Morphological Studies on Epididymis and Vas Deferens of One - Humped Camel Bull (*Camelus dromedarius*), Uda Ram and Red Sokoto Buck

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To cite this article:

Muhammad Abdullahi Mahmud, Josephat Onu, Sani Abdullahi Shehu, Aminu Umaru, Abubakar Danmaigoro, Mohammed Shaibu Atabo. Morphological Studies on Epididymis and Vas Deferens of One - Humped Camel Bull (*Camelus dromedarius*), Uda Ram and Red Sokoto Buck. *American Journal of Bioscience and Bioengineering*. Vol. 3, No. 5, 2015, pp. 65-71. doi: 10.11648/j.bio.20150305.17

Abstract: This study was aimed at comparing morphology of the epididymis and vas deferens of One - humped Camel bull (OCB), Uda ram (UR) and Red Sokoto buck (RSB). Fifteen testes and vas deferens were collected, organs grossly examined, measured for weight or length and processed for histology. In OCB, epididymal tail was the largest of the three segments while a reverse of that was obtained in UR and RSB. In OCB, the vas deferens was found to be coiled all through while in both UR and RSB, it was found to be highly coiled initially but became straightened as it coursed down to form ampulla. Gross morphometrically, the weight and length of epididymis and vas deferens in the three species differed significantly ($P < 0.05$) from one another. Histomorphologically, in the corpus epididymis, stereocilia were prominently observed in UR followed by RSB and least in OCB. The proximal segment of vas deferens in the three species was found to consist of three histological layers; tunica mucosa from which, many folds extended, tunica muscularis and tunica serosa. Histomorphometrically, all measured parameters in both corpus epididymis and proximal segment of vas deferens in the three species differed significantly from one another. It was concluded that although results show that the studied animals are different ruminant species they exhibit some similarities and interesting morphological differences in epididymis and vas deferens compared to the majority of mammals. The basic morphological characterizations done in this study are important for future studies, such as comparison with other species of ruminants (whether true or pseudo).

Keywords: Morphology, Epididymis, Vas Deferens, Gross, Morphometry, Histomorphology, Histomorphometry

1. Introduction

The importance of epididymis and vas deferens in sperm production, storage and maturation and sperm transport respectively in domestic animals cannot be overemphasized, since survival of a species largely depends on its ability to reproduce its own kind [1]. This might be the reason why they received more attention especially the epididymis than any other segments of the excurrent duct of the testes. Recent studies on mammalian epididymis include those by [1, 2, 3, 4, 5, 6, 7, 8]. Vas deferens on the other hand had received very little attention recently especially in one - humped

camel, sheep and goat [9, 10]. The epididymis is a highly convoluted tubule which connects the testis to the ductus deferens and is an important segment of the excurrent duct system of the testes that performs a variety of functions [11]. Various studies on mammalian epididymis have shown that it can be divided into distinct regions according to the biochemical, morphological and morphometric characteristics of its segments [12]. Various divisions have been proposed and the most widely used is that dividing the organ into the initial segment, caput, corpus and cauda epididymis [13]. The vas deferens is the tubular structure which conducts spermatozoa from the epididymis to the urethra. After crossing the ureter in the abdominal cavity it

dilates into a spindle shaped enlargement, the ampulla [9]. Comparative anatomical and histological studies on epididymis and vas deferens of local domestic ruminants are not available, except few references that are available in the textbook on ruminants in general [14, 15]. Therefore, this work highlights comparatively; gross anatomy, gross morphometry, histomorphology and histomorphometry of epididymis and vas deferens in one - humped Camel Bull, Uda ram and Red Sokoto buck.

2. Materials and Methods

Fifteen testes and vas deferens of apparently healthy adult one - humped Camel bulls, Uda rams and Red Sokoto bucks (five samples per species) were collected from Sokoto metropolitan abattoir. Sokoto metropolis is located on latitudes 10° N and 14° 50' N and longitudes 7° E, east of the equator, in the extreme northwest of Nigeria. It covers an area of approximately 2, 823, 237 square kilometres. The last National census reported the state population to be 3, 696, 999 [16]. Following the collection, they were transported to Veterinary Anatomy Laboratory, Usmanu Danfodiyo Sokoto, Nigeria where gross features of the epididymidis and vas deferens were examined and recorded. The epididymis was dissected out from the testes. The weights (g) and lengths (cm) were measured using a weighing balance (Shimadzu AW320, Germany), metre rule and thread respectively. Photographs were taken using digital camera (Samsung ES95, 16.2 megapixels). The sample tissues for histology in epididymis were obtained from the body (corpus) while that of vas deferens was taken from the proximal region (between the vaginal ring and ampulla). They were immediately fixed inside 10% neutral buffered formalin, labelled and kept for two days, followed by preservation in 70% ethyl alcohol. They were dehydrated through ascending grades of ethanol (70%, 95% and absolute ethanol), cleared in xylene and embedded in paraffin wax. Serial sections of 5µm were cut and stained with Haematoxylin and Eosin (H &E) [17]. Micrographs were conducted with a Light microscope connected to a video based, computer – linked system (Tuscan CMOS Camera: IS500, Resolution: 5.0 megapixels) at x400 magnification. Histomorphometry was equally done using the same programme software. The following measurements were performed in epididymis: epithelial height (excluding the stereocilia), tubular and luminal diameters and stereocilia height. In the vas deferens, the following measurements were done: thickness of tunica mucosa, thickness of tunica muscularis, thickness of tunica serosa and luminal diameter. The analysis was performed at x100 magnification with a scale bar of 100µm, for both corpus epididymis and proximal vas deferens segments. The epididymal measurements were carried out according to the procedure of [12]. Briefly, the epithelial height was taken as the linear length of the principal cells, from the base of the epithelium (basal lamina) to the apical edge (excluding the stereocilia), the luminal diameter as the longest measurement from one apical edge to the other, and the tubular diameter as

the longest distance between basal - basal laminae

Data analysis: Epididymis and vas deferens data obtained were expressed as Mean ± SEM (Standard Error of Mean) and subjected to statistical analysis using Statistical Package for the Social Sciences (SPSS) version 17.0. One - Way Analysis Of Variance (ANOVA) at 95% confidence interval (CI) was used to determine level of significant difference in mean values of the data. Values of ($P \leq 0.05$) were considered significant. Where there were differences in means, they were separated by Tukey's Honestly Significant Difference (HSD).

3. Results

3.1. Gross Morphology

3.1.1. Epididymis

The epididymides of the three species are shown on Plates 1, 2 & 3. The epididymis was observed in the three species to be an elongated and convoluted tube that lies firmly attached laterally to the testis. The convoluted tube covers the superior pole of the testis and connects by a narrow corpus epididymis to cauda epididymis projecting from the distal border of the testis. In all the three studied species, the epididymis was macroscopically divided into three main segments; caput (head), corpus (body) and cauda (tail). In the one - humped camel bull, the tail was found to be the largest of the three segments, with the head curving on the cranial pole of the testes. The entire epididymis was loosely attached to the testes, with a soft consistency. In the Uda ram the epididymal head was found to be the largest of the three segments with the head markedly outlined and pointed dorsocranially with a faint neck outline. The terminal portion of the body was found not be attached to the testes and the tail was found to be attached in a caudodorsal direction. In the Red Sokoto buck, the general characteristic described above for the Uda was observed except that the head had a distinct neck with a smaller testicular bursa around the terminal portion of the body. The colour of epididymis was yellowish in both Uda rams and Red Sokoto buck though more in the buck, while that of the one - humped camel bull was greyish white.

3.1.2. Vas Deferens

The vas deferens of the three species is shown on Plates 1, 2 & 3. The vas deferens was observed in the three species to be a continuation of the caudal epididymis which was more straightened, more tubular and more muscular than the epididymis. It extends from the tail of the epididymis to the pelvic urethra. In the one - humped camel bull, the vas deferens was found to be coiled all through but more before it became enlarged at its terminal portion to form an ampulla, ventral to the prostate. In both Uda ram and Red Sokoto buck, the vas deferens was found to be highly coiled initially but became straightened as it coursed down to form ampulla. The vas deferens was observed to be longest in one - humped Camel bull, followed by that of the Uda ram and least in the Red Sokoto buck.

Plates 1, 2 and 3. Photograph of testes of OCB. 2. Photograph of testes of UR. 3. Photograph of testes of RSB, showing testes (T), epididymal head (EH), epididymal body (HB), epididymal tail (ET) and vas deferens (VD).

3.2. Gross Morphometry

Means $\pm$ SEM of weights and lengths of epididymis and vas deferens of One - humped Camel bull (OCB), Uda ram (UR) and Red Sokoto buck (RSB) are shown in Table 1.

3.2.1. Epididymis

The results of the mean epididymal length of OCB, UR and RSB indicated that the means differed significantly ($p \leq 0.05$) in the three species. The mean epididymal length of 19.00 ± 1.72 cm in UR differed significantly ($p \leq 0.05$) from those of OCB (14.88 ± 1.33 cm) and RSB (13.06 ± 0.35 cm).

The results of the mean epididymal weight of OCB, UR and RSB indicated that the means differed significantly ($p \leq 0.05$). The mean epididymal weight of 46.55 ± 1.28 g in OCB was significantly ($p \leq 0.05$), the highest in the three species, followed by that of UR (24.21 ± 3.67 g) and least in RSB (7.18 ± 0.35 g).

3.2.2 Vas deferens

The results of the mean vas deference length of OCB, UR and RSB indicated that the means differed significantly ($p \leq 0.05$). The mean vas deferens length of 35.67 ± 1.23 cm in OCB was significantly ($p \leq 0.05$), the highest in three species, followed by that of UR (29.81 ± 0.76 cm) and least in RSB

(15.00 ± 0.53 cm).

Table 1. Mean $\pm$ SEM of weights (g) and lengths (cm) of different parts of Reproductive Tracts of the one - hump camel bull (OCB), the Uda ram (UR) and Red Sokoto buck (RSB).

Parameters	OCB	UR	RSB
EL	$14.88 \pm 1.33b$	$19.00 \pm 1.72a$	$13.06 \pm 0.35b$
EW	$46.55 \pm 1.28a$	$24.21 \pm 3.67b$	$7.18 \pm 0.35c$
VDL	$35.67 \pm 1.23a$	$29.81 \pm 0.76b$	$15.00 \pm 0.53c$

a, b, c Means within the same row without the same superscript letters are significantly different ($p \leq 0.05$) from each other. EL=Epididymal length, EW= Epididymal weight, VDL = Vas deferens length.

3.3. Histomorphology

3.3.1. Corpus Epididymis

The mucosae of the corpus epididymis were found to comprise pseudostratified columnar epithelium with stereocilia, basal cells at the lamina propria, muscularis, vascular connective tissue as well as spermatozoa in the lumen. The stereocilia were found to be more extensive in UR than in the RSB and least prominent in the OCB (Plates 4, 5 and 6).

Plates 4, 5 and 6. Micrographs of epididymis (transverse sections) of OCB, UR and RSB, showing the mucosa lined by pseudostratified columnar epithelium (P) with stereocilia (S), muscularis (M), and presence of spermatozoa (SP) in the epididymal lumen (H & E x400, Scale bar = 100 μ m).

3.3.2. Proximal Vas Deferens

The vas deferens was observed to be thick with mucosal folds, resulting in irregular outline of the lumen. Also observed were thick layers of smooth muscle of muscularis

externa and serosa. The mucosa was observed to be thickest in OCB and least in RSB. The number of mucosal folds was observed to be highest in UR and least in OCB (Plate 7, 8 and 9).

Plates 7, 8 and 9. Micrographs of vas deferens (transverse section) of OCB (7), UR (8) and RSB (9), showing lumen (L), mucosal folds (F), thick layer of smooth muscle of muscularis externa (E) and adventitia (A) (H & E x40, Scale bar = 100 μ m).

3.4. Histo - Morphometry

The various histomorphometric dimensions of the corpus epididymis and the proximal vas deferens in the three studied species are shown in Figs. 1 and 2 respectively.

Fig. 1. Histomorphometric parameters of Corpus Epididymis.

3.4.1. Corpus Epididymis

The mean epithelial height of RSB ($173.26 \pm 19.46 \mu\text{m}$) significantly differed ($p \leq 0.05$) from those of UR ($76.84 \pm 3.28 \mu\text{m}$) and OCB ($107.71 \pm 7.06 \mu\text{m}$).

The mean tubular diameters of $776.21 \pm 23.03 \mu\text{m}$ in OCB and $742.80 \pm 21.97 \mu\text{m}$ in RSB significantly differed ($p \leq 0.05$) from a mean value of $626.00 \pm 30.42 \mu\text{m}$ recorded in UR.

The mean luminal diameters of $611.80 \pm 32.25 \mu\text{m}$ and $491.62 \pm 34.95 \mu\text{m}$ in OCB and RSB respectively significantly

differed ($p \leq 0.05$) from each other while that of UR ($531.07 \pm 29.82 \mu\text{m}$) did not differ ($p > 0.05$) from any of them.

The stereocilia heights of $150.61 \pm 14.53 \mu\text{m}$ and $130.48 \pm 15.45 \mu\text{m}$ in UR and RSB respectively did not differ significantly ($p > 0.05$) from each other. However, it is numerically higher in UR than RSB.

3.4.2. Proximal Vas Deferens

The sizes (thickness) of tunica mucosa and tunica muscularis of proximal vas deferens in the three species were found to be significantly different ($p \leq 0.05$) from one another. RSB was found to have the thickest t. mucosa and t. muscularis with mean values of $257.74 \pm 30.88 \mu\text{m}$ and $1202.29 \pm 49.31 \mu\text{m}$ respectively. This was followed by UR

with mean values of $180.60 \pm 27.29 \mu\text{m}$ and $755.25 \pm 70.85 \mu\text{m}$ respectively. The least values of $92.47 \pm 11.29 \mu\text{m}$ and $560.74 \pm 51.60 \mu\text{m}$ respectively were recorded for OCB.

The mean thickness values of the t. serosa of $245.88 \pm 17.90 \mu\text{m}$ and $401.87 \pm 60.45 \mu\text{m}$ in UR and RSB respectively were found to be significantly ($p \leq 0.05$) different from that of OCB ($1561.41 \pm 92.07 \mu\text{m}$).

The mean values for vas deferens luminal diameter though not significantly ($p > 0.05$) different in the three species, numerically, the OCB had highest mean value of $1567.77 \pm 534.37 \mu\text{m}$, followed by UR with a mean value of $1327.79 \pm 241.90 \mu\text{m}$ and least in RSB with a mean value of $1209.36 \pm 37.55 \mu\text{m}$.

Fig. 2. Histomorphometric parameters of Vas Deferens.

4. Discussion

The macroscopic division of the epididymis in the three species into three segments; the head (caput), the body (corpus) and the tail (cauda), follows general pattern as earlier described by [18] in mammals, [19] in one - humped camel and [14] in small ruminants. In this present study, the OCB epididymal tail which was found to be the largest of the three segments agrees with the reports of [19] in OCB. The result however, is contrary to the earlier reports of [20], who reported that the body of the epididymis of one - humped camel accounts for almost 50% of the total epididymal weight. It is also not in agreement with the reports of [21] and [22] in bulls and bucks respectively, who reported that the caput is the largest of the three segments.

The result of this study on the vas deferens of OCB found to be coiled all through, but more before it became enlarged at its terminal portion where it formed ampulla, agrees with the reports of [23] who said, the vas deferens in one - humped camel is coiled initially. He further said its distinct ampullary enlargement is present in the terminal 4.0 - 5.0 cm which enters the urethra ventral to the corpus prostate. The present finding is however, contrary to the reports of [24], who reported that, the vas deferens in one - humped camel is

remarkably twisted for much of its initial course, but becomes fairly straight towards the end portion.

The mean epididymal weight ($24.21 \pm 3.67 \text{ g}$) of the UR obtained in this study is close to the mean value of $25.5 \pm 0.28 \text{ g}$ earlier reported by [25] in Nigerian Uda ram but higher than mean weights of $134.48 \pm 2.28 \text{ g}$ and $19.12 \pm 0.12 \text{ g}$ respectively reported by [26] for West African dwarf ram. This difference may be attributed to genotype, as the West African dwarf ram had been described as the smallest breed of indigenous sheep in Nigeria [27].

The mean length of vas deferens of $35.67 \pm 1.23 \text{ cm}$ found in this study for OCB is below the range of 45 - 50 cm reported by [19] in one - humped camels. The mean vas deference in RSB of 15.00 ± 0.53 found in this report is contrary to the earlier reports by [9] of $29.86 \pm 1.38 \text{ cm}$, $31.71 \pm 0.84 \text{ cm}$ and $40.18 \pm 1.59 \text{ cm}$ in pre - pubertal, pubertal and post pubertal Gaddi goats respectively.

The findings on least prominence of stereocilia observed in the corpus epididymis of OCB agree with earlier reports of [28] in one - humped camel. [29] have mentioned that, in one - humped camel, the proximal part of the middle epididymis possesses high epithelium with long stereocilia, the intermediate part is characterized by the cytoplasmic vacuoles, mass collections of spermatozoa in the lumen and

short stereocilia. Whereas the distal part of the middle epididymis, is the longest of all the segments and extended over most of what is classically described as the corpus epididymidis. It is characterized by low epithelium; the lumen is very wide packed with spermatozoa and shortest stereocilia. This could mean that there might be less absorptive and secretory functions of the tubular epithelium of the corpus epididymis in OCB compared to the corpus epididymides of the other two species.

The results on histomorphometry could not be adequately compared to the findings of other workers due to paucity of available relevant literature. However, some histomorphometric results obtained in this study are of much higher values than what were earlier observed in Malabari goats [30], Black Bengal bucks [31] and camel [29]. The different results may be due to sampling from different positions, different magnification or different calibrations.

5. Conclusions

The results show that although the studied animals are different species of ruminant they exhibit some similarities and interesting morphological differences in epididymis and vas deferens compared to the majority of mammals. The basic morphological characterizations done in this study are important for future studies, such as comparison with other species of ruminants (whether true or pseudo).

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