Os cordis of mature Dromedary camel heart (Camelus dromedaries) with special emphasis on cartilago cordis

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ABSTRACT

The present study was conducted to investigate the bony structure (i.e., os cordis) in the heart of mature Dromedary camel. Ten hearts were collected from healthy mature camels of both sexes aging between 7-9 years. Grossly, the heart contained one large, elongated bone that was embedded inside the heart wall where the cardiac muscles were inserted and fixed. The bony structure was located at the adjacent areas between aorta and atria. Also, it was lying in the atrioventricular plane near the junction of the interatrial and interventricular septa, and was extended anteriorly into the atrioventricular valve rings. Histologically, the os cordis was composed of fibrous connective tissue, small foci and pieces of hyaline cartilage, calcified cartilage, and a large piece of spongy bone. The spongy bone contained red and white bone marrows with numerous red blood cells, adipocytes and osteocytes. This is the first detailed study on os cordis of Dromedary camel heart with special reference to cartilago cordis.

Keywords
Aortic ring, Camel heart, Camelus dromedaries, Cardiac skeleton, Cartilago cordis, Os cordis

INTRODUCTION

The cardiac skeleton (i.e., os cordis) is the dense fibrous connective tissue where the cardiac muscle fibers are inserted. Also, it supports the heart valves. Cardiac skeleton is composed of three types of structures: (i) annuli fibrosi rings that surround heart valve, (ii) fibrous trigones that fill the spaces between the rings, and (iii) septum membranaceum consisting of extensions of cardiac skeleton into the interatrial and interventricular septae (Ghallab, 2000; Dellmann and Eurell, 1998; Gartner and Hiatt, 2007; Samuelson, 2007; Junqueira et al., 2007). The fibrous rings are mainly composed of interwoven bundles of collagen fibers and a few elastic fibers surrounding the atrioventricular openings and the openings of aorta and pulmonary artery. The fibrous trigones consist of areas of dense connective tissues located between the aorta and the atrioventricular openings, which are connected by the aortic-mitral curtain. The right trigone, together with the membranous septum, constitutes the central fibrous body. In certain mammalian species, the trigones contain fibro-cartilage, hyaline cartilage, and even bony material. The latter forms a right and left cardiac bone (os cordis) related to the trigone between atrioventricular ostia and the aortic ostium, respectively. Moreover, the right cardiac bone is regularly larger as compared to the left (Gray, 1995).

In the heart of several animals like cattle, sheep, goat and pig, bony structures may present (Egerbacher and Weber, 2001; Malik et al., 1972; Mia, 1973; Schmack, 1974; Nickel et al., 1984). A single cartilage was reported in Otter (Lutra lutra) heart (Zogall, 1992). The main function of this bony structure is to stabilize the heart during contraction and relaxation. In most mammalian species, cardiac skeleton is composed of coarse collagen fibers, fibrocartilage, and pieces of hyaline cartilages. Bony structure (i.e., os cordis) is a normal constituent in the ruminant heart.
Increasing amounts of calcified cartilage and bone are correlated with the advancement of age (Egerbacher et al. 2000). Aretz (1981) reported that cartilage is ossified with the advancement of age in sheep and goats. Frink and Merrick (1974) found right cardiac bone in sheep heart; however, occasionally an additional second bone was also noted on the left. Daasch (1925) clarified that in the pig heart, 2 pieces of cartilages were lying close to the fibrous ring of the aorta. These cartilages tend to calcify and ossify in the animals of aging >3 years. Similarly, cartilage was found in the heart skeleton of dog and other canidae and felidae (Schmack, 1974; Simic, 1938). In addition, cartilage is present in the heart of mice and rats (Hueper, 1939), rabbits (Kern, 1927), Syrian hamsters (Kelsall and Visci, 1970), and serpents (Young, 1994). Benninghoff (1994) described a cartilage-like structure is present in the center of the right fibrous trigone of the human heart.

In a number of vertebrates, cartilago cordis, a cartilaginous element is present in heart (Young, 1994). Cartilago cordis varies in the size, shape, and location. However, presence of cartilago cordis is not dependent on body size, taxonomic relationships, or habitat preference. Murata and Yamada (1986) illustrated that the glycosaminoglycans are present in the cartilage of the porcine heart. The cartilage of the porcine heart also contains hyaluronic acid, chondroitin, chondroitin sulfate A and/or C, and keratin sulfate.

Nawal et al. (1998) studied morphology of heart in the Dromedary camel with special reference to the coronary vessels and conduction system of heart. Also, presence of os cordis has been reported by Nawal et al. (1998). However, there are very few reports that describes in detail histological studies on os cordis of Dromedary camel. Therefore, this study was aimed to investigate the fibrous skeleton of the camel heart (Camelus dromedaries) with special reference to cartilago cordis.

**MATERIALS AND METHODS**

Hearts of ten healthy mature camels of both sexes aging between 7-9 years were collected from Zagazig Slaughter house located in Sharkia province, Egypt. The specimens were immediately fixed in 10% buffered neutral formalin and decalcified using Ethylene Diamine Tetra Acetic Acid (EDTA). Then, the specimens were dehydrated in ascending grades of ethanol series, cleared in benzene and embedded in paraffin. Histological sections of 5-7 μm in thickness were prepared and mounted on glass slides. The sections were dewaxed in xylene, hydrated in descending grades of ethanol series and stained with Harris's Hematoxylin and Eosin (H&E) for routine histological studies (Bancroft and Gamble, 2001; Ghonimi et al., 2014a). The microphotographies were taken using a digital Dsc-W130 super steady cyber shot camera (Sony, Japan) connected to an Olympus BX21 light microscope.

**RESULTS AND DISCUSSION**

The bony structure (os cordis) present in camel heart is a large elongated bone, resembling finger like projection that embedded inside the heart wall where the cardiac muscles are inserted. This resembles the elongated os penis of dog. In this study, we found the os cordis in the aortic ring; especially in the adjacent area between the aorta and the left & right atrium. It was lying in the atrioventricular plane near the junction of the interatrial and interventricular septa extending anteriorly into the atrioventricular valve rings. These findings were in agreement with Nawal et al. (1998) who reported that the heart of camel contained only one bone (os cordis). Several other research groups reported that the connective tissue skeleton present in heart might be composed of dense and irregular connective tissue in pigs and cats, fibrocartilage in dogs, hyaline cartilage in horses or bone in large ruminants (Samuelson, 2007; Junqueira et al., 2007; Budras and Habel, 2003; Chairman, 2005; Egerbacher et al., 2000; James et al., 1995).

Histologically, the camel heart skeleton was consisted of fibrous connective tissues (mainly coarse collagen fibers) illustrated in the Figures 1-5. These findings were similar to the reports of Egerbacher et al. (2000) and Egerbacher and Weber (2001) in Otter (Lutra lutra). In this study, highly vascularized connective tissues with numerous adipocytes were found that surrounded and supported the cartilage and the bony structures (Figure 1A, 1B). Besides, a well-developed ossified hyaline cartilage was clearly seen as a cap for the spongy bone (Figure 1C, 2A). The cartilago cordis was consisting of numerous chondrocytes which were surrounded by lacunae and sometimes by cell nest within the basophilic matrix. Furthermore, calcification of the matrix material occurred in the central areas of the cartilage. These deposits minerals stained deep-blue with hematoxylin. The chondrocytes present within the areas of calcification were morphologically inconspicuous vital cells (Figure 2A, 2B, 3B). Small foci of the hyaline cartilage were not clearly defined, and
Figure 1: Os cordis showing the spongy bones; (1A) Hyaline cartilage (arrow head), cardiac muscle (C). Stain: H&E (25x), (1B) Higher magnification of Figure 1A showing the adipose tissue (A), large blood vessels (arrow). Stain: H&E (200x), (1C) Higher magnification of Figure 1A showing the spongy bones (long arrow), the calcified hyaline cartilage (arrow head), bone marrow (short arrow). Stain: H&E (50x).

Figure 2: Os cordis showing the spongy bones; (2A) Photomicrograph showing the spongy bones (B), the calcified hyaline cartilage with chondrocytes nest (arrow), single chondrocytes (arrow head) and bone marrow (BM). Stain: H&E (200x), (2B) Higher magnification of Figure 2A showing the chondrocytes nest (arrow), single chondrocytes (arrow head), calcified cartilage (C). Stain: H&E (400x), (2C) Bone marrow showing the adipose tissue (A), RBCs (arrow head), osteocytes (arrow) and bone (B). Stain: H&E (400x).

The perichondrial layer was missing having unclear chondrocytes and lacunae (Figure 5A, 5B). Moreover, hyaline cartilage was continuously transformed into surrounding fibrocartilage, and subsequently transformed into coarse connective tissue (Figure 5A). These made a strong attachment and fixation of cardiac muscles with the os cordis and cartilago cordis (Figure 5A). These investigations were similar to that of Otter (Lutra lutra), reported by Egerbacher et al. (2000).

The developmental stages of bone showed conversion of normal hyaline cartilage pieces with basophilic matrix to calcified cartilage. The calcified cartilage was gradually converted to spongy bone having irregular bone lamellae. Moreover, the spongy bone revealed the presence of red and white bone marrow with numerous red blood cells, adipocytes and osteocytes. The ossification rate might be depended on the increasing rate of calcium ions precipitation (Figure 1C, 2A, 2B, 2C, 3A, 3B, 3C). These investigations were in
Figure 3: Hyaline cartilages of heart; (3A) Calcification of the hyaline cartilage (arrow head), the hyaline cartilage with chondrocytes (arrow). Stain: H&E (100x), (3B) Higher magnification of Figure 3A showing the chondrocytes nest (arrow), single chondrocytes (arrow head) with basophilic matrix. Stain: H&E (400x), (3C): Higher magnification of Figure 3A showing the conversion of the hyaline cartilage to bone with presence of chondrocytes (arrow) Stain: H&E (400x).

Figure 4: Spongy bone and hyaline cartilage of heart; (4A) Spongy bone (B), hyaline cartilage (arrow head), collagen fibers (arrow), and cardiac muscle (C). Stain: Azan (50x), (4B) higher magnification of Figure 4A showing the spongy bone (B), hyaline cartilage with chondrocytes nests (arrow head) and the collagen fibers (arrow). Stain: Azan (100x).

Figure 5: Cardiac muscle of camel; (5A) Hyaline cartilage (long arrow), fibrous CT. (short arrow), cardiac muscle (C). Stain: H&E (50x), (5B) Higher magnification of Figure 5A showing the hyaline cartilage with basophilic matrix and chondrocytes (arrow). Stain: H&E (200x), (5C) High magnification of Figure 5A showing the cardiac muscle bundles (C). Stain: H&E (200x), (5D) Cardiac muscle cells. Stain: H&E (400x).

support of Egerbacher et al. (2000) who clarified that in addition to the bones, 1-3 pieces of cartilages and cartilage with initial ossification were frequently found in Otter (*Lutra lutra*).

The os cordis was attached strongly with the surrounding cardiac muscles by coarse fibrous connective tissue mainly of collagen fibers (Figure 1A, 4A, 4B). In our study, we found that sex and size of the camel were not related with calcification rate and formation of bony structures. This investigation was in agreement with Dellmann and Eurell (1998) and Gartner and Hiatt (2007) who stated that the nature of the heart connective tissue skeleton was species and age dependent.
The cardiac myocytes were attached with the cartilage, and the bony structure was appeared in the cross sections as an irregular polygonal cells of various sizes with a large, round, pale-staining, euchromatic, centrally placed, single nucleus and rarely with binucleated cells. The cardiac muscle sarcoplasm was eosinophilic and full of contractile myofibrils (Figure 5A, 5C, 5D). Similar findings were reported by Ghonimi et al. (2014b) in the camel left atria, and Ghonimi et al. (2014a) in the camel moderator bands.

CONCLUSIONS

Os cordis is the strongest part of the cardiac skeleton that stabilizes the heart during contraction and relaxation. This bony structure is consisting of fibrous connective tissue, pieces of hyaline cartilage, calcified cartilage and large piece of spongy bone. However, in hyaline cartilage, perichondrial layer is missing, but slightly-defined chondrocytes and lacunae are present.

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