Histological Study on the Fibrous Architecture of the Adrenal Gland During Postnatal Development in Buffalo

Vishnudeo Kumar1*, Ram Saran Sethi2, Opinder Singh3

ABSTRACT
The study was conducted on the adrenal glands of 29 buffaloes ranging from day old to one year of age. The adrenal gland was fully differentiated into cortex and medulla in the day old buffalo calf and was completely surrounded by a dense fibrous capsule at this stage. The capsule consisted of outer layer of loose connective tissue and inner layer of thick collagen fibres. The connective tissue trabeculae made up of collagen and reticular fibres arose from the inner aspect of capsule and infiltrated the cortical parenchyma of the gland. The reticular fibres dominated in the inner aspect of the capsule and the proportion of reticular fibres was more in the zona glomerulosa and outer zone of medulla, however, elastic fibres were not observed in any part of the gland up to one year of age. The average thickness of the capsule increased with progression of age which was due to increase in the density of collagen fibres especially in the inner zone of capsule.

Keywords: Adrenal, Buffalo, Histological, Postnatal.

INTRODUCTION
This research work was conducted at the Department of Anatomy, College of Veterinary Science, GADVASU, Ludhiana, Punjab-141004, India. The histological observations on the fibrous architecture of the adrenal gland during the postnatal development had been studied in goat (Hakeem et al., 1993, Kale et al., 2003), sheep (Stokoe, 1959), camel (Nagpal et al., 1991) and dog (Saleh et al., 1974). There is little information on the adrenal gland of buffalo (Sohal and Chaturvedi, 1962, Prasad and Yadava, 1972), however, no literature is available on the fibrous architecture of the buffalo adrenal gland with advancement of age. Hence, the present study was undertaken.

MATERIALS AND METHODS
The study included adrenal glands from 29 buffaloes aging from day old to one year and based on their age the animals were divided into three Groups viz., Group-I (day old to 3 months), Group-II (more than 3 months to 6 months) and Group-III (more than 6 months to 1 year). The glands were fixed in 10 percent neutral buffered formalin and orth fluid immediately after collection. The paraffin blocks were prepared by acetone benzene schedule (Luna, 1968) and sections of 5-6 µm thickness were obtained on glass slide with the help of rotatory microtome. The sections were stained with haematoxylin and eosin method for routine histomorphism (Luna, 1968), Masson’s trichrome method for collagen fibres (Luna, 1968), Gridley’s method for reticular fibres (Sheehan and Hrapchak, 1973), Verhoeff’s method for elastic fibres (Sheehan and Hrapchak, 1973) and Holme’s method for Nerve cells and fibres (Luna, 1968).

RESULTS AND DISCUSSION
The adrenal gland of day old buffalo calf was completely surrounded by a dense fibrous capsule as reported earlier in the adrenal gland of adult buffalo (Sohal and Chaturvedi, 1962 Prasad and Yadava, 1972), sheep (Stokoe 1959), goat (Hakeem et al., 1993, Kale et al., 2003), camel (Nagpal et al., 1991), pig (Kumar and Sharma, 2019), foal (Kumar and Sharma, 2013) and dog (Saleh et al., 1974). The capsule was made up of outer layer of loose connective tissue and inner layer of thick collagen fibres in the adrenal gland of day old buffalo calf. The collagen bundles of the capsule run parallel to the surface of the gland (Fig 1). The outer part of

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capsule contained blood vessels and neuronal elements. The connective tissue trabeculae arose from the inner aspect of capsule and descended deep into the cortical parenchyma. In some locations the trabeculae even reached up to the medulla. The trabeculae accompanied the medium size blood vessels and nerves. Similar type of connective tissue trabeculae were also reported in the adrenal gland of adult buffalo (Sohal and Chaturvedi, 1962, Prasad and Yadava, 1972), goat (Hakeem et al., 1993, Kale et al., 2003) and camel (Nagpal et al., 1991). In the present study, the collagen fibres were found in the zona glomerulosa and were present in high proportion in the inner zone of medulla. Prasad and Yadava (1972) also observed few fine bundles of collagen fibres entering into the zona glomerulosa from the inner layer of capsule in the adrenal gland of young calves and adult buffalo as reported by Hakeem et al. (1993) and Nagpal et al. (1991) in the adrenal gland of Indian goat and camel, respectively. In the adrenal capsule of buffalo fine collagen fibres were found between the cortex and medulla which appeared to form a capsule around the medulla, however it was incomplete. The connective tissue capsule at corticomedullary junction varied in thickness from one part to another (Fig. 2) and was

**Fig 1:** Dense fibrous capsule (C), zona glomerulosa (G) large number of sinusoidal capillaries and hemorrhages (arrow) in the foetal cortex (f) in day old buffalo calf. Masson's trichrome stain x 100X

**Fig 2:** Collagen fibres in outer part of medulla (Mo) and condensation of collagen fibres (arrows) at the corticomedullary junction in three months old buffalo calf. Masson's trichrome stain x 200X

**Fig 3:** Condensation of connective tissue at corticomedullary junction in form of incomplete capsule (arrows) containing round to elongated eosinophilic cells in five months old buffalo calf. Also seen are outer (Mo) and inner (Mi) zone of medulla. Masson's trichrome stain x 200X.
thin and irregular. The presence of a layer of connective tissue at the corticomedullary junction had also been reported in young calves and adult buffalo (Prasad and Yadava, 1972) and foal (Kumar and Sharma, 2013). Similarly Holmes (1968) observed an incomplete corticomedullary boundary in the adrenal gland of monkey. In the present study the proportion of collagen fibres was more in the inner zone of medulla as reported earlier in young calves and adult buffalo (Prasad and Yadava, 1972) and goat (Hakeem et al., 1993, Kale et al., 2003). The connective tissue capsule present at the corticomedullary junction was more defined in the five months old buffalo calves and often contained round to elongated cells with eosinophilic cytoplasm (Fig. 3). Holmes (1968) also found two types of cells in the corticomedullary capsule viz: round cell with eosinophilic cytoplasm and cells with elongated nuclei resembling fibroblast. Howard-Miller (1927) suggested that degeneration of foetal cortex during the postnatal period was associated with development of corticomedullary fibrous zone and this zone disappeared with advancement of age.

The proportions of collagen fibres were more in the zona glomerulosa and outer and inner zone of medulla in Group-III (Fig. 4). The connective tissue capsule at corticomedullary junction also showed an increase in thickness and cellular density of eosinophilic cells, however, distinct medullary connective tissue capsule was not observed as earlier reported in the adrenal gland of ferrets and monkey (Holmes, 1961, 1968).

In the day old buffalo calf the inner aspect of the adrenal capsule had a higher proportion of reticular fibres which entered into the parenchyma of the gland (Fig. 5). The proportion of reticular fibres was more in the zona glomerulosa and outer zone of medulla in Group- I. Similarly Prasad and Yadava (1972), Hakeem et al. (1993) and Nagpal et al. (1991) also observed the presence of the reticular fibres in the adrenal gland of adult buffalo, goat and camel, respectively. Further Prasad and Yadava (1972) reported the blending of collagenous, elastic and reticular fibres along with the smooth muscle fibres in the inner part of the capsule of adrenal gland of adult buffalo. The reticular fibres intermingled with loose collagenous, elastic and smooth muscle fibres in the capsule of adrenal gland of goat (Hakeem et al., 1993). In the present study the reticular fibres became more organized in the capsule and parenchyma in Group –II and Group-III. The proportion of reticular fibres appeared to be more in the zona glomerulosa and outer

![Fig 4: Collagen fibers in capsule (C), zona glomerulosa (G), outer (Mo) and inner (Mi) zone of medulla in eight months old buffalo. Also seen are zona fasciculata (F), zona reticularis (R) and central vein (Cv). Masson's trichrome stain x 40X.](image1)

![Fig 5: Reticular fibers in capsule (C), zona glomerulosa (G), foetal cortex (f) and medulla (M) in day old buffalo calf. Gridley's stain x 40X.](image2)
Fig 6: Reticular fibers in capsule (C), zona glomerulosa (G), outer (Mo) and inner (Mi) zone of medulla in one year old buffalo. Gridley’s stain x 40X.

zone of medulla (Fig. 6), which supports the earlier findings of Prasad and Yadava (1972) in adrenal gland of Indian buffalo.

The Verhoeff stain sections of buffalo adrenal gland in all the Groups did not show the presence of elastic fibres in any part of gland up to one year of age but the elastic fibres were observed in blood vessels. However, Prasad and Yadava (1972), Hakeem et al. (1993) and Nagpal et al. (1991) reported the elastic fibres in the adrenal capsule of adult buffalo, goat and camel, respectively.

ACKNOWLEDGMENT
Authors are thankful to the Professor & Head and staff of Department of Veterinary Anatomy, College of Veterinary Science, GADVASU, Ludhiana for providing necessary facility.

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