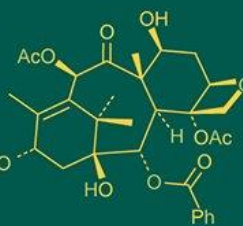
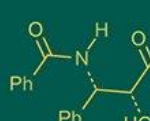


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Anjali

Veterinary Officer, Veterinary
Dispensary, Veterinary
College, KVAFSU, Karnataka,
India

Ashok Pawar

Head of the Department,
Department of Veterinary
Anatomy, Veterinary College,
KVAFSU, Karnataka, India

Girish MH

Assistant Professor,
Department of Veterinary
Anatomy, Veterinary College,
KVAFSU, Karnataka, India

Biradar SM

Veterinary Officer, Veterinary
Dispensary, Veterinary
College, KVAFSU, Karnataka,
India

Anand

Veterinary Officer, Veterinary
Dispensary, Veterinary
College, KVAFSU, Karnataka,
India

Corresponding Author:**Anjali**

Veterinary Officer, Veterinary
Dispensary, Veterinary
College, KVAFSU, Karnataka,
India

Comparative histological and histochemical study of cricoid cartilage in Deccani sheep and Bidri goat

Anjali, Ashok Pawar, Girish MH, Biradar SM and Anand

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Abstract

The present comparative histology and histochemical study of cricoid cartilage was carried out to know the structural differences between cricoid cartilage of Deccani sheep and Bidri goat. The cricoid cartilage in Deccani sheep and Bidri goat consisted of lamina epithelia, lamina propria-submucosa, muscular layer, a cartilaginous layer and tunica adventitia. The epithelium in both species was lined by pseudostratified columnar epithelium with goblet cells. There was more number of goblet cells in the epithelium of Deccani sheep than Bidri goat. The lamina propria-submucosa of both species were continuous and composed of loose connective tissue collagen, elastic, collagen and reticular fibers and consisted of fibroblast, fibrocytes, mast cells, macrophages, blood vessels and nerve fibers. A large number of plasma cells and lymphatic aggregations were also noticed beneath the epithelium. The glands were tubulo-acinar type, sero-mucous in nature. The glandular lobules were separated from each other by connective tissue fibers consisted of collagen, reticular and elastic fibers in both species. The muscular layer consisted of skeletal muscle between the submucosa and cartilaginous layer was observed in both the species. The cartilage was of hyaline type surrounded by perichondrium in both species. The cartilage was continuous plate like structure surrounded by perichondrium, which had two distinct layers Viz., chondrogenic layer consisting of elliptical lacunae and fibrous layer consisted of collagen and elastic fibers. Tunica adventitia composed of loose connective tissue in both the species. Histochemical observation of cricoid cartilage of both species revealed moderate PAS positive reaction in all secretory units of the glands and epithelium. The epithelium and some of the glandular cells showed strong Alcian Blue reaction at pH 2.5 in both species.

Keywords: Cricoid cartilage, histology, histochemistry, Bidri goat, Deccani sheep

Introduction

Socioeconomic growth and national economy of the country has been significantly backed by small ruminants sector. Small ruminants contribute about 10% GDP in the total value of livestock sector. Sheep and goat are the important source of food (milk & meat) and main source of income for small and marginal farmers. The heavy mortality in the small ruminants especially in goats has been reported due to respiratory tract disease. The larynx is a conducting part of respiratory system which protects the lower respiratory passage against inundation. Cricoid cartilage being one of the important cartilage and unpaired one. The predominant breeds of sheep and goat in Bidar district are Deccani sheep and Bidri goat respectively and limited scientific work has been carried out, hence the present study was undertaken to study and to correlate any histological and histochemical variations in the Cricoid cartilage of Deccani sheep and Bidri goat.

Materials and Methods

The materials for the study were collected from Six Deccani sheep and Six Bidri goat immediately after slaughter from local slaughter house. The collected Cricoid cartilage samples were washed in normal saline and fixed in different fixatives like 10% Neutral buffered formalin, Zenker's fluid and Bouin's fluid. The samples were processed in isopropyl alcohol-xylene sequence and embedded in paraffin by routine method (Luna, 1968)^[10]. Sections were cut at 5-6 µm thickness and were utilized for histomorphological and histochemical studies. The Cricoid cartilage tissue fixed in Neutral buffered formalin were pre-frozen for 1-2 hrs and embedded in freezing media; using cryostat 6-7 µm thickness sections were cut and utilised for lipid studies.

The histomorphological study of Cricoid cartilage was carried out by Ehrlich's Haematoxylin and Eosin stain (Luna, 1968) ^[10], Van Geison's stain for collagen fibers (Bancroft *et al.*, 2008) ^[3], Gomori's method for reticular fibers (Luna, 1968) ^[10], Verhoeff's method for elastic fibers (Bancroft *et al.*, 2008) ^[3] and Toluidine blue method for mast cells (Singh and Sulochana, 1996) ^[12] whereas histochemical study was carried out using PAS technique for mucopolysaccharides (Bancroft, *et al.*, 2008) ^[3], Alcian blue pH 2.5 for sulphated sulfomucins and sialomucins (Bancroft *et al.*, 2008) ^[3] and Oil red O for lipids (Luna, 1968) ^[10].

Results and Discussion

The cricoid cartilage in Deccani sheep and Bidri goat consisted of lamina epithelia, lamina propria-submucosa, muscular layer, a cartilaginous layer and tunica adventitia (Sinha *et al.*, 2012) ^[13] (Fig. 1 and Fig. 4).

The Cricoid cartilage in Deccani sheep and Bidri goat was lined by pseudostratified columnar epithelium containing goblets cells within them (Fig. 2). There were more number of goblet cells in the epithelium of Deccani sheep than the epithelium of Bidri goat (Chunhui *et al.* 2009 and Sinha *et al.* 2012) ^[4, 3]. However, it differs with the earlier reports in ruminants (Eurell and Frappier, 2006) ^[7] in sheep and goat (Alsadi, 2006) ^[1] in Camel Dromadarius (Ebrahimi *et al.*, 2010) ^[5]. The lamina propria and submucosa were continuous with no distinct boundary. lamina propria-submucosa was composed of loose connective tissue fibers collagen, elastic and reticular fibers, and consisted of fibroblast, fibrocytes, mast cells, macrophages, blood vessels and nerve fibers were also noticed (Fig. 1, Fig. 3, Fig. 5, Fig. 6, Fig. 7 and Fig. 8). More number of plasma cells were noticed in this region. Large amount of fat cells were observed in the lamina propria-submucosa (Fig. 1) (Trautmann and Fiebiger, 2002, Eurell and Frappier, 2006, Chunhui *et al.*, 2009, Sinha *et al.*, 2012, Arup, 2014 and Eshra *et al.*, 2016) ^[14, 7, 4, 13, 2, 6]. The presence of large number of plasma cells and diffuse lymphatic aggregation below the epithelium indicates role in immune response against the infection and also play role in defense mechanism.

The glands were sero-mucous in nature (Fig. 9) (Trautmann and Fiebiger, 2002, Alsadi, 2006, Eurell and Frappier, 2006, Chunhui *et al.*, 2009, Arup, 2014 and Eshra *et al.*, 2016) ^[14, 1, 7, 4, 2, 6]. The sero-mucous glands help in secretion of the watery and mucous fluids that moistens the surface epithelium and the air. The glandular lobules were separated from each other by connective tissue fibers consisted of collagen, reticular and elastic fibers (Fig. 3, Fig. 5 and Fig. 7). However, it differs in camels where serous glands were observed (Eshra *et al.*, 2016) ^[6]. The glands were tubulo-acinar type of glands (Fig. 9). However, in Bactrian camel glands were branched coiled tubulo-alveolar types of glands (Chunhui *et al.*, 2009) ^[4]. This may be due to species variation.

The acini and tubules of serous secreting units were lined by pyramidal to columnar shaped cells with basally placed nuclei. The mucous acini and tubules were lined by pyramidal shaped cells with basally placed flattened nuclei. The acini consisted of small round lumen whereas, the

tubular portion contained long narrow lumen (Eurell and Frappier, 2006) ^[7]. The skeletal muscle was observed between submucosa and cartilage. The cartilage was of hyaline type and continuous plate like surrounded by perichondrium, which had two distinct layer Viz., chondrogenic layer consisting of elliptical lacunae and fibrous layer consisting of collagen and elastic fibers (Fig. 1 and Fig. 4). The lacunae were polyhedral in shape contained chondrocytes. The chondrocytes were separated from each other by interterritorial matrix. The perichondrium consisted of two layers chondrogenic layer made up of elliptical lacunae, and the other consisting of collagen and elastic fibers (Fig. 4) (Trautmann and Fiebiger, 2002, Alsadi, 2006, Eurell and Frappier, 2006, Chunhui *et al.*, 2009, Konig and Liebich, 2009, Ebrahimi *et al.*, 2010, Sinha *et al.*, 2012, Arup, 2014 and Eshra *et al.*, 2016) ^[14, 1, 7, 4, 9, 5, 13, 2, 6].

The histochemical study of Cricoid cartilage in Deccani sheep and Bidri goat showed moderate PAS positive reaction in the gland and epithelium indicating the presence of mucopolysaccharides (Fig. 10 and Fig. 12). The epithelium and some glandular cells showed strong positive reaction for Alcian Blue reaction at pH 2.5 (Fig. 11) indicating the occurrence of sulfated sialomucins and sulfomucins (Majid, 1986 and Kahwa and Purton, 1996) ^[11, 8] whereas acidic and neutral reaction for Alcian blue-PAS stains was noted in Bactrian camel (Chunhui *et al.*, 2009) ^[4].

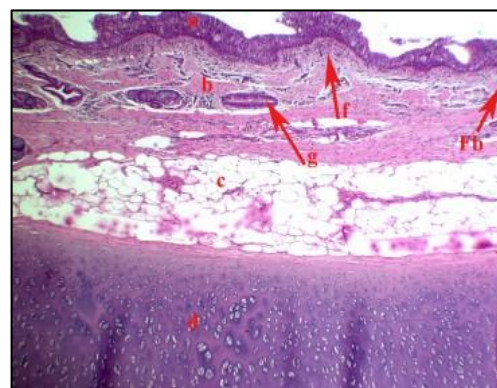


Fig 1: Photomicrograph of cricoid cartilage of Deccani sheep showing lamina epithelia (a), lamina propria-submucosa (b), fat cells (c), cartilage (d), lymphatic aggregation (f), blood vessel (g) and fibroblast (fb). H&E stain X 4

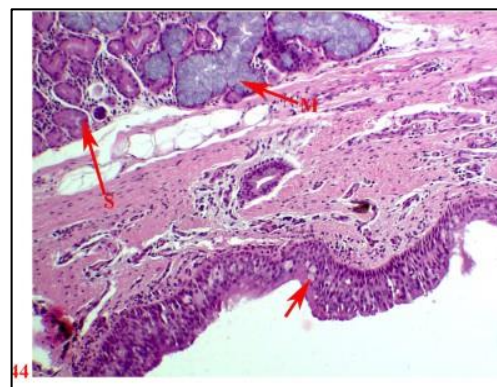


Fig 2: Photomicrograph of cricoid cartilage of Deccani sheep showing pseudostratified columnar epithelium with goblet cells (arrow), serous (S) and mucous (M) in secretory units. H&E stain X 10

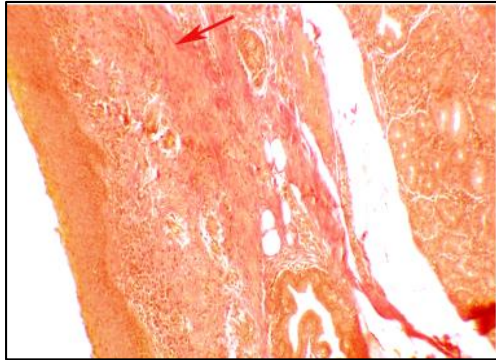


Fig 3: Photomicrograph of cricoid cartilage of Deccani sheep showing collagen fibers (arrow) in the lamina propria. Vangieson's stain X 10

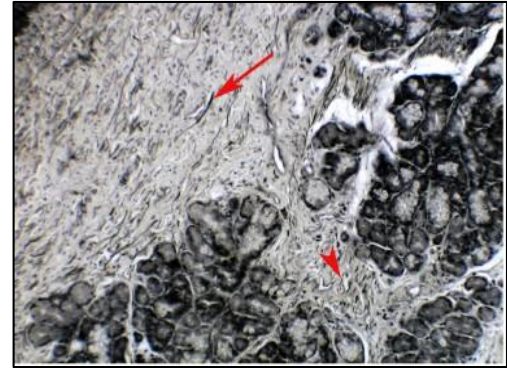


Fig 7: Photomicrograph of cricoid cartilage of Bidri goat showing elastic fibers (arrow) in the lamina propria and (arrowhead) separating the glandular lobules. Verhoeff's stain X 4

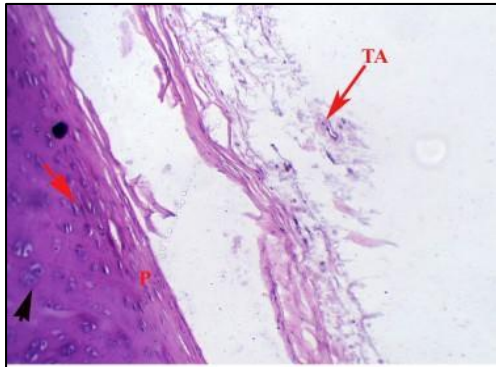


Fig 4: Photomicrograph of cricoid cartilage of Deccani sheep showing tunica adventitia (TA), perichondrium (P), elliptical lacunae (red arrow) and polyhedral lacunae (black arrow). H&E stain X 10

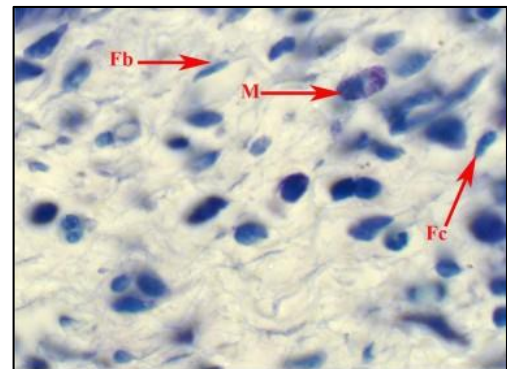


Fig 8: Photomicrograph of cricoid cartilage of Deccani sheep showing fibroblast (Fb), fibrocyte (Fc) and mast cell (M) in the lamina propria. Toluidine blue stain X 40

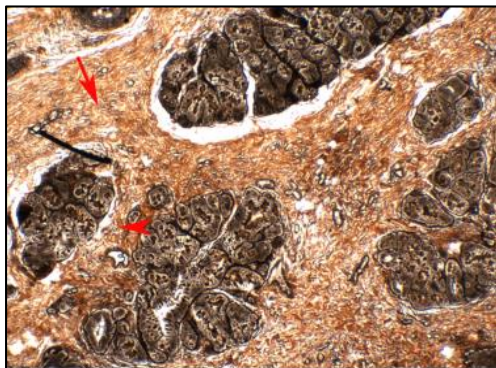


Fig 5: Photomicrograph of cricoid cartilage of Bidri goat showing reticular fibers (arrow) in the lamina propria and (arrowhead) separating the glandular lobules. Gomori's stain X 4

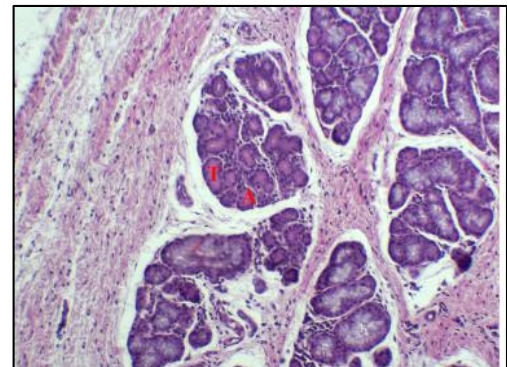


Fig 9: Photomicrograph of cricoid cartilage of Bidri goat showing tubular (T) and acinar (A) type seromucous glands. H&E stain X 10

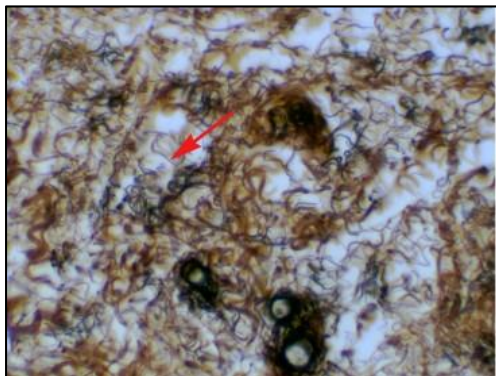


Fig 6: Photomicrograph of cricoid cartilage of Deccani sheep showing reticular fibers (arrow) in the lamina propria. Gomori's stain X 40

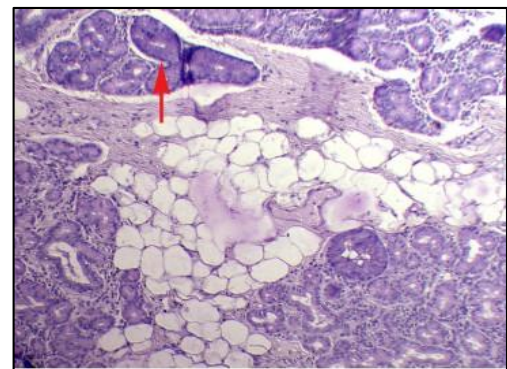


Fig 10: Photomicrograph of cricoid cartilage of Deccani sheep showing PAS positive reaction in secretory cells (arrow). PAS stain X 10

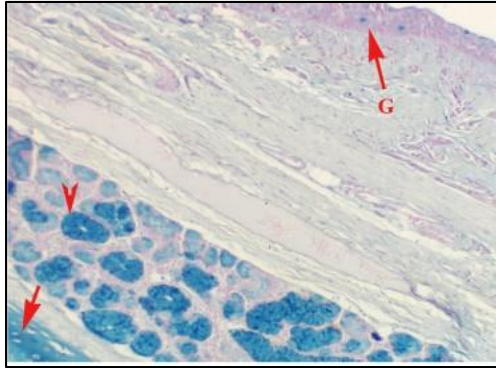


Fig 11: Photomicrograph of cricoid cartilage of Bidri goat showing positive reaction in cartilage (arrow), goblet cell (G) and few secretory cells (arrow head) for Alcian blue. Alcian blue at pH 2.5 X 10

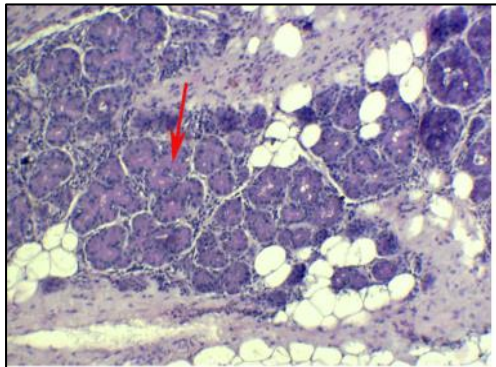


Fig 12: Photomicrograph of cricoid cartilage of Bidri goat showing PAS positive reaction in secretory cells (arrow). PAS stain X 10

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