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Reproductive Hormonal profile in NARI Suwarna ewes at different stages of pregnancy and diestrus stage

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ABSTRACT
The present study was undertaken to assess the reproductive hormonal profile in NARI Suwarna ewes at different stages of pregnancy and diestrus stage of estrous cycle. Around two to four year aged naturally mated ewes at second month of pregnancy were selected. The pregnancy was confirmed by using trans-rectal and trans-abdominal ultrasonography based on foetal head diameter, anechoic embryonic vesicle, fetal heart beat, gestational sac diameter and placentomes. The animals were categorized in to five groups including control. Ewes (n=7) after the synchronization treatment came to estrous stage within 12 hours and followed by diestrus stage served as control group (Group I). These pregnant ewes were subjected to the longitudinal study which were designated as Group II (second month of pregnancy), Group III (third month of pregnancy), Group IV (fourth month of pregnancy) and Group V (fifth month of pregnancy) as the pregnancy advanced for these seven pregnant ewes. Serum hormonal profile like progesterone, estradiol and prolactin were determined by commercially available ELISA kit. The serum concentration of these hormonal concentrations were significantly (P ≤0.05) increased as pregnancy advanced. From the results of the present study it was concluded that the reproductive hormones like progesterone, estradiol and prolactin concentrations change during different physiological stages. The increased concentrations are necessary to bring developmental changes like preparation of uterus for implantation, growth and development of embryo and fetus, maintenance of pregnancy and birth of healthy lambs.
Key words : NARI Suwarna ewes, pregnancy, progesterone, estradiol and prolactin.

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INTRODUCTION

Sheep husbandry has been a traditional occupation and profitable backyard profession in Indian subcontinent. At present there is more demand for high productive sheep breeds. To fulfil this demand and to enhance fertility rate of local sheep breeds of Maharastra Dr. B. V. Nimbkar has evolved NARI Suvarna breed. It is the cross of Deccani (90%) and high prolific breed Garole (10%) or Deccani (60%), Madgyal (30%) and Garole (10%) [2]. This breed is known for multiple lambing, good mothering ability and play important role in improving economic status of the poor farmers. The information on reproductive hormonal level would be useful for pregnancy diagnosis, to know the accurate gestational age, number of foetuses and for nutritional management of the ewes to avoid pregnancy toxaemia and effective flock management and segregation of pregnant and non pregnant ewes to reduce the production losses. The concentration of reproductive hormones such as progesterone, estrogen and prolactin varies with the physiological stage of the animal to co-operate and support for the particular need of the stage of gestation. The perusal of literature revealed scanty information on reproductive hormonal profile of NARI Suvarna ewes. Therefore, the present study was undertaken to establish the various reproductive hormonal profile at different physiological stages of the NARI Suvarna ewes.

MATERIALS AND METHODS

The present study was conducted to estimate the components of reproductive hormonal profile in non-pregnant and pregnant healthy NARI Suwarna ewes maintained at the Department of Veterinary Gynaecology and Obstetrics, Veterinary College, Hebbal, Bengaluru, Karnataka, under Rashtriya Krishi Vikas Yojana (RKVY) project. Seven healthy non pregnant ewes were selected and subjected to estrus synchronization by using intra-vaginal progesterone sponge for 11 days. After removal of sponge,

pregnant mare serum gonadotropin 150 IU per animal and 75 microgram of cloprostenol per animal was given by intramuscular route. These seven clinically healthy non-pregnant NARI Suwarna ewes which were in diestrus stage of estrous cycle were selected to serve as control group (Group I). Seven naturally mated animals were selected and different stages of pregnancy was confirmed by using trans-rectal and trans-abdominal ultrasonography based on foetal head diameter, anechoic embryonic vesicle, fetal heart beat, gestational sac diameter and placentomes. Seven clinically healthy pregnant NARI Suwarna ewes which were at second month of pregnancy were selected for the study. These pregnant ewes were subjected to the longitudinal study which were designated as Group II (second month of pregnancy), Group III (third month of pregnancy), Group IV (fourth month of pregnancy) and Group V (fifth month of pregnancy) as the pregnancy advanced for these seven pregnant ewes. Five ml of blood samples were collected from seven non pregnant ewes at diestrus stage (Group I). In pregnant animals, the blood samples were collected at monthly intervals from second month of pregnancy to fifth month of pregnancy to serve as different groups in the present longitudinal study. The samples were collected during morning hours between 9.30 and 10.30 AM before they were allowed for grazing. The samples were processed for the separation of serum by centrifugation. Separated serum samples were stored at -80 °C for analysis of certain reproductive hormonal profile. All the hormones were analysed with the help of commercially available ELISA kits. The values were expressed as Mean ± Standard Error and the data obtained in the present study were subjected to statistical analysis by using the GraphPad Prism Version (2007), by applying one way ANOVA with Tukey's post test. The level of significance or non-significance was determined at P value of 0.05.

RESULTS AND DISCUSSION

Significantly ($P<0.05$) higher serum progesterone levels were observed in pregnant groups compared to non-pregnant group(Table 1). Further, the serum progesterone levels in mid and late pregnancy were significantly ($P<0.05$) higher compared to early stage of pregnancy. The findings of the present study were in conformity with Hamon and Heap [9] in Barbary sheep, Mugerwa and Viviani [14] in Menz sheep, Sawada *et al.* [16] in goats, Nessim *et al.* [18] in Egyptian ewes, Alwan and Ibrahim [1] in ewes, Eastwood *et al.* [6] in Romney ewes, Talebi *et al*. [22] in Markhoz does, Debbarama *et al.* [5] in Osmanabadi goats, Shakir *et al.* [19] in Kari ewes, Nawito *et al*. [15] in sheep and goat, who recorded increased serum progesterone concentration with advancement of the pregnancy in different breeds of ewes and goats. The increased progesterone during pregnancy was attributed to secretion of progesterone from corpus luteum, adrenal gland and majority of it is secreted by placenta. The serum estradiol levels during late gestation were significantly ($P<0.05$) higher compared to early and mid-pregnancy and non-pregnant ewes. These findings were in agreement with Thompson and Wagner [23], Kandiel *et al.* [10], Mondal *et al*. [13], Kumar *et al.* [12], Sharma *et al*. [20] who reported increased estradiol levels during late pregnancy in ewes, goats, black Bengal goats and Himalayan Gaddi sheep, respectively. Estradiol concentration was increased abruptly five days before parturition in goats [17] which could be attributed for the synthesis of oxytocin receptors in myoepithelium that prepares for easy parturition upon action of oxytocin. In the present study, significantly ($P<0.05$) higher levels of estradiol in NARI Suwarna ewes during late gestation were in concurrence with the Bazer *et al.* [4] who hypothesized that the estrogen receptors expression will increase as the pregnancy advanced. Further, the findings of the present study were in accordance with Sharma *et al*. [20] who concluded that the fetal cortisol stimulates estrogen production during last three weeks of gestation and Ashmawy [3] who related increased estrogen levels during late pregnancy to the secretion of prostaglandin $F_{2\alpha}$ which is required for interrupting progesterone secretion. Goff [8] who reported that the increased estradiol levels in ruminants at pregnancy stimulate prolactin production from anterior pituitary, prolactin receptor synthesis in the mammary cells, growth of uterus and mammary gland for successful reproduction. The significantly increased levels of estradiol recorded in the present study during late gestation could be attributed to demand for estradiol for the rapid growth of mammary gland during last stage of pregnancy and to help in the initiation of parturition.

The serum prolactin levels in late pregnant ewes were significantly ($P<0.05$) higher compared to other groups. These findings were similar to the findings of Kendall [11] who opined that in Romney ewe maternal plasma prolactin levels remained low and relatively stable throughout most of pregnancy and increased rapidly during last month of gestation and it was reached maximal levels around the time of parturition. Increase in prolactin during late gestation could be attributed to altered responsiveness of pituitary leading to change in the pattern of prolactin secretion. Further, higher level of prolactin in ewes could also be due to higher levels of thyrotropin releasing hormone during late gestation [7]. The significantly increased levels of prolactin in the present study, where it was stated that the prolactin is

necessary for growth and development during pregnancy, required for various aspects of reproduction, mammogenesis by way of renewal of alveoli, lactogenesis and galactopoiesis [21].

CONCLUSION

In conclusion, In the present study, significantly increased progesterone levels were found and significantly increased level of prolactin along with the estradiol was observed during fourth and fifth month of gestation. These hormones are involved in supporting the pregnancy, developing of alveoli the mammary gland, proliferation of cells in the duct system and expression of maternal behaviour. Further, the measurement of these hormones will help to diagnose the pregnancy in NARI Suwarna ewes.

Table. 1. Reproductive hormonal profile in NARI Suwarna ewes at different stages of gestation and diestrus period (n=7).

Sl No.	Parameters	Group I	Group II	Group III	Group IV	Group V
1	Progesterone (ng/ml)	1.90 ± 0.24^{a}	14.26 ± 1.03^{b}	33.63 ± 4.16^{c}	34.13 ± 3.52^{c}	42.20 ± 1.56^{c}
2	Estradiol (pg/ml)	5.04 ± 0.50^{a}	53.10 ± 5.03^{a}	76.91 ± 3.98^{a}	267.40 ± 17.24^{b}	609.90 ± 50.60^{c}
3	PRL (ng/ml)	0.58 ± 0.14^{a}	3.77 ± 0.33^{a}	14.09 ± 1.5^{ab}	49.96 ± 4.60^{b}	151.20 ± 21.42^{c}

Note : The values with different superscripts within row differ significantly ($P<0.05$).

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