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Anatomical studies on the cystic artery of sheep (*Ovis aries*)

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Abstract

Cystic artery in the present study was the branch of right hepatic artery it gave small branches to the duct and lateral part of the right lobe and quadrate lobe. The observed mean external diameter of cast preparations was 0.96 ± 0.05 mm. Similarly, morbid arteriography revealed 0.87 ± 0.05 mm. The endothelium was lined by a single layer of flat squamous type cells. The nucleus shape was oblong and resembled tear drop. The subendothelial layer was very thin. The internal elastic membrane was continuous. The tunica media constituted the bulk of the arterial wall and chiefly consisted of 10 to 15 smooth muscle cells. The external elastic lamina was indistinct. There was a no evidence of vaso vasorum and nervi vasorum. The tunica intima, media, adventitia and total wall thickness was 4.48 ± 0.63 , 16.64 ± 1.9 , 10.88 ± 1.18 and 32.03 ± 2.75 respectively while the long and short luminal diameters were 75.52 ± 2.56 and 12.8 ± 1.9 respectively. The endothelium, basement membrane and externa of cystic artery exhibited intense alcian blue activity which indicated the presence acid mucopolysaccharides while weak alcian blue activity was observed in media and strong PAS reaction was noticed in tunica externa. The frozen sections revealed intense AKP activity in endothelium and tunica externa while moderate and scattered in tunica media. The ACP activity was very weak in endothelium and media while moderate in tunica externa.

Keywords: Cystic artery, morphology, histology, histochemistry

Introduction

Recent liver anatomy studies lay stress on the organization, course and ramification of vascular system. Such approach may provide clarity not only on the functional significance and differential abilities in the lobes of liver but also provide surgeon with useful information and knowledge for safe removal of the affected parts of the liver (Dyce *et al.*, 2010) [3]. The arterial diameter is importance especially due to development of new techniques for liver transplantation and histomorphological organization determines their physico-mechanical properties and is influenced by hemodynamic forces of the luminal blood flow including, pulse rate, arterial flow velocity and resistance to flow in vascular segments and supplied organs. There has been limited scientific work carried on the cystic artery in small ruminants, hence the present research work has prompted to take up and establish a more precise and detailed information on the cystic artery which would give a comprehensive and useful data for interdisciplinary works.

Material and methods

“Anatomical studies on the cystic artery of Sheep (*Ovis aries*)” was carried out in the Department of Veterinary Anatomy, College of Veterinary Science, Rajendranagar, Hyderabad. The liver samples along with coeliac artery were collected from twenty two apparently healthy sheep irrespective of sex from local slaughter houses in and around Hyderabad. Age of the animals and weight of liver samples were between 1.5 to 2 years and 450 to 700 grams, respectively. Liver specimens were transported to the department for undertaking various anatomical studies as per standard procedure (Culling, 1974) [2].

Gross morphology and morphometry

1. Gross dissection

The coeliac artery was dissected by fixing the liver in 10% NBF to enable the study of course and branching. Both corrosion cast technique and morbid arteriography was conducted to enable these studies.

2. Corrosion casting

Prepare the vascular corrosion cast. The Cold cure self polymerizing acrylic (methyl methacrylate) solution was reconstructed in 1:2 ratio with few drops of red color dye. This solution was injected into the hepatic artery. The vessels were clamped and the injected liver specimens were kept at room temperature for effective polymerization. Subsequently the specimens were digested in 75% hydrochloric acid till the corrosion process was completed. The specimen was cleaned under running water to remove macerated tissue leaving the vessels exposed. The morphometry of the cystic artery in terms of length and diameter were recorded by using digital calipers.

3. Morbid arteriography

The radio-opaque suspension was prepared by dissolving red lead oxide @ 20 mg/100 ml of soap solution and was injected through coeliac artery under steady and constant digital pressure with help of cannula and syringe while keeping the specimen towards gravity. The injected liver was radiographed immediately at 10 MAS, 42 KVP and 70cm FFD. Arteriography readings were measured by using FUZI System with IWCR (Image Works Computer Radiography) software at Dept. of Radiology, College of Veterinary Science and Hyderabad.

Histology, histometry and histochemistry

1. Histology

The samples were fixed in 10% neutral buffered formalin and processed by routine paraffin method (Singh and Sulochana, 1998)^[7]. About 5-6 μm thick sections were cut and subjected to the following routine and special staining techniques. Harris haematoxylin and eosin staining for micro-architecture (Singh and Sulochana, 1998)^[7], Masson's trichrome method for collagen and muscle fibers (Singh and Sulochana, 1998)^[7], Verhoeff's method for demonstration of elastic fibers (Singh and Sulochana, 1998)^[7] and Van-Giesons method for demonstration of collagen fibers (Singh and Sulochana, 1998)^[7].

2. Histometry

Histometry was evaluated by factorizing the Occular with Stage micrometer. Following parameters were recorded, analyzed (George and Cochran, 1994)^[4].

- a) Thickness of the arterial wall tunics.
- b) Luminal diameter of the cystic artery.

The myosytic density in tunica media was measured from histological sections by the OLYMPUS A X 70 System with advanced cell senses software at Ruska Labs, College of Veterinary Science, Hyderabad.

3. Histochemistry

Histochemical investigation in the cystic artery of sheep was studied from routine paraffin and cryo-sections. The paraffin processed sections were subjected to study the localization of neutral (Luna, 1968)^[6] and acid mucopolysaccharides (Singh and Sulochana, 1998)^[7] while fresh Cryo-sections of 10-15 μm thickness from prefixed chilled formal calcium were subjected to study the alkaline and acid phosphatase activity in the tunics of cystic artery (Singh and Sulochana, 1998)^[7].

Results and discussion

1. Gross morphology and morphometry

Cystic artery in the present study was the branch of right hepatic artery which was loosely attached to the dorsal aspect of the cystic duct (Fig.1 & 2). This finding is in consonance with the observations of Anuradha *et al.*, (2002)^[1] in buffalo calves, Teh *et al.*, (2007)^[8] in sheep and Thamke and Rani, (2017)^[9] in human cadavers, where they mentioned that this artery was branch of RHA. However, Kuru, (2016)^[5] in rabbit reported that the left hepatic branch gave the cystic artery to the gall bladder in humans. In the present study we observed small branches were given to the duct and lateral part of the right lobe and quadrate lobe (Fig.3). At the neck of the gall bladder it divided into two or three branches which coursed up and supplied blood to the free margin, right and left margins of gall bladder. These arteries were divided into further smaller branches which had tortuous paths around the wall of the gall bladder (Fig.3).

The observed mean external diameters of cystic artery in these cast preparations was 0.96 ± 0.05 mm. Similarly, morbid arteriography revealed 0.87 ± 0.05 mm. Such detailed split data was not available in the literature reviewed.

2. Histology, histometry and histochemistry

The tunica intima of cystic artery consisted of endothelium resting on the basement membrane. The endothelium was lined by a single layer of flat squamous type cells which were placed on thin basement membrane. The nucleus shape was oblong and resembled tear drop. The subendothelial layer was very thin. The internal elastic membrane was continuous (Fig.4). The tunica media was the most developed of the three tunics which constituted the bulk of the arterial wall and chiefly consisted of 10 to 15 smooth muscle cells. The myocytic density per 500 μm^2 area was 24.40 ± 1.02 . They were arranged circularly and concentrically held together by collagen fibers and few elastic fibers (Fig.5&6). The external elastic lamina was indistinct. The tunica externa comprised of collagen fibers and elastic fibers. There was a no evidence of vaso vasorum and nervi vasorum (Fig.5&6).

The cystic artery thickness was evaluated by histometry in the present study. The CA tunica intima, media, adventitia and total wall thickness in the present study was 4.48 ± 0.63 , 16.64 ± 1.9 , 10.88 ± 1.18 and 32.03 ± 2.75 respectively while the long and short luminal diameters were 75.52 ± 2.56 and 12.8 ± 1.9 respectively. No data is available for comparison.

The histochemical study revealed that intense alcian blue activity was observed in the endothelium, basement membrane of externa of cystic artery which indicated the presence acid mucopolysaccharides while weak alcian blue activity was observed in media and strong PAS reaction was noticed in tunica externa (Fig.7). The frozen sections revealed intense AKP activity in endothelium and tunica externa while moderate and scattered in tunica media (Fig.8). The ACP activity was very weak in endothelium and media while moderate in tunica externa (Fig.9). The present findings could not be compared as such reports were not available in the literature reviewed so far.

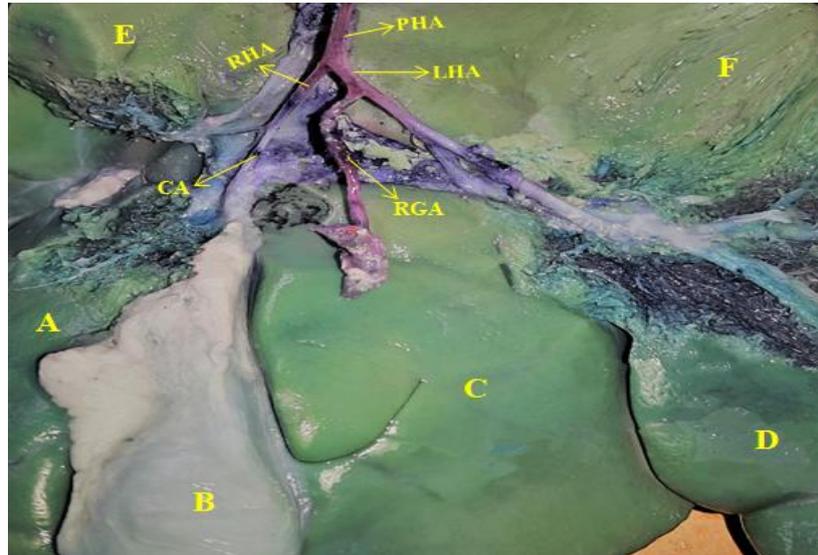


Fig 1: Photograph showing the origin of the cystic artery
 Right lobe B. Gall bladder C. Quadrate lobe D. Caudate lobe E. Caudate process. F. Caudate lobe.
 Proper hepatic artery (PHA), Right hepatic artery (RHA), Left hepatic artery (LHA), Right gastric artery

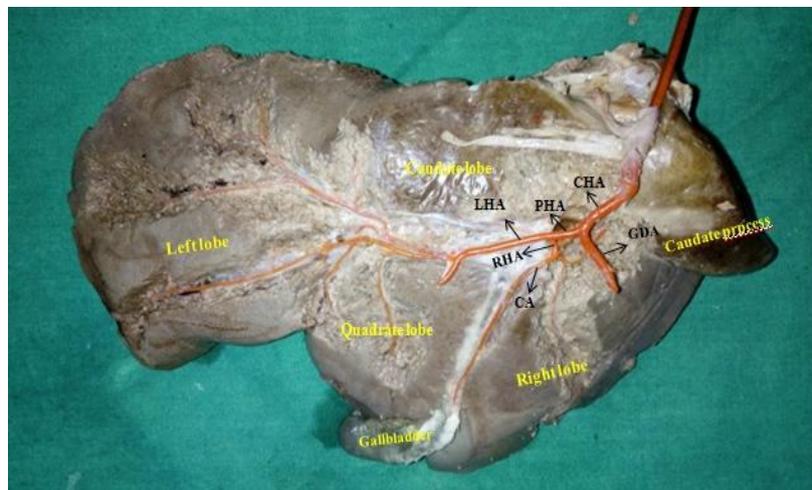


Fig 2: Photograph of Acrylic cast showing origin of cystic artery (visceral view)

Common hepatic artery (CHA), Proper hepatic artery (PHA), Right hepatic artery (RHA), Left hepatic artery (LHA), Gastroduodenal artery (GDA), Cystic artery (CA)

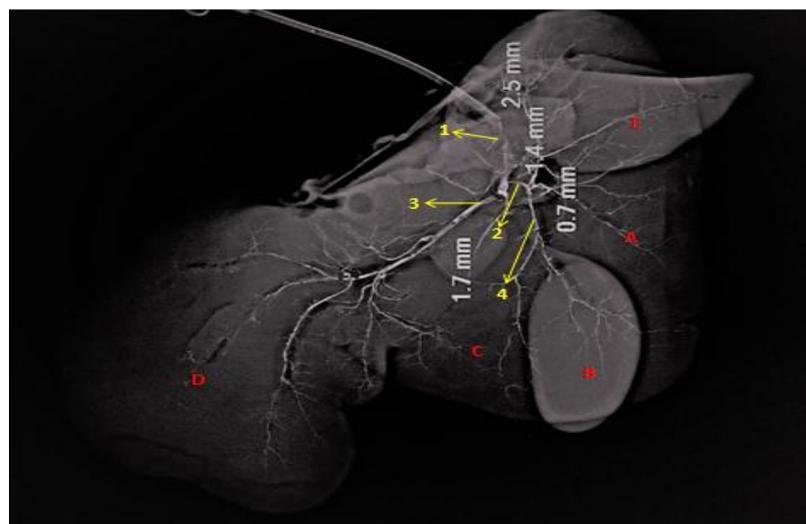


Fig 3: Morbid arteriogram showing the origin and divisions of cystic artery
 A. Right lobe, B. Gall bladder, C. Quadrate lobe, D. Left lobe, E. Caudate process of caudate lobe.
 1. Common hepatic artery. 2. Right hepatic artery. 3. Left hepatic artery. 4. Cystic artery

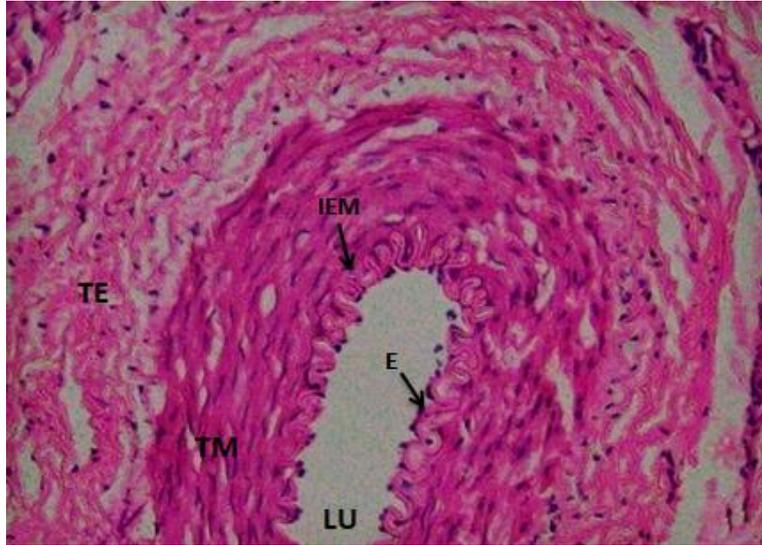


Fig 4: Photomicrograph of cystic artery showing three tunics Endothelium (E), Tunica media (TM), Tunica externa (TE), Internal elastic membrane (IEM), Lumen (LU). H & E X 20



Fig 5: Photomicrograph of cystic artery showing tunics, collagen and muscle cells Endothelium (E), Tunica media (TM), Tunica externa (TE), Internal elastic membrane (IEM), Lumen (LU), Smooth muscle (*), Collagen fibers (CF). Vangieson X 20

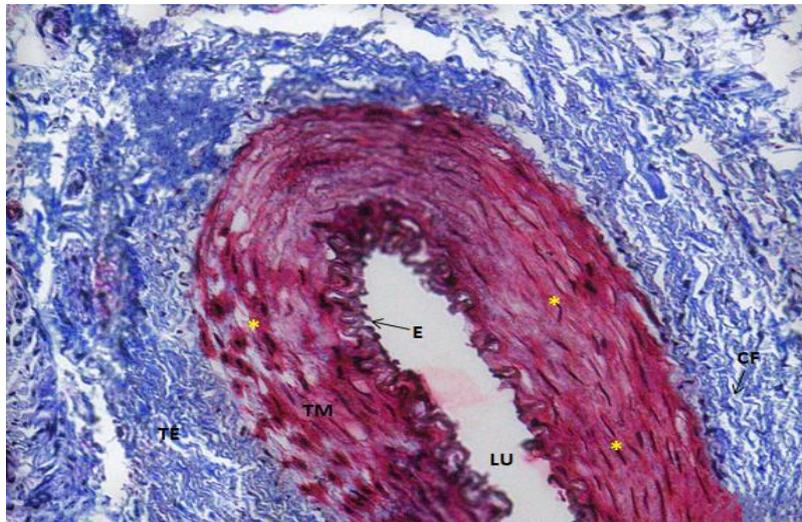


Fig 6: Photomicrograph of cystic artery showing tunics, collagen and muscle cells Endothelium (E), Tunica media (TM), Tunica externa (TE), Lumen (LU), Smooth muscle (*), Collagen fibers (CF). Masson's trichrome X 20

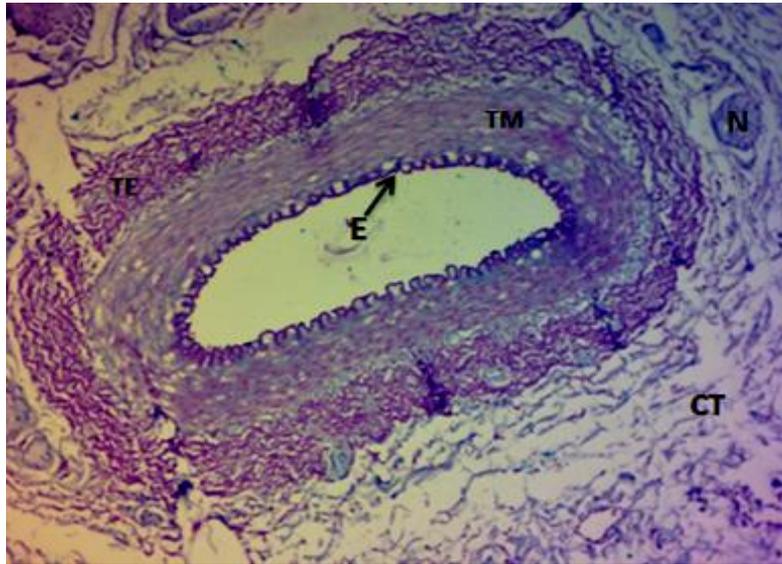


Fig 7: Photomicrograph of cystic artery showing alcian blue activity Endothelium (E), Tunica media (TM), Tunica externa (TE), Nerve (N), Connective tissue (CT). PAS-AB X 10

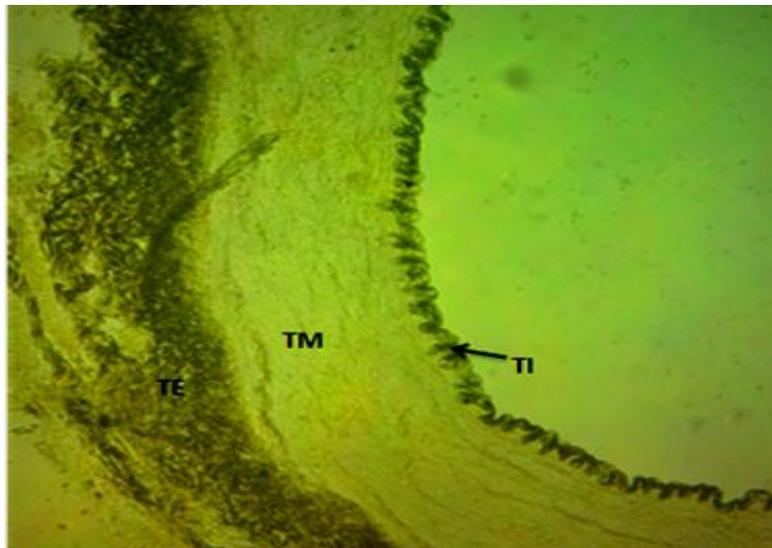


Fig 8: Photomicrograph showing AKP activity in cystic artery. Tunica intima (TI), Tunica media (TM), Tunica externa (TE). AKPX 10

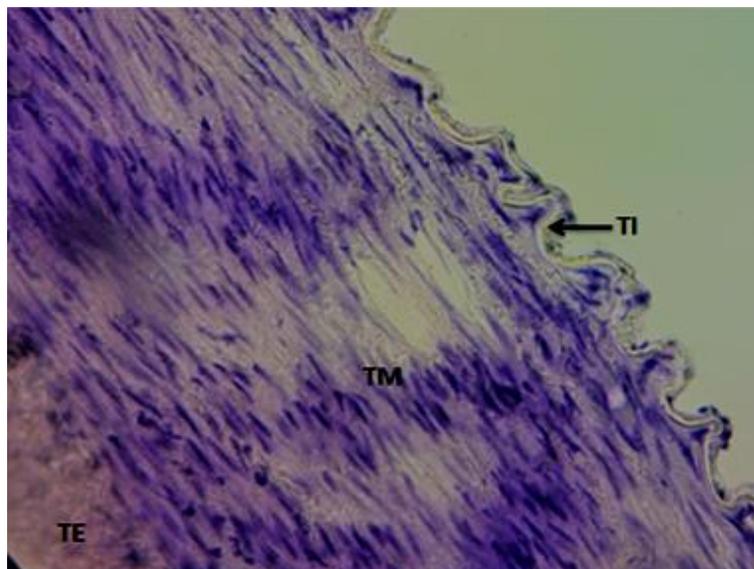


Fig 9: Photomicrograph showing ACP activity in cystic artery. Tunica intima (TI), Tunica media (TM), Tunica externa (TE). ACPX 40

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