POSTNATAL AGE RELATED HISTOCHEMICAL STUDIES ON THE TESTES IN ASSAM GOAT (CAPRA HIRCUS)

Kamal Sarma*, S.N. Kalita2 and J. Devi3
Department of Veterinary Anatomy, College of Veterinary Science
Assam Agricultural University, Khanapara, Guwahati-781 022, Assam, India

ABSTRACT

The present study was conducted on eighteen male Assam goats ranging in age from day old to 10 months divided into six post natal age groups viz. group-I (0-day), group-II (2 months), group-III (4 months), group-IV (6 months), group-V (8 months) and group-VI (10 months) consisting of three animals in each group. Tissues were processed for paraffin sections. Sections were cut at 5 μ thickness and stained with different stains for demonstration of various histochemical substances. The connective tissue of the capsule and septula testis was weakly reactive to basic proteins in day-old kids and moderately reactive in 2 months old kids (group-II). The nuclei of the spermatogenic cells more particularly of the primary spermatocytes showed a strong reaction for basic proteins which were probably due to their increased activity during meiotic phases of spermatogenesis at 4 to 8 month of age. The basement membranes of the seminiferous tubules were strongly PAS reactive in day-old kids, moderate in 2 months old kids and weak reaction was seen in the older goats, where as the Sustentacular or Sertoli cells showed a very weak reaction in all the age groups under study. The capsule and septulae testes showed a faint reaction for nucleic acids in the male goats in all the age groups. The nuclei of various spermatogenic cells were Fuellgen positive and the intensity of reaction increased with the advancing age of the kids. The connective tissue of the capsule and septula testis showed faint reactions for acid mucopolysaccharides in the male goats of all the age groups. The Leydig cells were also weakly reactive to acid mucopolysaccharides in the kids up to 2 month of age, but showed moderate reactions in older kids.

Key words: Testis, post natal development, histochemistry, Assam goat

Introduction

The economic advancement of early puberty in male is important and the age when animal reaches puberty has a direct effect on the age when it can select for progeny testing. Testis is the main organ of male reproductive system and the testicular parenchyma is composed of seminiferous tubules from which spermatooza are produced that maintain generation and Leydig cells that produce testosterone which is responsible for male sexuality and secondary male sex characteristics (Dellmann and Wrobel, 1998 and Hafez, 2000). Testicular architecture has been disorganized in various diseases involving the gonads such as hypogonadotropic eunuchoidism, Sertoli-cells-only syndrome (Heller and Nelson, 1948). In several cases of testicular failure, the problem may be quantitative only, with a reduction in the number of one or more of the different types of germ cells (Paulsen, 1968). Quantitative testicular histology has been used to determine daily sperm production in the boar (Kennelly and Foote, 1964) and short horn bull (Swienstra,1968).

Post natal anatomical studies on the male genital system at various ages, particularly the testis and its tubular system are important to know the anatomical growth and development. A few anatomical studies on testes are reported in goats viz. post natal development of testes in Malabari goat from 4 to 11 months of age (Bilaspuri and Singh, 1992), biometry of the testes in Sirohi goats (Mishra et al., 1984), testicular measurements in Assam local X Beetal goats (Sarmah et al., 1998) and testicular growth in British Saanen, Alpine and Toggenburg breeds of bucks (Ahmad and Noakes, 1996). Some works were also conducted in other ruminants elucidating morphology and biometry of the testes such as buffalo (Pal and Bharadwaj, 1983) and rams (Sergeev and Zabolotskii, 1976).

This work is the first of its kind reporting the histochemistry of the testis in Assam goat at various ages during post natal development which will provide valuable information to the anatomist, pathologist and theriogenologist.

Materials and Methods

A total of 18 male Assam goats varying in age from 0-day to 10 months were used in the present study. The animals were divided into six age groups viz. group-I (0-day), group-II (2 months), group-III (4 months), group-IV (6 months), group-V (8 months) and group-VI (10 months) consisting of three animals in each group. The age of the goats were estimated from birth records. Each animal was weighed using spring balance to record the body weight. The animals were sedated by giving intramuscular injection of Siquil (Triflupromazine hydrochloride) @ 1 mg/kg body weight and subsequently anaesthetized by administering intravenous injection of Intravel Sodium (pentobarbital sodium) @ 15 mg/kg b. wt. (Hall et al., 2000). After induction of proper level of anesthesia, the animals were sacrificed.

Tissue pieces were collected from three different

1Associate Professor and corresponding author, email: kamalsarma73@yahoo.com; 2Retd. Professor & Head, Division of Veterinary Anatomy & Histology; 3Associate Professor, Division of Veterinary Physiology, F.V.Sc. & A.H., S.K. University of Agricultural Sciences & Technology, Jammu-181 102 (J&K).
regions of the testis *viz.*, upper, middle and lower and subsequently fixed in Bouin’s solution prepared as per (Luna, 1968). All the tissues were processed for paraffin sections (Luna, 1968) by alcohol- xylene method using ceder wood oil. Sections were cut at 5 μ thickness using a rotary microtome (Thermo, Germany) and stained for various stains for demonstration of various histochemical substances *viz.* McManus method for glycogen, Alcian Blue method at Ph 1.0 for acid mucopolysaccharides, Fuelgen reaction for nucleic acids and Mercuric Bromophenol Blue method for Protein (Humason, 1967).

**Results and Discussion**

In the present study, the connective tissue of the capsule and septula testis was weakly reactive to basic proteins in day-old kids (group-I) and moderately reactive in 2 months old kids (group-II). A moderate to strong reaction exhibited by the basement membranes of the seminiferous tubules indicated presence of a good amount of basic proteins at various ages. These findings were in corroboration with the observations of Bordoloi and Dhingra (1983) and Pyne and Sinha (1989) in goats, and Singh (1996) in buffaloes. The nuclei of the spermatogenic cells more particularly of the primary spermatocytes showed a strong reaction for basic proteins which were probably due to their increased activity during meiotic phases of spermatogenesis at 4 to 8 month of age (groups-II to V). Bilaspuri (1978) also reported that the activity of protein in the spermatocytes increased from leptotene to deplotene stages in buffalo. Further, Rajani *et al.* (2001) observed that the reaction for basic proteins in all the tissues of rat testis increased with the advancement of age as also seen in the present study. An weak reaction observed in the nuclei and cytoplasm of the lining epithelium of the tubuli recti and rete testis at various ages in the study might be due to their less activity as compared to the spermatogenic cells of the seminiferous epithelium.

The connective tissue stroma of the capsule of the testes showed weak to moderate PAS reaction in all the age groups. Chandra Pal and Bharadwaj (1989) also reported that the connective tissue fibres of the testes of buffalo calves at different ages exhibited mild to moderate PAS reaction. Again, basement membranes of the seminiferous tubules were strongly reactive in day-old kids (group-I), moderate in 2 months old kids (group-II) and weak reaction was seen in the older goats. The weak reactivity of the basement membranes in older subjects as compared to the younger ones was reported earlier in buffalo calves (Goyal and Dhingra 1973), Black Bengal goats (Pyne and Sinha, 1989) and rat (Rajani *et al.* 2001). The Sustentacular or Sertoli cells showed a very weak reaction in all the age groups under study. As regard to the reactivity of the Sertoli cells to PAS reaction, Goyal and Dhingra (1973) reported that the Sertoli cells completely lacked PAS active materials in the testis of buffalo. On the contrary, Dhingra (1980) found a positive reaction for PAS active carbohydrates in the Sertoli cells of goats. This variation in the reactivity of the Sertoli cells to PAS reaction might be due to breed or species variations.

The Leydig cells showed weak to moderate PAS reaction in the testes of 4 months old (group-III) kids. Similar observations were also made by Karmore *et al.* (2001) stating that the Leydig cells showed mild PAS positive reaction in the
testis of goat. On the contrary, Goyal and Dhinigra (1973) reported that the Leydig cells completely lacked PAS reactive material in buffalo, which might draw the same conclusion of species variation. In this study, the basement membranes of the tubuli recti and rete testis exhibited a moderate reaction in younger kids and weak reaction in older goats. These findings were in conformity with that observed in rabbit (Dhinigra and Barnwal, 1977), goat (Dhinigra, 1989) and buffalo (Chandra Pal and Bharadwaj, 1990).

In the present study, the connective tissue of the capsule and septulae testes showed faint reactions for acid mucopolysaccharides in the male goats of all the age groups as also observed in Black Bengal goats (Pyne and Sinha, 1989). However, Chandra Pal and Bharadwaj (1984) obtained a mild to moderate reaction for acid mucopolysaccharides in the connective tissue stroma of the testes in Indian buffaloes. Again, a faint reaction to acid mucopolysaccharides was seen in the basement membranes, spermatogenic and Sertoli cells of the seminiferous tubules in the male Assam goats. The Leydig cells were also weakly reactive in the kids up to 2 month of age (group-II), but showed moderate reactions in older kids as also reported by Karamore et al. (2001) in goats. In this study, the basement membranes and the lining epithelium of the rete testis and tubuli recti showed weak reactions to acid mucopolysaccharides. On the contrary, Chandra Pal and Bharadwaj (1990) obtained a mild to moderate reaction for acid mucopolysaccharides in the epithelium of the tubuli recti and rete testis in buffaloes.

The capsule and septulae testes showed a faint reaction for nucleic acids in the male goats in all the age groups. The nuclei of various spermatogenic cells were Fuelgen positive and the intensity of reaction increased with the advancing age of the kids. These findings were in conformity with the observations made in buffalo (Chandra Pal and Bharadwaj, 1990), goat (Kakade and Singh 1990) and rat (Rajani et al. 2001). In this study, the fibrocytes and Leydig cells of the intertubular tissue of the testes showed a mild to moderate Fuelgen reaction. Similar findings in regard to the reactivity of the Leydig cells to Fuelgen reaction was also reported in goats (Bordoloi 1979).

References


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