

Sex differentiation in goat fetus

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Summary

Reproduction in domestic animals, as a major source of food and other products for human, has great importance and study of related subjects including sex differentiation and gonadogenesis during fetal life can solve many questions on normal development and various disorders of urogenital system. Since studies on sex differentiation in goat fetus are scarce, this study was performed. Twenty-five goat fetuses with 5–40 mm crown-rump lengths (CRL) were obtained from slaughter-house and fixed in 10% formalin solution immediately. The development and features of external genitalia of these fetuses as well as their sex differentiation were then examined. In externally indifferentiated fetuses, the cranial half of the body was cut away at the diaphragmatic level and serial sections, 6-micron thick, were prepared and the ovarian and testicular differentiation was studied. Results showed that considering the development of the external genitalia, external sex differentiation occurs at 31 mm CRL in male fetuses and at 33 mm CRL in female fetuses. Microscopic studies suggested that testicular tissue differentiation occurs at 23 mm CRL with formation of tunica albuginea and medullary cords. Ovarian tissue differentiation occurs at 38 mm CRL with establishment of cortical and medullary regions.

Key words: Sex differentiation, Fetus, Goat

Introduction

In many parts of the world, goats are raised for production of milk, meat, hide and down. Goats have high adaptability and can live in different climates. During recent decades, universal concentration on goats has increased considerably and new initiatives have been starting for development of related industries. In Iran, traditional goat rearing is customary because of low maintenance costs and adaptation with different climatic conditions. Therefore, conduction of basic and applied research regarding various aspects of goats seems necessary.

Sex differentiation has been studied in human (Baker and Scrimgeour, 1980), swine (Inomata *et al.*, 1993), dog (Evans, 1979), bovine (Erickson, 1966), buffalo (Ghannam and Deeb, 1967, 1969; Ranjbar, 1998) and horse (Sakai, 1955). However, only one study on ovarian differentiation in goat fetus was reported. (Harshan *et al.*, 1994). Because there is no comprehensive study on

external and gonadal sex differentiation in goat fetus and in view of the fact that studies on various aspects of Iranian native goats including fetal development are necessary, the present study was conducted to determine sex differentiation in goat fetus.

Materials and Methods

Twenty-five pregnant goat uteri in early stages of pregnancy were collected from a slaughter-house in Ahvaz, southern Iran. Fetuses were expelled after splitting of uterus and separating of fetal membranes. crown-rump lengths (CRL) of fetuses were measured so that the minimum CRL was 5 mm and the maximum CRL was 40 mm. Approximate ages (y) of fetuses were estimated using the CRL of fetuses (x) according to the following equation: $y = 2.74x + 30.15$, presented by (Gall *et al.*, 1994). Fetuses were then fixed in 10% formalin solution and were examined macroscopically to study the growth of external genitalia. In male fetuses, presence

of long anogenital raphe between anus and genital tubercle, and existence of urogenital orifice; and in female fetuses, absence of anogenital raphe and urogenital orifice, growth of vulvar labia and posterior direction of clitoris were considered as the criteria for sexual differentiation. In fetuses with CRL less than 30 mm, the cranial half of the body at diaphragmatic level was cut away. Then, serial sections, 6-micron thick, were prepared from the rest of the body and after staining with haematoxylin and eosin, their gonadal differentiation were studied microscopically. Formation of tunica albuginea and medullary cords in testis and formation of cortical and medullary regions in ovary were considered as criteria for

gonadal differentiation.

Results

We found that goat fetuses were macroscopically sexually undifferentiated externally until 31 mm CRL, both sexes have genital tubercle and cloacal folds. Likewise, at the posteriolateral of cloacal membranes, there were genital or labioscrotal swellings. Male goat fetuses differentiated at 31 mm CRL and had long anogenital raphe between the anus and base of genital tubercle and also urogenital orifice (Fig. 1). In female goat fetuses, external sexuality differentiation occurred at 33 mm CRL with formation of vulvar labia, absence of urogenital orifice and anogenital raphe, as

Fig. 1: Anogenital raphe of male goat fetus with 31 mm CRL (arrow), (×20)

Fig. 2: Vulvar labia of the female goat fetus with 33 mm CRL (arrow), (×20)

Fig. 3: Differentiation of testis in fetus with 23 mm CRL. Medullary cords are formed (arrow), (H&E, ×400)

Fig. 4: Differentiation of ovary in fetus with 38 mm CRL. Cortex (arrow) and medulla (arrow head) are formed (H&E, ×100)

well as short distance between anus and base of genital tubercle (Fig. 2). Microscopic studies showed that gonads were not differentiated before 23 mm CRL. However, at this time, in testis, tunica albuginea were formed under the surface of epithelium, and medullary cords were well-established (Fig. 3). Differentiation of ovary was seen at 38 mm CRL with formation of clear cortex containing cortical (secondary) cords and medulla having blood vessels and connective tissue (Fig. 4).

Discussion

Mammalian sex determination proceeds

in three distinct phases. In the first stage, genetic sex is determined at the time of fertilization by the chromosomal complement of the fertilizing spermatozoon. Later, during embryonic development, this genetic information is translated into gonadal sex that determines the growth of either a testis or ovary from a bipotential early indifferent gonad. The third stage which is the phenotypic sex determination, begins in fetal or early post-natal life and continues through puberty, a period in which endocrine products of the gonads direct the differentiation of the accessory sex ducts and external genitalia (Loffler and

Koopman, 2002).

External and gonadal sex differentiation has been studied in human and domestic animals with different results.

In male human fetus, at 7th week of fetal life, under the influence of Y chromosome, primary sex cords containing proliferated coelomic epithelium and primordial germ cells continue their proliferation and form medullary (testicular) cords. Synchronously, a thick fibrous layer called tunica albuginea forms due to proliferation of mesenchyme. The female gonads differentiate later than male. Because of the absence of Y chromosome, the primary sex cords degenerate and coelomic epithelium proliferates and form cortical or secondary sex cords that are characteristics of early female gonad (Soleimani Rad, 2000).

The findings of the present study showed that in male goat fetuses, the tunica albuginea and medullary cords are distinctive at 23 mm CRL (age of 36 days). However, tunica albuginea organize in swine at 30 days of fetal life, sheep at 35 and cow at 45 (Noden and Delahunta, 1985).

Testicular differentiation in buffalo occurs in 20–21 mm CRL (approximately at 45 days of fetal life, Ghannam and Deeb, 1967, 1969; Ranjbar, 1998). The first signs of ovarian differentiation in female goat fetuses are distinguishable at 38 mm CRL (age of 40.5 days) by organized cortical and medullary regions and absence of tunica albuginea. However in another study, Harshan *et al.*, (1994) demonstrated that ovarian differentiation occurs at 40 mm CRL. In female sheep fetus, at days of 38, cortical and medullary regions form and their thickness increase gradually (Sawyer *et al.*, 2002). Bascom (1923) reported that in the cow, ovarian cortical cords develop poorly at 25 mm CRL and become better organized at 35 mm CRL. Nonetheless, Sakai (1955) observed the cords only at 150 mm CRL. Differentiation of ovary in buffalo was seen at 23 mm CRL (Ranjbar, 1998).

In ruminants, vulvar labia that form from urogenital folds are equivalent to minor labia in human (Inomata *et al.*, 1982). In swine, the external sex differentiation has been observed at 25–30 mm CRL (Inomata *et al.*, 1993) and in buffalo at the age of 56 days (Ranjbar, 1998).

The sooner sex differentiation in male than female fetuses is due to the masculinization effects of testis on indifferent urogenital system, ovary does not affect the urogenital system at early stages of development. Finally, comparison of various species shows that many aspects of sex differentiation are similar in goat and sheep.

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