blood biochemical profile. Progesterone level did not correspond to value indicating pregnancy. A strong thinning of the uterine wall, aseptic fluid in the uterine cavity and persistent corpus luteum in one ovary were found out. In the endometrium, there were extensive lesions and strands dilatation of superficially located uterine glands.

In order to diagnose hydrometra in sheep, we recommend two echographic examinations and visualization of intrauterine anechoic fluid, thinned uterine wall and lack of embryo and placentomas.

Key words: sheep; hydrometra

Introduction

Hydrometra or pseudo pregnancy is a common disease of the genital tract in goats, described by a number of investigators (1,2,3,4). It is characterized with accumulation of aseptic fluid into the uterus, presence of persistent corpus luteum in the ovaries, high blood progesterone concentration and is observed in goats with spontaneous ovulation or after oestrus synchronization (5,6,7).

The hydrometra in sheep is a genital pathological state leading to permanent infertility (8). It is rarely encountered and that is why is not studied in details.

In threefold echographic pregnancy examination of 1,411 sheep from the Rambouillet breed and its crosses, hydrometra was observed only in 2.9% of cases (9). After the second examination, the cases of

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hydrometra were found to increase and it is pointed out that the examination stress could be a cause for the onset of the diseases

In a study on genital tracts of 1,042 slaughtered Awassi sheep, hydrometra was registered in 0.3%(10). Another authors have investigated the genital tract of 9,970 culled and 23,536 non-inseminated sheep and detected hydrometra in 18 and 61 animals respectively (8).

Case history

A case of hydrometra in a Blackhead Pleven sheep at the age of 4 years, weighing 62 kg, reared at a place situated at latitude of 42° 25' N.

The animal has given birth thrice, and the last parturition and the postparturient period were normal. In June 2008, echography of the genital tract of the sheep was performed. No pathologies have been observed.

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The oestrus was synchronized by means of Chronogest® intravaginal sponges (30 mg Cronolone), that stayed into the vagina for 12 days and treatment with Folligon® (500 UI PMSG). Artificial insemination was done twice on hours 48 and 56 after sponge removal.

By the 40th and the 60th day after the insemination, the sheep was examined for pregnancy by transrectal and transabdominal echography with an echograph Aloka SSD 500 Micrus (Aloka Co. Ltd, Tokyo, Japan) supplied with a 5 MHz probe.

A physical examination was performed and blood samples for haematological, biochemical and hormonal analysis were obtained.

Haematological parameters were assayed on an automated haematological analyzer (Serono Plus, Germany), the biochemical profile on a biochemical analyzer BA 88 (Mindray, China), and progesterone level – by ELISA (Elisa kit for Progesterone, Human GmbH, Germany) with a sensitivity >0.03 ng/ml and a standard curve from 0 ng/ml to 40 ng/ml.

After the second echography, the sheep was slaughtered, a gross examination of the genital tract was performed and the volume of uterine fluid was determined.

Samples for histological examination were obtained from the uterine horns, fixed in 10% solution of neutral formalin and processed according to routine histological techniques (11). After dehydration, embedding in paraffin, preparation of histological cross sections on a microtome (Reichert–Jung, Austria) and staining with haematoxylin-eosin, they were examined under light microscope NU–2 (Carlzeiss, Jena, Germany) at magnifications from 1:50 to 1:250 (12).

Results

Physical examination: Rectal body temperature - 38.7°C, heart rate - 52 beat per minute, respiratory rate - 18/ min, ruminal movements - 8/5min.

Hematology, blood biochemistry and hormonal assay: RBC - 9 T/L, Hb - 120 g/l, HCT - 0.32 l/l, WBC - 12.5 x 10 9 /l, AST - 146, ALT - 78, Ca - 2.32 mmol/l, P - 1.46/mmol/l, Mg - 0.72 mmol/l, total protein - 55 g/l, cholesterol - 1.26 mmol/l, P₄ - 8.2 ng/ml.

Echography: Two echoic luteinized structures with a diameter of > 8 mm and four anechoic follicle of a diameter of 3–5 mm were observed in the left ovary (Fig.1a). In the right ovary, multiple follicles with diameters < 5 mm were present. In the uterus, several anechoic zones, located ventrocranially to

the urinary bladder and thinned echoic uterine wall without visible embryo and clearly manifested placentomas were observed (Fig.1b).





Figure 1: Echography of ovary (a) and uterus (b) in a sheep with hydrometra: 1 - corpora lutea; 2 - follicles; 3 - thinned uterine wall and anechoic uterine fluid

Gross anatomy and histological examinations: In the left ovary, two corpora lutea and several small follicles, and in the right ovary – multiple small and medium-sized follicles were observed. The left uterine horn was enlarged, with a strongly thinned and transparent uterine wall at the horn's tip (Fig. 2). The uterus was filled with 26.4 ml clear fluid without pus or blood, and on the surface of the endometrium in the left uterine horn, loci of darker colour and millet to lentils size were established.

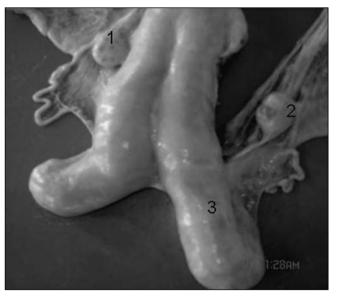


Figure 2: Uterus in a sheep with hydrometra: 1 - right ovary with small follicles; 2 - left ovary with corpora lutea; 3 - dilated uterine horn

The histological examination showed no alterations in the microstructure of the perimetrium and myometrium.

There was a thinning of the endometrium in the areas with hydrometra. Extensive lesions were present in lamina epithelialis mucosae.

The epithelial cells of the single-layered or double-layered prismatic epithelium that were in contact with altered areas, possessed microstructural changes. In these areas, Lamina epithelialis was built most frequently from one layer of cells, whose height was lower than that of cube-shaped cells.

In both types of lining epithelial cells – those with initial height reduction and those having completed this process, the cell cytoplasm was partially or totally vacuolated. Various amounts of diffusely located, basophilically stained granules were detected.

The most perceptible changes were observed in the uterine glands of the endometrium that was in contact with hydrometra. Significant differences in the number, density, and the extent of branching, lumen width and functional state were observed between uterine glands situated in the deep and the superficial endometrial zones (Fig. 3). Uterine glands were strongly dilated, with the width of their lumen exceeding many times that of deeply located ones.

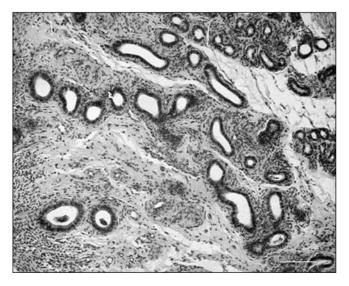


Figure 3: Endometrium with desquamated lining epithelium and structural changes in uterine glands in a sheep with hydrometra (H/E; 1:250, bar= $12.5 \,\mu m$)

The glandular epithelium in some glands showed destructive changes and initial vacuolization. Single intraepithelial glands of a various size, at some places transformed into intraepithelial cysts, were observed.

Discussion

The presented case evidenced that hydrometra in sheep could be detected after oestrus synchronization and artificial inseminations in the anoestral season. In goats with this state was registered or without insemination (13), regardless of whether they were fertilized or not, whereas (14) found it after oestrus synchronization. These reports support the above mentioned statement.

The disease could pass without symptoms, without discharge from the external genitals and deviations in the haematological and blood biochemical indices. Only leukocytes were slightly elevated, but could not be a precise marker of inflammation of the uterus.

The blood serum progesterone concentration of 8.2 ng/ml showed the presence of corpus luteum in the ovaries, but was lower than the levels indicated a gestation of 40 days in this sheep breed as determined in previous studies of ours (15).

These results showed that the prolonged effect of progesterone is able to induce hydrometra in sheep too, not only in goats (3,16).

The echography of ovaries and the uterus is the most precise means to detect hydrometra in sheep *in vivo*. The visualization of corpus luteum in the ovaries, the thinned uterine wall, the lack of embryo and the clear visualization of placentomas are all proofs for the occurrence of hydrometra.

In agreement with the previous studies (9), we also believe that two echographies are needed for the correct diagnosis.

The diagnostic error is highly probably in case of a single echography, especially in the early gestational stages. Moreover, when non-fertilized or in cases of early embryonic death, sheep do not exhibit a regular sexual cycle, as they are not in the breeding season.

The gross examination of the genital tract did not show remnants of foetal membranes or an embryo, excluding early embryonic death as a cause. The nature of the fluid and the uterine changes suggest that this was not a case of pyometra and they were similar to those reported by previous authors (8).

In our view, the extensive lesions observed throughout the histological examination, could be probably due to the pressure exerted by the fluid, resulting in cell desquamation and lack of lining epithelium. The basophilic granulation and the total vacuolization of cellular cytoplasm in Lamina epithelialis provided evidence for destructive pro-

cesses in the cells. The increased incidence of cross sections of glands in areas of desquamated epithelium or reduction indicated that they were excessively coiled or branched. Similar changes in the endometrium are described in goats with hydrometra (13).

In conclusion, hydrometra in sheep could be detected after oestrus synchronization and artificial insemination in the anoestral season. There were not clear clinical manifestations of the disease, or any deviations in the haematological and blood biochemical parameters. The Histological changes of the endometrium consisted in vast lesions with destructive events and altered height of epithelial cells situated around them. The strong dilatation of the lumen of superficially located uterine glands is specific for the disease.

In order to diagnose hydrometra in sheep *in vivo*, two echographic examinations at 20-day interval are recommended, as well as visualization of persistent corpus luteum in the ovaries, anechoic fluid in the uterus, thinned hyperechoic uterine wall, lack of embryo and placentomas.

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HIDROMETRA PRI OVCAH PO SINHRONIZACIJI GONITVE IN UMETNI OSEMENTIVI V OBDOBJU NEGONITVE

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Povzetek: Pri plevenski črnoglavi ovci je po sinhronizaciji gonitve in umetni osemenitvi v obdobju negonitve prišlo do hidrometre. V članku so obravnavani vzroki bolezni, klinični znaki ter ugotovitve, pridobljene s pomočjo ultrazvoka. Analizirani so bili tudi hematološki in krvni biokemijski parametri ter koncentracija progesterona. V spolovilih ovce so bile opazne patološke in histološke spremembe

V opisanem primeru smo pokazali, da hidrometre pri ovci niso spremljali odkloni splošnega kliničnega zdravja ter hematološkega in krvnega biokemijskega profila. Raven progesterona ni ustrezala vrednosti, ki kaže na brejost. Maternična stena je bila močno stanjšana, v maternici je bila prisotna aseptična tekočina ter trajajoče rumeno telesce na enem jajčniku. V steni maternice so bile spremembe razširjene, opazno je bilo povečanje površinskih materničnih žlez.

Za diagnostiko hidrometre pri ovcah priporočamo dva pregleda z ultrazvokom, pri katerih je potrebno biti pozoren na prosto tekočino znotraj maternice, stanjšanje maternične stene, neopaznost zarodka in placentomov.

Ključne besede: ovca; hidrometra