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Comparative Histology and Histometry of the Renal Capsule in Dromedary She Camel (*Camelus dromedarius*), Cow (*Bos indicus*) and Ewe (*Ovis aires*)

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ABSTRACT:

Kidneys from adult she camels, cows and ewes (eight from each) were investigated using histologic and histometric techniques to study the renal capsule. The capsule in the kidney of the three species was made up of two layers, outer and inner. The outer layer of the renal capsule was composed mainly of dense collagenous fibres arranged in wavy bundles. The outer layer was the thicker in she camel (396 µm) as compared to cow (39 μm) and ewe (36 μm). The outer layer represented about 82% of the total thickness of the capsule in she camel and cow whereas it was only 37% in ewe. The inner layer in the three species was mainly formed of smooth muscle fibers, and fine collagen and reticular fibres. The reticular fibres in the inner layer of the renal capsule were markedly extensive in ewe and cow compared to she camel. Smooth muscle fibres formed a distinct thick layer in ewe and a thin one in cow; however, in her camel they were randomly distributed. No elastic fibres were present in all capsules studied. The inner layer was observed to be thick in ewe as it constituted about 63% (61.2 µm) of the total thickness of the capsule compared to only 18% in both cow (8.4 µm) and she camel (84.3 µm). The study revealed that the renal capsule was thickest in she camel (480.4 µm) compared to ewe (97.5 μ m) and cow (47.3 μ m). It is concluded that the renal capsules of she camel, cow and ewe showed remarkable variation in thickness as well as the amount of the histological components. It is suggested that such variation might influence the function of the renal capsule.

Keywords: Histology, renal capsule, domestic animals

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INTRODUCTION

The renal capsule is a sheath covering the outer surface of kidney (Eurell and Frappier, 2006). The main function of this capsular layer is to support the kidney mass and protect kidney from possible damage and injuries (Orchard and Nation, 2015). The renal capsule also believed to assist kidney resisting fluctuation of volume and pressure that accompany variations in its function (Ross *et al.*, 1995).

It is known that the renal capsule of domestic and experimental animals has two layers, an outer layer composed of dense collagen fibres, and an inner layer of loose connective tissue with some smooth muscles (Gartner and Hiatt, 2014). However, many studies carried out on different mammalian species have shown remarkable controversy in the histological structure of the two layers forming the renal capsule, particularly the amount of the capsular connective tissue fibres and smooth muscles. In addition, several studies indicated the variation in the total thickness of the renal capsule of different animals (Bulger et al., 1979; Mbassa, 1988; Xu et al., 2009; Singh, 2013; Sikarwar et al., 2016).

Search in the literature revealed that only few studies have been carried out to compare the renal capsule of different animals (Kobayashi, 1978). Moreover, comparative studies on the renal capsule of the domestic animals are virtually lacking. Hence, the present study was conducted to investigate the histologic structures and histometric measurements of the renal capsule in she camel, cow and ewe.

MATERIALS AND METHODS

Animals and tissues: The study was carried out in adult eight apparently healthy dromedary camels, cattle and sheep of female animals slaughtered at Assalam abattoir in Khartoum state, Sudan. Five blocks of kidney samples (about 1 cm^3) were collected from the capsule and underlying cortex for each animal.

Histology: Samples were immediately fixed in 10% neutral buffered formalin and were processed for the routine histological techniques. Then, sections

were stained with Hematoxylin and Eosin for studying the general structure of the renal capsule. Connective tissue fibres were investigated by applying the following stains: Masson's trichrome for Collagen fibres; Verhoeff's stain for elastic fibres: Gomori's silver impregnation technique stain for reticular fibres. Alcian blue stain was employed to determine smooth muscles, whereas their basement membranes were demonstrated by Periodic Acid Schiff's (PAS) stain (Bancroft and Gamble, 2008).

Histometry: For histometric investigation, the thickness of the capsule was measured using a light microscope (Olympus BX63-Japan) connected to a digital camera (Olympus DP72). The measurements were carried out utilizing video image software (Cell Sens 510- Olympus) to record (in µm) the thickness of the two layers of the renal capsule. For each parameter, three readings were measured per tissue sample with a total of 120 readings from the eight animals. Mean of the readings was then calculated.

RESULTS

The capsule of the kidney in the three species was made of two layers; outer and inner (Figure 1). The outer layer of the capsule was composed mainly of dense collagenous fibres arranged in wavy bundles (Figure 2). The inner layer in the three species was mainly formed of smooth muscle fibres and to a lesser extent fine collagen and reticular fibres. Smooth muscle fibres formed a distinct thick inner layer in ewe and a thin one in cow. In the she camel, however, muscle fibres were less extensive. Generally, the basement membranes of the smooth muscles were always observed positive to PAS stain (Figure 3). In cow and ewe, the reticular fibres were extensive and arranged in parallel lines amongst the smooth muscles. In she camel, they more or less appeared as a reticulum of fine and loose fibres (Figure 4). Elastic fibres were not present in all of the capsules studied (Figure 5).



Figure 1: Light photomicrographs showing the outer layer (OC) and inner layer (IC) of the renal capsule and underlying cortex stained with H&E.



Figure 2: Light photomicrographs showing the connective tissue (CT) of outer layer of the renal capsule consisting of dense irregular collagen fibres stained with Alcian blue (she camel) and Masson's trichrome (cow and ewe). Fine collagen fibres (arrows) were scattered in the inner layer of renal capsule.



Figure 3: Light photomicrographs showing the PAS-positive basement membranes (arrows) of smooth muscle cells of the renal capsule in the three species studied. PAS stain.



Figure 4: Light photomicrographs showing the reticular fibres (arrows) clear in the inner layer of the renal capsule of ewe and cow compared to she camel. Silver impregnation technique.



Figure 5: Light photomicrographs showing the absence of elastic fibers in renal capsule of the three species. Verhoeff's stain.

As it can be seen in Table 1, the renal capsule of she camel was the thickest (480.4 μ m) when compared to ewe (97.5 μ m) and cow (47.3 μ m).

The outer layer of the renal capsule of she camel was remarkably thicker (396 μ m) than those of cow (39 μ m) and ewe (36 μ m). This layer also represented

about 82% of the total thickness of the capsule in she camel and cow but only 37% in ewe. The inner layer was observed remarkably thick in ewe as it constituted about 63% (61.2 μ m) of the total thickness of the renal capsule compared to only 18% in both cow (8.4 μ m) and she camel (84.3 μ m).

Table 1: The thickness $(\pm SD)$ and percentage of the inner and outer capsule of she camel, cow and ewe.

Animal	Measurements			Percentage (%)		
	OL	IL	ТС	OL	IL	ТС
She camel	396.1±53.1	84.3±11.1	480.4 ± 58.8	82.4	17.6	100
Cow	39.1±5.1	8.4 ± 2.3	47.5±5.2	82.2	17.8	100
Ewe	36.3±5.4	61.2±6.5	97.5±7.2	37.3	62.7	100

OL: Outer layer IL: Inner layer TC: Total thickness of the renal capsule

DISCUSSION

The current study showed that, the kidneys of the three species were covered by connective capsule composed of outer and inner layers. Similar findings were reported by Abdalla (1973), Xu *et al.* (2009) and Singh (2013).

In camel, the present investigation showed that the inner layer had scattered smooth muscle fibres. In ewe and cow, unlike she camel, the smooth muscles were distinct and formed most of the inner layer of the renal capsule. The presence of such muscle fibres was confirmed by their positive PAS basement membranes. This is in agreement with the previous accounts given by Abdalla (1973) and Xu et al. (2009) in camel and Bacha and Bacha (2000) in domestic animals who reported the presence of smooth muscles within the renal capsule. Furthermore, smooth muscle fibres were also reported in experimental human and animals (Gartner, and Hiatt, 2007; Al-Samawy, 2012; Al-Jebori et al., 2014). In contrast, Zaghloul (1999) and Wenhui and Huaitao (2000) stated that smooth muscles were not present in the entire renal capsule of camel.

Several studies have shown that the camel produces concentrated urine (Siebert, and Macfarlane, 1971; Kataria *et al.*, 2007). In rat, it has been suggested that the renal capsule affects renal interstitial hydrostatic pressure (Khraibi and Knox, 1989). It was reported that the increase of this pressure resulted in elevation of sodium and water excretion (Granger *et al.*, 1988; Farrugia *et al.*, 1992). Moreover, Kobayashi (1978) suggested that the intrarenal pressure was adjusted by the contraction of the smooth muscle cells of the renal capsule. Therefore, it is plausible that the few

amount of capsular smooth muscles of she camel in the present study might reflected on reduction of the renal interstitial pressure; and subsequently decreasing the amount of and increasing concentrating of urine.

In the present study, reticular fibres were observed within the renal capsule, mainly the inner layer, of the three species studied. The reticular fibres were also reported in the renal capsule of sheep (Singh, 2013) and rabbit (Al-Jebori *et al.*, 2014). It had been stated that reticular fibres were the main components of the basement membranes surrounding and supporting muscle cells (Kulkarni, 2012). Thus, it is suggested that reticular fibres in the present study were most likely related to the basement membranes of the smooth muscle.

The renal capsule of all domestic species in the current study revealed no elastic fibres. This finding is in accord with Frohmüller et al. (1987) who stated that the renal capsule is extremely inelastic. The main function of the elastic fibres is to provide connective tissues with elasticity and resilience properties (Kielty et al., 2002). Therefore, the lack of elastic fibres in the present study may indicate limited elasticity of the renal capsules currently investigated. On the contrary to the present findings, the presence of elastic fibres within the renal capsule was reported in sheep Singh (2013) and human (Gartner and Hiatt, 2007: Orchard and Nation, 2015). Further immunohistocemical studies are needed to investigate the presence, or absence, of the elastic fibres within the renal capsules of camel, cattle and sheep. The present study revealed great variation in the thickness of the renal capsule of she camel (480 µm), ewe (98 μ m) and cow (48 μ m). The variation in the thickness of renal capsule was also

evident in other animals. For instance, the total thickness of the renal capsule measured 347 μ m in bacterian camel (Wenhui and Huaitao, 2000), 750 μ m in giraffes (Maluf, 2002) and reached up to 900 μ m in dog (Bulger *et al.*, 1979).

According to the arrangement of the medullary pyramids, and its associated cortex, it is well known that the kidneys of the domestic animals are of different types (König and Liebich, 2004; Eurell and Frappier, 2006). In this regard, the three species in this study have different types of kidneys. The she camel (Abdalla, 1973) and ewe (Banks, 1993; König and Liebich, 2004) have unipyramidal kidneys that are characterized by a smooth external surface, whereas the kidney of cow is multipyramidal with a lobulated surface. Interestingly, although the she camel and ewe have the same type of kidneys, yet their renal capsules showed a noticeable difference in the total thickness of capsule and the percentage of the outer and inner layers. On the other hand, despite the fact that the she camel and cow have different types of kidneys, yet they showed similar percentages in the thickness of the outer and inner layers of the renal capsule.

It is concluded that the renal capsules of she camel, cow and ewe showed remarkable variation in thickness as well as the amount of the histological components. It is suggested that such variation might influence the function of the renal capsule.

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