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Review Article

OVARIAN BIOMETRY WITH ROLE AND DISTRIBUTION OF TRACE ELEMENTS IN OVARIAN FOLLICULAR FLUIDS OF GOAT: AN OVERVIEW

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Follicular fluid is an avascular compartment within the mammalian ovary, separated from the perifollicular stroma by the follicular wall that constitutes a 'blood-follicle-barrier' and also maintains proper environment for growth and maturation of the oocyte. It plays a role in the physiological, biochemical and metabolic aspects of the nuclear and cytoplasmic maturation of the oocyte. Information on the biometrics of the reproductive system of livestock animal is necessary in order to improve the fertility, the reproductive performance as well as enabling the adoption of other assisted reproductive technologies. Trace elements are highly essential for health and immunity, and also their contribution to growth, production and reproduction. Their deficiency affects both steroid and thyroid hormone production.

Key words: Blood-follicle-barrier, Follicular fluid, Oocyte, Reproduction, Trace elements

In all mammals follicles begin to grow from a pool of primordial follicles constituted early in life, continuously throughout the life of the female. Primordial follicles are the least developed and more numerous follicles of the ovary. Each consists of primary oocyte surrounded by a layer of simple squamous follicular cells which in late stages develop to primary, secondary and graaffian follicles respectively (Banks, 1993). Follicular fluid is an avascular compartment within the mammalian ovary, separated from the perifollicular stroma by the follicular wall that constitutes a 'blood-

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follicle-barrier' (Abd Ellah *et al.*, 2010 and Albomohsen *et al.*, 2011). Besides meeting nutritional requirement of the growing oocyte, follicular fluid also maintains proper environment for growth and maturation of the oocyte. Besides a transudate of serum, follicular fluid is partially composed of locally produced substances, which are related to the metabolic activity of follicular cells (Gerard *et al.*, 2002). This metabolic activity, together with the 'barrier' properties of the follicular wall, is changes significantly during the growth phase of the follicle (Bagavandoss *et al.*, 1983; Wise, 1987 and Gosden *et al.*, 1988). Therefore, it is expected that biochemical composition would vary with size of follicles. Follicular fluid contains specific constituents such as steroids and glycoprotein synthesized by the cells of the follicle wall. It plays a role in the physiological, biochemical and metabolic aspects of the nuclear and cytoplasmic maturation of the oocyte (Hafez and Hafez, 2000). The constituents of follicular fluid are considered as a regulating factor in follicular development and steroidogenesis. Mc Gauphey (1972) suggested that the follicular fluid contents vary prior and after oocyte maturation and it might be related to meiotic process. Follicular fluid composition was under intensive investigation to know the oocyte maturation (O' Callaghan *et al.*, 2000), fertilization and embryonic development (Choi *et al.*, 1998) and follicular atresia (Lebedeva *et al.*, 1998). Chang *et al.* (1976) reported that the fluid in ovarian follicles has long been suspected of playing role in differentiation and steroidogenic capacity of ovarian cells and the regulation and maturation of oocyte.

Information on the biometrics of the reproductive system of livestock animal is necessary in order to improve the fertility, the reproduction and the performance as well as enabling the adoption of other assisted reproductive technologies (Okoye *et al.*, 2017). To maintain a good reproductive performance a clear idea about the reproductive organs of small ruminant is necessary. The biometry of genital tracts of the female reflects the overall well being of the animals and is essential to perform artificial insemination, pregnancy diagnosis and dealing with the infertility problems along with treatment (Kumbhar *et al.*, 2003 and Kumar *et al.*, 2004). For successful *in vitro* production (IVP) of embryos, evaluation of ovaries, efficient collection and grading of oocyte is very important (Gupta *et al.*, 2011).

Trace elements are required in small quantities as low as 100mg/kg dry matter which are usually available in feeds and fodders and are present in very minute quantities in animal serum usually less than 2 ppm (McDowell, 1992 and Suttle, 2010). Though required in small amounts they are highly essential for health and immunity and also contributing to growth, production and reproduction (Boland, 2003; Andrieu, 2008; Siciliano-Jones *et al.*, 2008; Spears and Weiss, 2008 and Hesari *et al.*, 2012). Arshad *et al.* (2005) also stated that minerals are essential for growth and reproduction and are involved in large number of digestive, physiological and biosynthetic processes within the body. Ovarian activities of ruminant are influenced by mineral deficiency and are also involved in hormones synthesis that is important for reproduction. Their deficiency affects both steroid and thyroid hormone production (Boland, 2003 and Abdollahi *et al.*, 2013). Most of the non conventional feeds are deficient in micro minerals and are likely to accentuate reproductive problems (Parnekar, 2003).

Biometry of ovary

The ovary is a very dynamic tissue and the dimension is greatly influenced by structures present on it (Noakes, 2001). Goat ovaries are almond-shaped, paired, and located on each side of the pelvic cavity. They produce the ova or female gametes and sex steroid hormones such as estrogens and progesterone. The follicle may be regarded as the functional unit of the ovary, consisting of both germ cells and endocrine cells. The mature ovary at any time contains varying numbers of follicles in various stages of development. This has prompted many investigations on the growth, morphology and morphometry of ovarian follicles (Cahill *et al.*, 1979 and Ireland, 1987), and changes during the stages of the oestrous cycle (Ireland *et al.*, 1979 and Ravindra *et al.*, 1994). Jaji *et al.* (2012) found that the diameter and thickness of the right ovary showed extremely significant increase ($p<0.01$) during the 14-20 weeks of gestation. Conclusively, in the Sahelian goat, pregnancy does not seem to have significant effect on the dimensions and weight of the left ovary to full term but causes extremely significant increases in the diameter and thickness of the right ovary during the 14-20 weeks of gestation (Jaji *et al.*, 2012). The diameter and thickness of the right ovary is significantly more than those of the left at the 14-20 weeks of gestation. The gravid and non-gravid right ovaries are larger in dimensions and heavier in weight as compared to left ones, which confirming the fact of right ovary being more active than the left one. In gravid doe-goats, the dimension of the ovary is influenced mostly by the corpus luteum as pregnancy anaestrous puts a negative feedback blocking progesterone and inhibiting the development of ovarian follicles to preovulatory follicles (Noakes, 2001). Furthermore, in the doe goat the corpus luteum may be protruding or embedded in the ovary to varying degrees (Rowell *et al.*, 1993) showing the variability in ovarian dimensions. Adigwe and Fayemi (2005) worked on Marodi goats of Nigeria and found that the lengths of the left and right ovaries were 1.71 ± 0.27 cm and 1.73 ± 0.27 cm respectively and the diameters of the left and right ovaries were 0.65 ± 0.15 cm and 0.66 ± 0.22 cm, respectively. Sangha *et al.* (2006) reported that average length, width and thickness based on the study of 373 goat ovaries were 1.575 ± 0.017 cm, 1.17 ± 0.01 cm and 0.69 ± 0.009

cm, respectively. Majority of the ovaries had the ovarian length between 1.1 to 1.5 cm and width 0.9 to 1.1 cm and thickness 0.6 to 0.7 cm. Islam *et al.* (2007) demonstrated that in different categories of ovaries, the mean weight, length and width were distinctly higher in right ovaries 0.66 ± 0.02 gm, 1.17 ± 0.02 cm and 0.77 ± 0.02 cm, respectively compared to that of left ovaries 0.64 ± 0.02 gm, 1.11 ± 0.02 cm and 0.74 ± 0.02 cm, respectively. According to these workers, the mean weight was significantly higher ($P<0.05$) and width was comparatively higher in the ovaries with corpus luteum than those of ovaries without corpus luteum, while the mean lengths were found higher in the ovaries without corpus luteum. Miranda-Moura *et al.* (2010) showed that the average length of right ovary in goat was 1.64 ± 0.21 cm in the 2nd day, 2.02 ± 0.16 cm in the 12th day, 1.90 ± 0.31 cm in the 16th day of post ovulation, whereas in left ovary it was 1.75 ± 0.27 cm, 1.96 ± 0.11 cm, 1.80 ± 0.25 cm in the similar days of post ovulation, respectively. The mean diameter of ovulatary follicles in Najdi goats (Mohammadi *et al.*, 2010) is 6.3± 0.13mm and was almost similar to White Polish (Schwarz and Wierzchos, 1999) and Serrana goats (Simoes *et al.*, 2005). Okoye *et al.* (2017) showed that there was no significant variation across the term of gestation except in the right ovary where its length during the third term of gestation was significantly higher than that of first term. Schwarz and Wierzchos (2010) demonstrated that the mean numbers of growing and large follicles on the corpus luteum bearing ovary were significantly lower than those on the corpus luteum free ovary. Gupta *et al.* (2011) stated that the differences between the different studies in goats might be due to breed differences. Differences in size of reproductive tract may also be due to climatic effects as young goats in the tropics have to contend with the effects of the first dry season when growth may be seriously retarded. Similarly, in Black Bengal goat (Singh *et al.*, 1974; Rahman *et al.*, 1977 and Dalai *et al.*, 2014) found that the mean weight, length and width of right ovaries are higher in comparison to left ovaries. Hence it can be assumed that right ovaries are more active in Black Bengal goat.

Role and distribution of trace elements in follicular fluid

Minerals are essential for growth and reproduction and are involved in a large number of digestive, physiological and biosynthetic processes within the body (Close, 1998). The most obvious function is as components of body organs and tissues and to provide structural support. In addition, they act as electrolytes, as constituents of body fluids and as catalysts in both enzyme and hormone systems and fulfil several important functions for the maintenance of animal growth and reproduction as well as health status (Underwood, 1981). The requirements of trace mineral in animals are variable and depend on age (Devi *et al.*, 2011), sex

(Yatoo *et al.*, 2012), stage of growth or production (NRC, 2001), breed and genotype (Waldroup, 2001 and Lukic *et al.*, 2009).

Among all trace elements the importance and role in reproduction and patterns of distribution among blood, ovarian tissues and follicular fluid of zinc, iron, copper and manganese are discussed here:

Zinc (Zn): Zinc plays a major role in the immune system and certain reproductive hormones (Capuco *et al.*, 1990). Zinc is known to be essential for proper sexual maturity, reproductive capacity, and more specifically, onset of estrus. Zinc has a critical role in the repair and maintenance of the uterine lining following parturition, speeding return to normal reproductive function and estrus (Goff, 1999). Zinc deficient animals have been shown to have lower concentrations of FSH and LH chiefly in males (Boland, 2003). The higher concentrations of Zn and Fe in small follicle as compared to large follicles were reported by Nandi *et al*. (2012) and Bordoloi *et al.* (2001). These variations are attributed to increased levels of steroid hormones (estrogen and progesterone) that induce increased hemodynamic pulses in the vascular shunt of the developing follicles (Sangha *et al.*, 1999). In ovulatory follicles, the declined iron level might be due to ischemia leading to rupture of follicle wall at stigma (Sharma and Vats, 1998). Dalai *et al.* (2014) also reported that the concentration of zinc decreased significantly in large follicles as compared to small and medium follicles and also noticed higher concentration of zinc in ovarian tissues compared to serum. The concentration gradient of zinc between serum and ovarian tissues might be due to an active inward transport of this cation for ovarian development by synthesizing Zn-binding protein and metalloenzymes (Jeckel *et al.*, 1996). Higher Zn concentration in small follicle than that of medium and large one might be indicating the essentiality of this element to the initial maturation of graffian follicle (Bordoloi *et al.*, 2001).

Iron (Fe): Khilare (2007) stated that iron is required for the synthesis of haemoglobin and myoglobin as well as many enzymes and cytochrome enzymes of electron transport chain. Iron functions in transport of oxygen to tissues, maintenance of oxidative enzyme system and is concerned with ferretin formation. A deficient animal becomes repeat breeders and require increased number of inseminations per conception and occasionally may abort due to anemia, reduced appetite and lower body condition. Dalai *et al.* (2014) found higher iron content in ovarian tissue as compared to serum as well as different types of ovarian follicles. Significantly higher concentration of iron in ovarian tissue might be due to its continuous mobilization against the concentration gradient for ovarian development. The role of iron in oxidative metabolism could explain its accumulation in ovaries and further transfer to eggs to sustain metabolic processes during oocyte growth and embryogenesis (Mendez *et al.*, 2001).

Manganese (Mn): Manganese appears to have a vital role in reproduction. It is necessary for cholesterol synthesis (Kappel and Zidenberg, 1999), which in turn is required for synthesis of the steroids, estrogen, progesterone and testosterone. Moreover reduced reproductive efficiency encountered loss of appetite due to magnesium deficiency (Kumar, 2003). A deficiency in manganese may be associated with suppression of oestrus, cystic ovaries and reduced conception rate (Patterson *et al.*, 2003). Even Manganese deficient goats were observed to exhibit no apparent sign of oestrus despite normal ovulation (Groppel and Anke, 1971). In males, the dietary deficiencies of manganese, leads to absences of libido, decreased motility of spermatozoa and reduced number of sperms in ejaculate (Kumar, 2003). Increase in concentration of Mn in follicular fluid along with increase in size of follicles has been reported by Dalai *et al.* (2014) and Bordoloi *et al.* (2001) which might be due to involvement of this ion in major energy producing reaction (Sikka, 1992).

Copper (Cu): Reproductive problems that relate to copper deficiency manifest themselves in inhibited conception rate even though estrus may be normal. Symptoms of a copper deficiency include early embryonic death, resorption of embryo, increased retained placentas and necrosis of the placenta (Patterson *et al.*, 2003). Dairy cows with higher serum copper levels had significantly less days to first service, fewer services per conception and fewer days to open (Jousan *et al.*, 2002). Proper copper supplementation of the sire is needed for production of quality semen (Patterson *et al.*, 2003). Copper is a mineral element that activates several enzyme systems, and though in less numbers than Zn, it is considered an essential nutrient (Minatel and Carfagnini, 2007). However, sheep and goats are not tolerant to high Cu levels in their diets, and it is thus considered a toxic element (Minson, 1990; McDowell, 2003 and NRC, 2005). The result obtained by Dalai *et al.* (2014) showed no significant difference in copper concentration among serum, ovarian tissue and follicles of Black Bengal goats while higher copper level in large follicles was found than that of medium ones which might be due to its some role in final maturation of oocyte.

In summary, to maintain a good reproductive performance a clear idea about the reproductive organs of small ruminant is necessary. The biometry of genital tracts of the female reflects the overall well being of the animals and is essential to perform artificial insemination, pregnancy diagnosis and dealing with the infertility problems. Most of the animals showed deficient serum mineral status. Mineral deficiency affects hormone status and impairs production potential of animals. Adequate minerals supplementation is required as most of the roughages, greens, concentrate and even most of commercial feeds are deficient in trace mineral elements. They are essential for functioning of a number of components of the immune system. They act as cofactors for a number of enzymes and proteins which are involved in many physiological and biochemical processes which are related to growth, production and reproduction. Cu, Fe, Mn and Zn directly affect reproductive events on goats, they directly influence events such as, expression of estrus, embryo implantation and reduction in spermatogenesis; indirectly, they affect overall animal health. Mineral mixture containing zinc, iron, manganese, copper and other trace minerals are recommended in the nutritional management of animals.

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