

MORPHOLOGICAL AND HISTOLOGICAL INVESTIGATIONS ON THE ADRENAL GLANDS IN BLACK BENGAL GOAT (*Capra hircus*)

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Abstract

Black Bengal goat is a highly prolific small ruminant well known for tender meat and good quality skin. A plenty of research work have already been conducted on the production performance of this goat breed but the research work to elucidate the histomorphology of different vital organs and glands of this breed is still in scarce. There is a close relation between the productivity and endocrine function. With an aim to study the histoarchitecture of adrenal gland of s/he goats, adrenal gland samples were collected from the government slaughter house of Sylhet, Bangladesh at the time of slaughtering. The differences in size and shape of the gland, the cortico-medullary ratio, cell size and nuclear diameters of the cortical and medullary tissues were studied in both sexes. For the histological examinations, the tissue pieces from adrenal glands were immersed in bouin's fluid. After fixation, tissue samples were dehydrated, cleared, and embedded in paraffin. Haematoxylin and eosin staining method was used to examine tissue sections. The adrenal gland consisted of capsule (3.31% and 2.83%), cortex (70.75% and 72.26%) and medulla (25.94% and 24.91%) in buck and doe. The weight of the left gland was more than the right. Cortical cells were sometimes found in the medullary portion. Medullary epinephrine and norepinephrine cells size and nuclear diameter was more in male but the total length of medulla was more in female. The aim of this study is to provide valuable information for further research on the adrenal gland of goat.

Keywords: Black Bengal goat, adrenal gland, histoarchitecture, cortex, medulla.

Introduction

Black Bengal goats are well-known for their adaptability, high prolificacy, superior chevon and quality skin, although poor milk production, slower growth rate and higher kid mortality have been reported in this breed (Honhold, 2001). The growth, development and productivity of individuals depend on the proper functioning of all the body systems. One of the most important and complicated system for the production performance of the livestock is the endocrine. Adrenal gland, one of the endocrine organs plays a vital role in all living being with the secretion of important steroid hormones. These paired glands are located anatomically above the kidney and regulates a variety of functions in fetal as well as postnatal life (Mecenas *et al.*, 1996; Smith *et al.*, 1998). In ruminants, the right adrenal gland is found medial to the cranial extremity of the right kidney while the left gland is less regular and less stable in position (Dyce *et al.*, 2002). This gland produces a variety of hormones and also maintains homeostasis and play role in response to stress (Humayun *et al.*, 2012). The two anatomical distinct units of adrenal cortex and medulla is originated from embryological mesoderm and neural crest ectoderm respectively and is surrounded by a connective tissue capsule (Dellman, 1993; Junqueira *et al.*, 1998). The adrenal cortex is subdivided into three (Dellman, 1993) or four (Bacha and Wood, 1990) distinct zones of epithelial cells. The cortical and medullary cells are different in their secretory functions. The adrenal cortex produces most of the mineralo-corticoids and glucocorticoids whereas the epinephrine and nor-epinephrine are secreted from the medulla in response to intense emotional reactions (Humayun *et al.*, 2012). During normal activity, the medulla continuously secretes small quantities of these hormones (Junqueira *et al.*, 1998).

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Both the glands are surrounded by a connective tissue capsule. A number of factors are responsible for the weight, length, width and thickness of the adrenal glands. The histology of the adrenal gland varies according to the sex and age of the animal. Several studies have been undertaken on the morphological and histological aspects in various livestock species with emphasis on the basic information such as in *Mus musculus* (Waring, 1935), *Tammar wallaby* (Call et al., 1980), *Macacamulata* (McNulty et al., 1981), *Ovisaries* (Naaman and Durand, 1997), *Papio species* (Leavitt et al., 1999) and *Homo sapiens* (Sirianni et al., 2005). However, no one to the best of our knowledge investigated the histomorphology the indigenous goat breed of Bangladesh. Therefore, this experiment was done focusing particularly on the gross and the histological feature variation of adrenal glands of Black Bengal buck and doe found in Bangladesh.

Materials and Methods

The experiment was conducted in the in the laboratory of the Department of Anatomy and Histology, Sylhet Agricultural University, Bangladesh. Both the right and left adrenal glands were collected from eight (8), Black Bengal goats (four, 4 from each sex) of 24 months old, slaughtered in the Govt. slaughter house, Sylhet, Bangladesh. The age of the goat was 24 months which was determined by using dentition (Chibuzo, 2006). The adrenal glands were dissected out aseptically immediately after slaughter. The glands were collected immediately after slaughter with aseptic measures using a scalpel. The shape, color and other morphometric data were measured and recorded. The absolute weights were measured by a digital balance.

For the histological examinations, the tissue pieces from adrenal glands were fixed in bouin's fluid. Following fixation, the tissues were then dehydrated, cleared, and embedded in paraffin. Sections of 5- μ thicknesses were cut using a rotary microtome. Haematoxylin and eosin staining protocol was used to stain the tissue sections. Following the protocol mentioned by Drury et al. (1976) the permanent slides were prepared. Capsular thickness, thickness of cortex (zonaglomerulosa, zonafasciculata and zonareticularis) and medulla, cellular height and nuclear diameter of 50 randomly selected cortical and medullary regions were measured using the stage and ocular micrometers. Finally using the SPSS (Statistical Package for the Social Sciences) program the recorded data were analysed. To find out the differences in different recorded data Student's paired *t*-test was performed considering $P < 0.05$ as significant.

Results and Discussion

Gross anatomy of the adrenal gland of goat

This present experiment illustrates an incipient data on morphological and histological features of adrenal glands of adult Black Bengal Goat of Bangladesh. Both the glands were found as bean shaped structures (Figure 1), the right gland was located along the cranial end of the medial side of the right kidney and the left one was closely attached to the left renal vein as reported by Getty (1975). Blood vessels profusely supplying the glands were seen necessary for active transport of hormones, which was similar to the findings of Dyce et al. (2002). The right gland was 1.5 cm long and 1 cm wide and the left gland was longer than the right one. The weight of the right adrenal gland in buck and doe were recorded as 0.79 ± 0.035 g and 0.74 ± 0.036 g whereas the left one were recorded as 0.97 ± 0.018 g and 0.80 ± 0.007 g, respectively (Table 1). A significant weight difference was recorded between buck and doe, though the finding of Panchal et al. (1998) was different while studying the histomorphology of adrenal gland of sheep.

Table 1. Mean (\pm SD) weight of the adrenal glands in relation to the body weight of goat (n=08)

Name of Species	No. of animals	Age (Months)	Av. Weight of the animals (kg g ⁻¹)	Av. Weight of the adrenals (g) \pm SD	
				Right	Left
Buck	4	24	15 kg	$0.79 \pm 0.035^*$	0.97 ± 0.018
Doe	4	24	12 kg	$0.74 \pm 0.036^*$	0.80 ± 0.007

* indicates significant difference (Paired *t*-test, $*P < 0.05$) from the adrenal gland of left side.



Fig. 1. Left and Right adrenal gland of goat

Histology of the adrenal gland of goat

Capsule and trabeculae: The thick capsule consisted of collagenous, reticular and elastic fibers covered the adrenal gland was richly supplied with blood vessels and nerves (Figs. 2 and 3). Thin connective tissue trabeculae originating from the capsule penetrated the cortex and also sometimes the medulla (Figs. 3 and 8). This observation is similar to the findings of several other authors in ram, sheep, Indian buffalo and in human (Pancha *et al.*, 1998; Stokoe, 1959; Prasad and Yadava, 1974 and Junqueira *et al.*, 1998). However, Dellmann (1993) reported a common thin capsule surrounds the adrenal glands of ruminants. Average thickness of the capsule was $69.58 \pm 2.59 \mu\text{m}$ and $54.79 \pm 2.393 \mu\text{m}$, possessing 3.31% and 2.83% of total thickness in buck and doe respectively (Table 2).

Table 2. Micrometric measurement (Mean \pm SD) in μm of the adrenal components and their ratio

Name of Species	Thickness of Capsule (μm)	Thickness of cortex (μm)	Thickness of medulla (μm)	Ratio of capsule to gland %	Ratio of cortex to gland %	Ratio of medulla to gland %
Buck	69.58 ± 2.59	1488.34 ± 34.307	545.83 ± 35.046	3.31	70.75	25.94
Doe	54.79 ± 2.393	1397.29 ± 61.276	481.63 ± 22.271	2.83	72.26	24.91

Cortex of the adrenal gland of goat: Our result showed that, three distinct cortical zones along with a thin zonaintermedia and centrally located medulla were found in the adrenal gland of goat, but distinct demarcation was lacking between those zones. Those findings were similar to the findings of Bacha and Bacha (2000) in mammals, Panchal *et al.* (1998) in sheep, Dellmann (1993) in ruminants, however Gued *et al.* (1982) reported two zones in mouse. The thickness of the cortex of Buck and Doe was recorded $1488.34 \pm 34.307 \mu\text{m}$ and $1397.29 \pm 61.276 \mu\text{m}$ (Table 2) where the cortico-medullary ratio was 73.17:26.83 and 74.37:25.63, respectively.

Table 3. Mean (\pm SD) thickness of different cortical zones irrespective to their sex (n=08)

Name of Species	Widths of different cortical zones Mean \pm SD (μm)			
	Zonaglomerulosa	Zonaintermedia	Zonafasciculata	Zonareticularis
Buck	$218.79 \pm 9.242^*$	8.26 ± 0.208	$1071.46 \pm 34.125^*$	$189.83 \pm 9.01^*$
Doe	243.41 ± 22.488	8.57 ± 0.128	927.50 ± 42.103	217.81 ± 9.489

* indicates significant difference (Paired *t*-test, **P*<0.05) from the adrenal gland of Doe.



Fig. 2. Adrenal gland of adult goat showing Capsule (C), Zonaglomerulosa (ZG), Fibroblast cell (FC), Sinusoid (SS) and Blood vessel (BV). H & E stains X 825

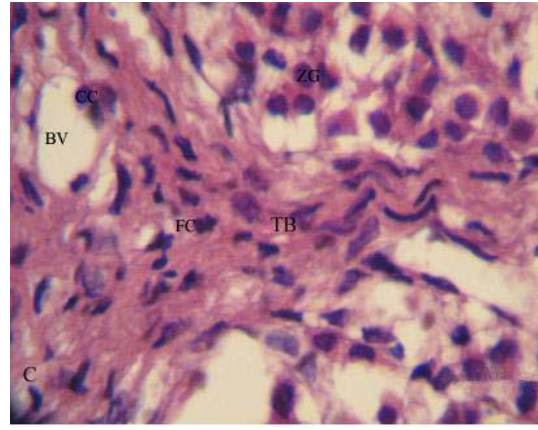


Fig. 3. Adrenal gland of adult goat showing Capsule (C), Zonaglomerulosa (ZG), Fibroblast cell (FC), Cortical cell (CC), Trabeculae (TB) and Blood vessel (BV). H & E stains X 825

The outermost zone just beneath the capsule was formed as zonaglomerulosa. It contained prismatic cells that formed irregular clusters or cords and contained lipid vacuoles (Figs. 2 and 3). The thickness of this zone was $218.79 \pm 9.242 \mu\text{m}$ thick in Buck and $243.41 \pm 22.488 \mu\text{m}$ in Doe covering 14.98% of the cortical and 8.25% in total adrenal gland thickness in Buck and 18.08% of cortical and 8.97% in total thickness in Doe (Table 3). The diameter of the cell and nucleus was recorded respectively $6.77 \pm 0.28 \mu\text{m}$ and $3.98 \pm 0.213 \mu\text{m}$ in buck and $8.0 \pm 0.112 \mu\text{m}$ and $4.06 \pm 0.322 \mu\text{m}$ in doe (Table 4). The average thickness of this zone and the cell and nuclear size goes similarly with the findings of several other researchers (Bacha and Bacha, 2000; Junqueira *et al.*, 1998; Panchal *et al.*, 1998; Dellmann, 1993; and Holmes, 1961).

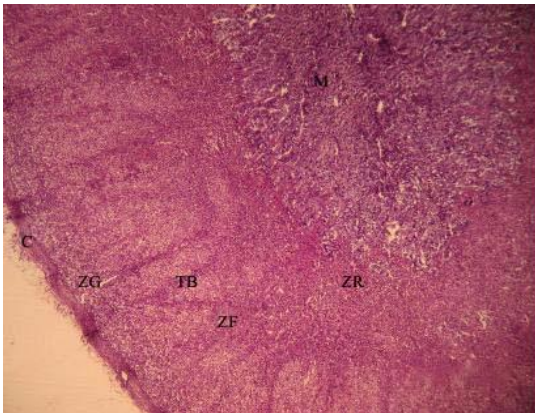


Fig. 4. Adrenal gland of adult goat showing Capsule (C), Zonaglomerulosa (ZG), Zonafasciculata (ZF), Zonareticularis (ZR), Trabeculae (TB) and medulla (M). H & E stains X 33

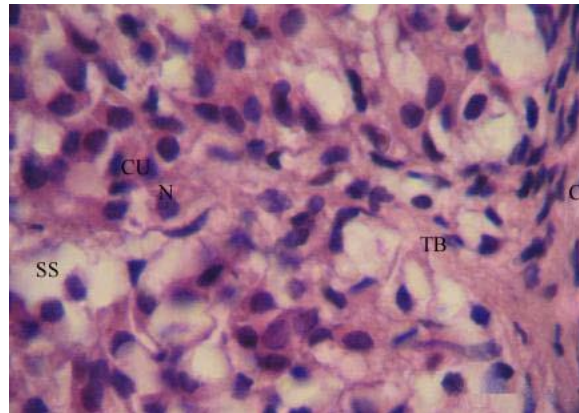


Fig. 5. Zonaglomerulosa of adrenal gland of adult goat showing Cuboidal cell (CU), Sinusoid (SS), Capsule (C) Nucleus (N) and Trabeculae (TB). H & E stains X 825

Next to zonaglomerulosa, a transitional zone of small-undifferentiated cells comprised zonaintermedia was reported in this present study. The diameter of the cell and nucleus were $8.26 \pm 0.208 \mu\text{m}$ and $3.97 \pm 0.418 \mu\text{m}$ in Buck and $8.57 \pm 0.128 \mu\text{m}$ and $4.22 \pm 0.459 \mu\text{m}$ in Doe (Table 4). This transitional zone was also reported by Bacha and Bacha (2000) in mammals, Dellmann (1993) in ruminants and Prasad and Yadava (1974) in Indian buffalo.

Table 4. Mean (\pm SD) (μ m) height of cell and diameter of nucleus of different cortical and medullary cells of adrenal gland in goat irrespective to their sex (n=08)

Parameters	Name of the species	
	Buck	Doe
Zonaglomerulosa	Cell height	6.77 \pm 0.28*
	Nuclear diameter	3.98 \pm 0.213
Zonaintermedia	Cell height	8.26 \pm 0.208
	Nuclear diameter	3.97 \pm 0.418
Zonafasciculata	Cell height	8.27 \pm 0.376*
	Nuclear diameter	4.10 \pm 0.422*
Zonareticularis	Cell height	7.97 \pm 0.478
	Nuclear diameter	4.47 \pm 0.276
Medullary epinephrine cell	Cell height	10.43 \pm 0.192
	Nuclear diameter	6.48 \pm 0.408*
Medullary norepinephrine cell	Cell height	7.25 \pm 0.175*
	Nuclear diameter	4.43 \pm 0.224*

*indicates significant difference (Paired *t*-test, **P*<0.05) from the adrenal gland of Doe.

The foamy zone, zona fasciculata was found as the widest adrenal cortical zone (1071.46 \pm 34.125 μ m thick in buck and 927.50 \pm 42.103 μ m in doe), comprising 50.93% and 47.96% of the total gland in buck and doe respectively (Table 3). The highest thickness was reported by several other authors (Bacha and Wood, 1990; Junqueira *et al.*, 1998; Yilmaz *et al.*, 2005). The zonafasciculata forms 60% of the cortex in the rabbit (Yilmaz *et al.*, 2005), 65% of the gland (Junqueira *et al.*, 1998) and the foamy appearance were considered due to existence of profuse lipid droplets (Bacha and Wood, 1990; Dellman, 1993; Junqueira *et al.*, 1998). This zone was composed of columnar cell in goat, arranged in radiating columns surrounding the sinusoid containing endothelial cell (Figure 4 & 6). They were mainly single cell column but double cell column may also found with frequent double nucleus. The size of the nucleus increased in size in the deeper part of the column. The size of the cells and the nucleus of the zonafasciculata were largest among all other cortical layers cells and nucleus. The average diameter of the cell and the nucleus were 8.27 \pm 0.376 μ m and 4.10 \pm 0.422 μ m, respectively in buck and 10.19 \pm 0.343 μ m and 5.65 \pm 0.903 μ m in doe (Table 4). The average cell size and nucleus which was measured by Prasad and Yadava (1974) is approximately similar to the present.

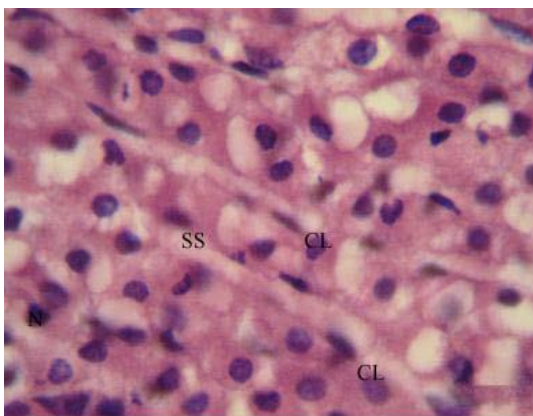


Fig. 6. Zonafasciculata of adrenal gland of adult goat showing Columnar cell (CL) and Sinusoid (SS). H & E stains X 825

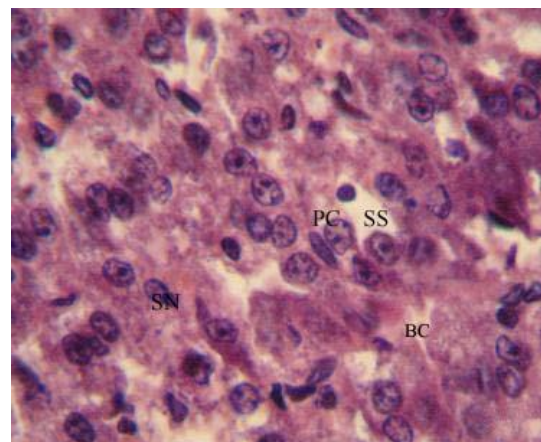


Fig. 7. Zonareticularis of adrenal gland of adult goat showing Polygonal cell (PC), Blood cell (BC) and Sinusoid (SS). H & E stains X 825

The innermost cortical layer, the zonarectularis contained polygonal cells with spherical nucleus arranged in small irregular groups around the sinusoids (Figs. 4 & 7). The cell and the nucleus size of the zonarectularis in buck and doe were 7.97 ± 0.478 , 4.47 ± 0.276 and 8.12 ± 0.31 , 4.32 ± 0.019 μm (Table 4). Its thickness was 189.83 ± 9.01 μm in buck and 217.81 ± 9.489 μm in doe (Table 3) and was about 9.02% of the gland thickness in buck and 11.26% in doe. A similar observation was found from the study of Bacha and Bacha (2000) in mammals, Junqueira *et al.* (1998) in human, Dellmann (1993) in ruminants and Prasad and Yadava (1974) in Indian buffalo.

Medulla of adrenal gland of goat: The average thickness of adrenal medulla of black Bengal goat was 545.83 ± 35.046 μm and 481.63 ± 22.271 μm , respectively in buck and doe (Table 2) comprising about one quarter (25.94% in buck and 24.91% in doe) of the total thickness of the gland. This area was composed of polyhedral cells, arranged in cords or clumps and supported by the reticular fiber network with profuse capillary supply (Figures 8, 9). These findings were similar to the observations of Jamdar and Ema (1982), though Junqueira *et al.* (1998) reported 13% adrenal medulla in human. Occasionally the cortical cells were also seen in the medulla.

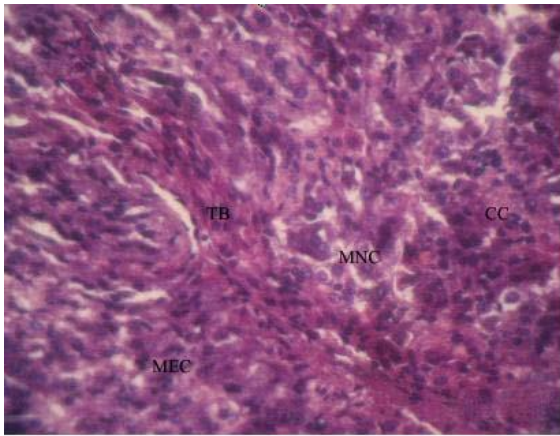


Fig. 8. Medulla of the adrenal gland of adult goat showing Medullary Epinephrine Cell (MEC), Medullary Norepinephrine Cell (MNC), Cortical cell (CT) and Trabeculae (TB). H&E stains X 82.5

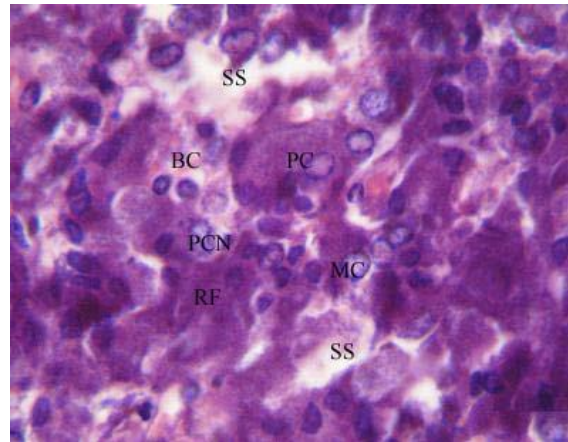


Fig. 9. Medulla of the adrenal gland of adult goat showing Medullary Cell (MC), Sinusoid (SS), Blood cell (BC), Polygonal cell (PC), Polygonal Cell Nucleus (PCN) and Reticular Fiber (RF).H & E stains X 330

This area was subdivided into two distinct zones. The outer zone, which was made up of large, intensely stained epinephrine secreting cells, were columnar type and the nucleus present in apical part of the cells. The average size of an epinephrine storing cell and nucleus in buck and doe were recorded 10.43 ± 0.192 μm , 6.48 ± 0.408 μm and 9.84 ± 0.221 μm , 6.02 ± 0.288 μm , respectively (Table 4). The inner zone, which was formed by, clusters of small, polyhedral cells with low staining affinities secrete norepinephrine with centrally placed nucleus. The norepinephrine storing cell and nucleus in buck and doe were 7.25 ± 0.175 μm , 4.43 ± 0.224 μm and 7.08 ± 0.505 μm , 3.81 ± 0.197 μm , respectively (Table 4). These findings were similar to the study of Bacha and Bacha (2000) in mammals, Junqueira *et al.* (1998) in human and Dellmann (1993) in ruminants. Coupland and Weakley (1968) identified epinephrine granules as being bigger compared to norepinephrine granules which was also reported in our findings.

In conclusion it can be mentioned that the present study demonstrates a general feature in the structures of adrenal cortex and medulla of black bangle goat. The findings related to shape and size of the adrenal gland of the buck and doe where medullary portion is more in doe than buck. This indicates epinephrine and norepinephrine are secreted in large quantities in female than the male in response to intense emotional reactions.

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