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Research Article

VAGINAL EXFOLIATIVE CYTOLOGY DURING NORMAL AND INDUCED ESTRUM IN NON-DESCRIPT GOATS (*CAPRA HIRCUS*) - A COMPARATIVE STUDY

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ABSTRACT

The study was conducted in does maintained at the Goat Breeding unit of Post Graduate Research Institute in Animal Sciences, Kattupakkam. Thirty parous, healthy and cycling does weighing 20-30 kg were utilized for this study. Transrectal ultrasonography was carried out to eliminate pregnancy and to assess the ovarian status of non-pregnant does. Does with evidence of corpus luteum in the ovary indicating cyclicity were randomly divided into three groups of ten each. The does in group I were subjected to Vaginal Exfoliative Cytology (VEC) once in two days until the onset of estrus by visual observation and thereafter twice daily till the end of estrum. In group II, Does were administered with a single total dose of 125 µg of Inj. Pragma, intramuscularly and VEC was done in all the does from the start of treatment twice daily until the end of estrum. In group III, Does were subjected to intravaginal insertion of TRIU C® and retained in situ for seven days and VEC was performed at the time of insertion. One day before the withdrawal (6th day), an intramuscular injection of 50 µg of Inj. Pragma was administered and on the seventh day, TRIU C® was removed, VEC was performed using Leishman stain starting from the time of withdrawal of the device twice daily until the end of estrus and the results are discussed.

Keywords: Estrus cycle, Estrus Synchronization, Goat, Vaginal exfoliative cytology

Success of Estrus Synchronization (ES) and Artificial Insemination (AI) in goats depends upon the timely detection of estrus. Vaginal cytology is a method used for the detection of the estrus, assisting in the artificial insemination in goats. In the normal cycling female livestock, morphologic, endocrine and secretory changes occurring in the ovaries and the tubular genitalia during the oestrous cycle usually depict stage of the cycle. These changes have been associated with levels of steroid sex hormones. In the absence of infections, the circulating levels of progesterone and oestradiol–17β are the major determinants of the cytology pattern of the vagina (Rahman *et al.*, 2008). Exfoliated cells in the vaginal lumen are the result of rising peripheral oestrogen which causes the vaginal wall to thicken. As the outermost layer moves further from the vascular supply, the cells keratinize and detach from the wall. Thus, the exfoliated cells are a normal occurrence during the oestrous (and menstrual) cycle of animals (and women). What has been found very useful is the morphology of the exfoliated cells, which has been utilized to determine the physiological and pathological status of the female animal as well as a tool for hormonal bioassay in several animal species (Selvaraju, 1994). However, there is meager information about vaginal exfoliative cytology in goats. Therefore, the present study has been designed to study the usefulness of vaginal exfoliative cytology for detection of estrus in goats.

MATERIALS AND METHODS

Experimental animals : The study was conducted in does maintained at the Goat Breeding unit of Post Graduate Research Institute in Animal Sciences, Kattupakkam. Thirty parous, healthy and cycling does weighing 20-30 kg were utilized for this study. All the does were maintained on uniform management with concentrate diet and 8 hours of grazing.

Ultrasonography: Transrectal ultrasonography using B-mode, Real time scanner (DP-2200 Vet, Shenzhen Mindray Biomedicals Ltd) equipped with 7.5 MHz linear transducer was carried out to eliminate pregnancy and to assess the ovarian status of non-pregnant does (Fig. 1). The flexible probe was made rigid using polyvinyl chloride pipe for better visualization of reproductive tract. The urinary bladder was taken as the acoustic window for viewing the ovaries. The ovaries were visualized cranial to the urinary bladder. Does with evidence of corpus luteum in the ovary indicating cyclicity were randomly divided into three groups of ten each. The does in group I, were subjected to Vaginal Exfoliative Cytology (VEC) once in two days until the onset of estrus and thereafter twice daily till the end of estrum. In group II, does were administered with a single total dose of 125 micrograms of PGF2α analogue, cloprostenol (Inj. Pragma, INTAS Pharmaceuticals Ltd.) intramuscularly and VEC was done in all the does from the start of treatment twice daily until the end of estrum. In group III, does were subjected to intravaginal insertion of progesterone device TRIU C® containing 160 mg of progesterone and retained in situ for seven days. At the time of insertion, VEC was done. One day before the withdrawal (6th day), an intramuscular injection of 50 µg of PGF2α analogue, cloprostenol was administered and on the seventh day, TRIU C® was removed, VEC was done starting from the time of

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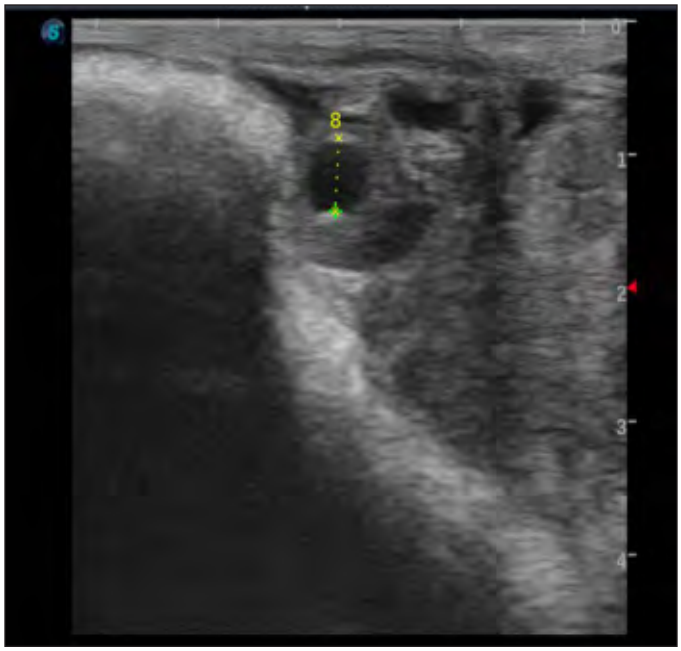

Fig. 1. Ultrasonographic View: Ovary with follicle

withdrawal of the device twice daily until the end of estrus.

Vaginal Exfoliative Cytology: The vulva was rinsed with normal saline and dried with a clean towel. Each doe was restrained in standing position by an assistant and the swab moistened with normal saline was gently inserted with the right hand while the left thumb and fore-finger were used to expose the vulvar lips. The swab was gently rolled in the caudal vagina and carefully withdrawn. The smear was prepared by rolling the swab on a glass slide and air dried. The smears were then stained with Leishman stain using standard protocol. One hundred cells were counted from random fields of each slide and the type, percentage of cells present during different stages of estrous cycle was recorded (Leigh *et al.*, 2010).

Heat detection and breeding: In all the three groups, the physiological signs and behavioural signs and VEC were used to identify the does in estrus which were bred naturally two times at an interval of 24 hours.

RESULTS AND DISCUSSION

In normal estrus does i.e. Group I, the VEC showed only 10 per cent intermediate cells on the day of onset of estrum and disappeared on day two. Then it showed a gradual increase up to 40 per cent towards the end of estrum. The superficial intermediate cells gradually decreased from 60 per cent from the day of onset of estrum to less than 10 per cent at the end of estrum. The superficial cells gradually increased from 20 per cent from the day of onset of estrus to reach a peak of 80 per cent on day 2 and decreased to less than 10 per cent towards the end of estrum. The cornified cells gradually increased from 10 per cent from the day of onset of estrum to 30 percent on day 2 and subsequently increased to 60 per cent towards the end of estrum. Similar distribution pattern of the vaginal exfoliated cell was also observed in group II does.

The intermediate cells were 10 per cent on the day of onset of estrum and showed a gradual decrease and remained at less than 10 per cent till the end of estrum. The superficial intermediate cells gradually decreased from more than 60 per cent from the day of onset of estrum to less than 10 per cent at the end of estrum. The superficial cells gradually increased from 10 per cent from the day of onset of estrus to reach a peak of 60 per cent on day 2 (morning) and decreased to less than 20 per cent towards the end of estrum. The cornified cells increased from 10 per cent on the day of onset of estrum to reach a peak of 80 per cent which continued to remain at more than 60 per cent till the end of estrum. Vaginal exfoliative cytology in the present study revealed a gradual increase of superficial cells from 20 per cent on the day of onset of estrus to reach a peak of 80 per cent on day 2 and then decreased to less than 10 per cent towards the end of estrum in group II. On the contrary, in group III the cornified cell (anuclear superficial cells) were found to increase from 10 per cent on the day of estrum to 80 per cent on day 2 and subsequently shows a decline toward the end of estrum. The peculiarity observed in vaginal exfoliative cytology of goats was the presence of 10-15 per cent of neutrophils during day 2 in all the groups (Fig. 2). The similar pattern of cell distribution in vaginal exfoliative cytology was also observed by earlier workers in goats (Safiriyu *et al.*, 2006). Pretorius (1977) reported that cytological changes in vaginal epithelium could be used to assess ovarian estrogenic activity and desquamation of vaginal epithelial cells reached a high level during late estrus when large numbers of cornified cells occurred in the smear.

The vaginal exfoliative cytology in the present study was used to clearly identify estrum which also corroborated with behavioural and physiological signs of estrum exhibited by the doe. VEC can be used as a complementary method for estrus detection and to monitor ovarian activity. The appearance of neutrophils in the

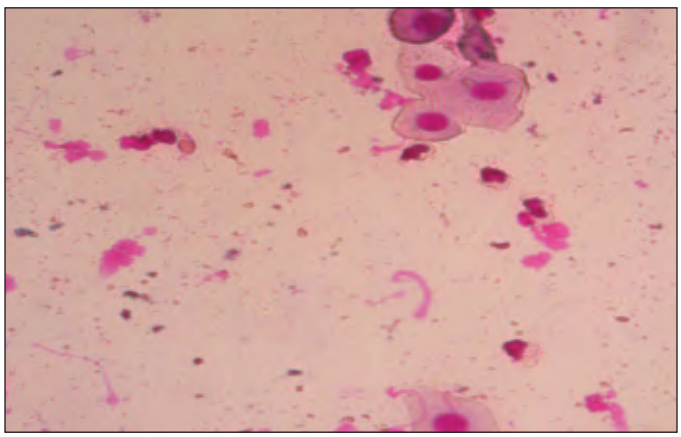

Fig. 2. Superficial cells with neutrophils

estrum smear observed in the present study may be due to a physiological response following mating to ward off possible infection as reported by Leigh *et al*. (2010).

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