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#### Purushottam

College of Veterinary and Animal science, Navania, Vallabhnagar, Udaipur-313601, Rajasthan University of Veterinary and Animal Sciences, Bikaner, Rajasthan, India

#### **Balwant Meshram**

College of Veterinary and Animal science, Navania, Vallabhnagar, Udaipur-313601, Rajasthan University of Veterinary and Animal Sciences, Bikaner, Rajasthan, India

#### Anita Kumari

College of Veterinary and Animal science, Navania, Vallabhnagar, Udaipur-313601, Rajasthan University of Veterinary and Animal Sciences, Bikaner, Rajasthan, India

#### Mamta Kumari

College of Veterinary and Animal science, Navania, Vallabhnagar, Udaipur-313601, Rajasthan University of Veterinary and Animal Sciences, Bikaner, Rajasthan, India

#### Nitika

College of Veterinary and Animal science, Navania, Vallabhnagar, Udaipur-313601, Rajasthan University of Veterinary and Animal Sciences, Bikaner, Rajasthan, India

#### Corresponding Author Purushottam

College of Veterinary and Animal science, Navania, Vallabhnagar, Udaipur-313601, Rajasthan University of Veterinary and Animal Sciences, Bikaner, Rajasthan, India

## Histomorphological and histochemical studies on the cystic duct in guinea fowl (*Numida meleagris*)

### Purushottam, Balwant Meshram, Anita Kumari, Mamta Kumari and Nitika

#### Abstract

The histomorphological and histochemical studies were conducted on cystic duct in Guinea Fowl (*Numida meleagris*). Three tunics i.e. tunica mucosa, tunica muscularis and tunica adventitia were noticed in the microscopic study of cystic duct. The lamina propria also noticed significantly. The tunica adventitia was observed as the undivided histological structures which provide means of access for the arterioles and venules. Weigert's method for elastic and collagen fibers has revealed the lamina propria and tunica adventitia as the major component of collagen fibers and rests of components were formed by intermixing of both collagen and elastic fibers. In the histochemical studies of cystic duct, Periodic Acid Schiff (PAS) for glycogen has shown the intense activity of mucus at the mucosal layer and pertaining to the acid phosphatase and alkaline phosphatase, it showed very mild activity at lamina propria and tunica muscularis.

Keywords: Bile, billiary apparatus, cystic duct, guinea fowl, histochemical, histomorphology

#### Introduction

Cystic duct is a component of extra hepatic biliary apparatus. Cystic duct reclines between neck of gall bladder and first part of small intestine viz. duodenum. This duct originates from cranio-dorsally or neck of the gall bladder, then turns backward and run parallel to the common hepatic duct. Cystic duct involves directly drainage of bile from liver to duodenum (Toouli and Bhandari, 2006) <sup>[8]</sup>, hence it may be confirmed as cysto-enteric duct. Cystic duct travels dorsal to the pancreas and some time it may pass through the inter-pancreatic lobules with common hepatic duct (CHD), unfastened anteriorly to the opening of CHD as oblique to this duct. Common bile duct is a form of these two-duct combination. Guinea fowl has come from Africa and it belongs to the oldest of the gallinaceous birds of Africa family Numididae in the order Galliformes. The Literature reveals meager information on this duct and hence histomorphological and histochemical studies of the cystic duct is studies on Guinea Fowl (*Numida meleagris*).

#### **Materials and Methods**

The extra hepatic duct is acquires from 12 Guinea Fowl birds (*Numida meleagris*) that collects from meat shops of Udaipur. The collected samples of various age groups are transfer to the laboratory for routine investigate the histomorphology and histochemical of cystic duct that is component of extra hepatic biliary apparatus. The methods subjected for histomorphological studies are haematoxyline and eosin and Weigert's method. (Singh and Sulochana, 1996) <sup>[7]</sup>. PAS, acid and alkaline phosphatases are used for studies of histochemistry of cystic duct.

#### **Results and Discussion**

The cystic duct is trench between the neck of gallbladder and small intestine. It carries the bile from liver to duodenum, which runs parallel to the CHD. Cystic duct made up of three tunics in its hollow structure, tunica mucosa, tunica muscularis and tunica adventitia with observance of lamina muscularis and submucosal layer without their presence. The sizable lamina propria also noticed. (Fig.1)

Longitudinal array and simple columnar epithelium with centrally located nucleus were observed in tunica mucosa of cystic duct, which aid in the bile regulation. At the gall bladder end of cystic duct simple alveolar duct present that is limiting in lamina propria in diminishing quantum, while duodenal end observed with significant lamina propria at proximity of

duodenum entrance. Caudal end of cystic duct also observed which reveals the lymphoid aggregation and lymphatic vessels along endothelial cells. (Fig.2)

Common bile duct travel posteriorly or even through the pancreas to get join the main pancreatic duct (duct of Wirsung), which is formed by the union of the common hepatic and cystic duct. The intramural portion in cystic duct have shown more dimension, because of inner longitudinal and outer circular muscle fibers, rather than CHD at opening end in duodenum. In cystic duct these two muscle fibers seen but sometime three different muscle fibers were also observed as to inner circular, middle longitudinal and outer circular particularly against the caudal end. Four muscle fibers also encountered, but in exceptional cases as to form inner most longitudinal, circular, longitudinal and outer most circular muscle fibers. Tunica adventitia mainly access for arterioles and venules, it is an undivided histological structure.

Tunica muscularis is thick at duodenal end than gallbladder end of cystic end. Collagen fibers observed mainly in boundaried structures as lamina propria and tunica adventitia, while other components were formed by intermingling of both collagen and elastic fibers. Elastic fibers prominently present in blood vessels. (Fig. 3)

The present study discovered that the complete length of cystic duct measurable from its union at neck of gall bladder to the unification of duodenum, and it observed as  $50.78\pm$  0.45. The width of cystic duct is mid portion of its length, that is observed as  $1.135\pm0.008$ . These observations were comparable with different researchers who were noticed the same in different species as Cai and Gabella (1983) <sup>[3]</sup> who had measured the cystic duct 8-10mm long in guinea pig, Abdalla *et al.* (2013) <sup>[1]</sup> were measured 2-3cm length and 2-3mm in diameter of cystic duct in humans.

The complete course of cystic duct is parallel to the CHD from starting to end. Cystic duct and CHD open obliquely to each other at duodenum. Cystic duct encountered longitudinal array in its mucosal layer that is maintains or regulate the pressure of bile in duct at the time of drafting from gallbladder to duodenum, however its flow was also maintained by the sphincter present at its duodenal opening. Abdalla et al. (2013) <sup>[1]</sup> and Chamberlain (2013) <sup>[4]</sup> has observed cystic duct mucosa lining by spiral folds i.e. known as spiral valve of heister. Union of boundaries of cystic duct, CHD, and the hepatic border formed Calot's triangle, which was noticed by various workers in different species. Anatomical structure Calot's triangle was not determined, because the cystic duct and CHD found running parallel and directly opens into the duodenum. However, Cachoeira et al. (2012)<sup>[2]</sup> were discovered cystohepatic triangle or Clot's triangle in Cadavers studies. Histochemically when the cystic duct was stained with PAS for glycogen, the mucus at mucosal layer has shown the intense activity. (Fig.4) Its appearance showed the functional maintenance as the source of energy at localized level. The acid phosphatases and alkaline phosphatases activity shows its very mild presence at lamina propria and tunica muscularis which was indicating the functional dynamics of cystic duct. (Fig. 5 and 6).

#### Conclusion

Cystic duct made up of three tunics in its hollow structure,

tunica mucosa, tunica muscularis and tunica adventitia with observance of lamina muscularis and submucosal layer without their presence. The intramural portion in cystic duct have shown more dimension, because of inner longitudinal and outer circular muscle fibers, rather than CHD at opening end in duodenum. In cystic duct these two muscle fibers seen but sometime three or four different muscle fibers were also observed particularly against the caudal end. Histochemically when the cystic duct was stained with PAS for glycogen, the mucus at mucosal layer has shown the intense activity. The acid phosphatases and alkaline phosphatases activity shows its very mild presence at lamina propria and tunica muscularis.

#### Haematoxylin and eosin 100X

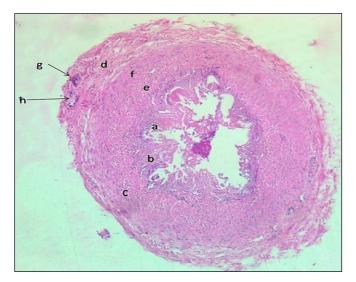


Fig 1: Photomicrograph is showing middle part of cystic duct. a. Tunica mucosa, b. Lamina propria, c. Tunica muscularis, d. Tunica adventitia, e. Inner longitudinal, f. Outer circular, g. Arterioles and h. Venules

#### Haematoxylin and Eosin 400X

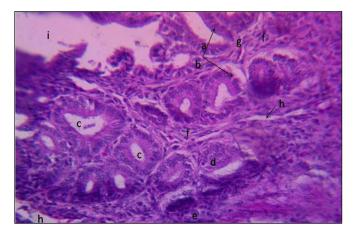


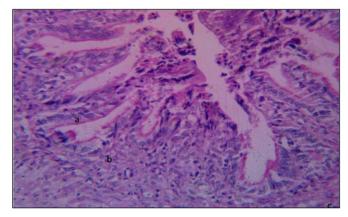
Fig 2: Photomicrograph is showing caudal end of the wall of cystic duct at intramural Portion. a. Tunica mucosa, b. Columnar cells, c. Exocrine glands (biliary glands), d. lymphoid aggregation, e. Lymphocytes, f. Lamina propria, g. Fibroblast, h. Lymphatic vessels and i. Lumen.

#### Weigert's method 40X



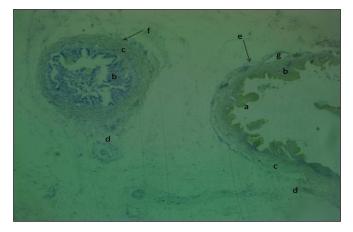
Fig 3: Photomicrograph is showing caudal end of the biliary apparatus with Collagen and elastic fibers. a. Cystic duct, b. Common hepatic duct, c. Pancreatic Duct d. Artery, e. Vein, f. Connective tissue, g. Tunica serosa, h. Lamina propria, i. Tunica muscularis, j. Tunica adventitia, k. Collagen and Elastic fiber, l. Capillary and m. Duodenum

#### Periodic Acid Schiff 400X



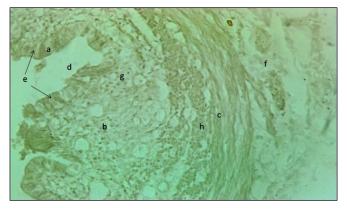
**Fig 4:** Photomicrograph is showing the PAS activity in wall of cystic duct at Caudal end. a. Tunica mucosa, b. Lamina propria and c. Tunica muscularis.

#### Acid Phosphates 100X



**Fig 5:** Photomicrograph is showing acid phosphates activity at the wall of cystic duct and body of gallbladder. a. Tunica mucosa, b. Lamina propria, c. Tunica muscularis, d. Tunica adventitia, e. Gallbladder, f. Cystic duct and g. Lymph vessels.

#### Alkaline phosphates 400X



**Fig 6:** Photomicrograph is showing alkaline phosphates activity at the caudal end of wall of cystic duct. a. Tunica mucosa, b. Lamina propria, c. Tunica muscularis, d. Lumen, e. Simple columnar epithelium and f. Tunica adventitia.

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