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Histomorphometric study of testis and epididymis of adult one humped camel *Camelus dromedarius* in AL-Muthanna province

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Abstract

Objective: This study aimed to focus light on the morphological and histological features of the testis and epididymis of mature camel. Materials and method; the total samples were fourteen testis and epididymis from adult healthy camel males collected from AL-Muthanna abattoir, seven samples to each morphological and histological study. Grossly: The testis of camel was elongated oval located within scrotum in the perineal region and directed caudo-dorsally, the epididymis was divided into head, body and tail. The testis and epididymis in left were larger than that in right. Histologically: The testis consists of a mass of seminiferous tubules, surrounded by fibrous capsule called tunica albuginea; which composed of dense collagenous tissue. A trabecula, pass inward from the tunica albuginea to form a framework for support of the seminiferous tubules gives off several trabeculae into the interior of the testis. These trabeculae divide the testis into lobules. Each lobule contains many seminiferous tubules. This capsule is covered by a tunica serosa, the connective tissue of which blends with the tunica albuginea. The tunica albuginea is continuous with the loose collagenous tissue of the mediastinum testis. Tunica albuginea is also continuous with the testis into the lobule testis; seminiferous tubules and rete testis are surrounded by loose collagenous tissue; Sertoli cells are elongated cells with irregular outlines extending from the basement membrane to the lumen. Leydig cells occurred in the interstitial space between the seminiferous tubules. The seminiferous tubules are associated

with a specialized type of myoid cell. These cells are located within the surrounding basal lamina. Spermatogonia undergo mitosis to produce A-spermatogonia, Intermediate spermatogonia and B spermatogonia. This cyclic mitotic division activate B spermatogonia to produce primary spermatocyte that pass through six different phases; preleptotene, leptotene, zygotene, pachytene, diplotene and diakinesis phases. The secondary spermatocyte enters the second meiotic division which gives rise to spermatids. Differentiated by fourteen stages which lead to form sperm; the epididymal duct was lined with pseudostratified columnar. We can conclude that the study has shown the significant differences in terms of morphometry, and histology of right and left testes of camel.

Key words; Camel, seminiferous tubules, Sertoli cells, epididymis; Leydig cell.

INTRODUCTION

Camels are one of the large animals on earth that are adapted to survive in the tough conditions of desolate environments (1). Camels are used for many purposes that are important in human life, including providing him with food (meat and milk) as, camel meat is one of the important tributaries of animal protein in the Arab countries, which depends on it to fill an important share of the growing demand for meat, especially in areas with poor pastures that can be better used in camel breeding, as small camel meat is comparable to veal meat in terms of taste quality and texture (2,3). Camel breeding in the country did not receive its abundant share of scientific studies for the purpose of raising the efficiency of its production and improving its performance, especially in the southern region, which is characterized by the spread of a good breed in it (4). The

camel is known to be a seasonal breeder, the male camel shows sexual activity during specific days of the year (breeding or rutting period), and have some aspects of the seasonal changes in the testis (5). Sexual behavior in male farm animals, including camel, is an expression of both sexual desire and the ability to mate. Male sexual behavior depends on the degree of sexual stimulation and preparation, which are greatly influenced by external stimuli such as sight, hearing, smell, and other stimuli (6-11). The male reproductive system in camel consists of the testes, which are the main glands or gonads of the male, and the vas deferens, which include the epididymis, the vasa deferens, and the penis. It also consists of accessory gonads (the prostate, Cowper's glands, the bulbourethral glands), which maintain the sperm, nourish them, and

produce the seminal fluid in which the sperm swim (12-15). Increasing animal output is mostly dependent on management and reproduction. Because it affects the number of children produced each year, reproductive efficiency is therefore one of the most important economic characteristics. A prevalent issue is low reproductive efficiency. Maintaining optimal reproductive performance requires а thorough understanding of a woman's genital organs, and using modern reproductive technologies requires knowledge of the organs' measures (16). Our knowledge of the camel's reproductive biology has also grown as a result of this study, which again highlights the need for more research and may aid in the identification and, consequently, the best possible treatment of the illnesses. In order to supply data for further research, this study attempts to shed light on the camel's testicles.

MATERIALS AND METHODS

Animals: For the morphological and histological study of the testis and epididymis conducted on 14 pairs of testes along with epididymis, healthy samples from adult camel (6-7 years) from slaughter in the Muthanna. Take five samples (0.5-0.7

mm) from different regions from each testis and fixated with 10% formalin and the testis samples were taken for histological analysis and many stains used H@E, Masson Trichrome, Verhoef's and Van Gesion (17).

STATISTICAL ANALYSIS; A one way analysis of a variance (ANOVA) tests was used to examine the study at the 5% significant levels. Data were processed and controlled using social science statistical tools (4).

RESULTS AND DISCUSION

The camel's left and right testes were long, oval-shaped, situated near the body in the perineal region, partially hidden by the animal's tail. They have a long axis that is oriented caudodistal obliquely, are somewhat compressed laterally, and are utterly and comparatively little in respect to the animal's size. Each testis contains two extremities (proximal and ventral), two surfaces (lateral and medial), and two borders (anterior and posterior) (Fig. 1). Because to its interaction with the septum scrota, medial surface was flattened and the lateral surface was convex. The epididymis is firmly linked to the posterior edge, while the anterior border is convex and free. The tail of the epididymis, which is connected to the ventral extremities by the epididymal ligament, covers the latter, which is somewhat thicker than the proximal (Fig.

1,2). This outcome was comparable to the one shown by (4,5) in camel and (14) in gazelle.

The left testis was larger and longer than the right (Fig. 1,2), these finding was in partial harmony with the findings of (3) resemble that in camel. Whereas (6) described that in goat both testes were elongated, (18) said that the paired testes of the alpacas had an oval shape and that the morphometric values of the left testis were higher than the right testis. (19,20) described the left testis were higher than the right testis in ram. While (21) stated that about size and weight vary considerably during the deferent season in sheep. (11) explained that the morphologically no considerable difference were found in right and left testicle in male goat.

The head is curved on the testicular cranial pole and is made up of a long epididymal duct distally and the efferent ductulus proximally. The dromedary camel epididymis is situated on cranial testicular border and goes from the lower end to approximately above the upper border. The three components of the epididymis are the head, body, and tail. It is shaped like tiny, extremely convoluted tubules (Fig. 2). (5) have demonstrated that camels have higher epididymal weight during the breeding period than during nonbreeding season. This could be explained by the fact that the animal's reproductive activity only increases during specific months when the quantity of spermatozoa in the epididymis rises, increasing the weight and volume of the organs relative to the nonbreeding season. Additionally, the testis and epididymis echotexture picture shows a ductulus and an increase in volume throughout the mating season (1).

asymmetrical capsule of A dense, connective tissue is formed by tunica albuginea of camel's testis. Collagen fibers make up the majority of it, with a little amount of elastic fibers, tunica albuginea has continuous connective tissue trabeculae, and there are occasionally smooth muscle cells present. The mediastinum testis is where the septula testis converge. These collagen fiber-based, narrow, and frequently incomplete trabeculae split the testis into a variable number of testicles lobules and contain arteries and nerves. The intralobular connective tissue and the septula are continuous (Fig. 3), this agrees with (22,23).

A layer of collagen fibers follows the basal lamina that envelops each convoluted seminiferous tubule. The parenchyma makes up the majority of the camel testis. The testis's parenchyma has a yellowish white hue. In the direction of the mediastinum, the collecting ducts and seminiferous tubules

converge. On their way to the mediastinum, the ducts anastomose, efferent ducts merge near the dorsal pole to form a single duct that connects to the epididymis by passing through the tunica albuginea. The tunica albuginea, a capsule made of thick collagenous tissue, encloses the testes. The connective tissue of the tunica serosa, which covers this capsule, melds with that of the tunica albuginea. The loose tissue of mediastinum is connected with the tunica albuginea. The testis and tunica albuginea continue into the lobules. There are reticular numerous fibers and loose collagenous tissue around the tubules of the rete testis and seminiferous tubules (Fig. 4-10), this agrees with results of (24,25). Testicular tissue is covered by a thick layer of dense, fibrous tissue called the tunica albuginea. The left and right testis had significantly different tunic mean thicknesses ($p \le 0.05$). The parenchyma was divided into lobules by thin septa that ran from tunica albuginea to mediastinum testis. Each lobule had tortuous seminiferous tubules. The left testicle's rounded seminiferous tubule had a significantly (P<0.05) greater mean diameter than the right testicle's (Table 1).

The seminiferous tubules are made up of seminiferous epithelium, a distinct basal lamina, and a tunica of the fibrous

connective tissue. Fibroblasts are found in multiple layers within the fibrous connective tissue tunic that envelops the seminiferous tubules. Flattened myoid cells with smooth muscle properties make up the innermost layer that adheres to the basal lamina (Fig. 7,8,9). The testis's interstitial tissue is formed by the connective tissue filling up the gaps between the seminiferous tubules. Numerous tiny blood vessels are also present in this interstitial tissue. interstitial tissue includes huge polyhedral cells, mast cells, macrophages, and fibroblasts in addition to tiny bundles of collagen fibers. These big cells have a single huge, spherical nucleus and are known as Leydig cells (Fig. 8). Spermatogenic cells and spermatozoa are found inside the seminiferous tubules. The heads of mature spermatozoa and spermatids are lodged in Sertoli cells, which also line the tubules. These cells have big, irregular nuclei with scattered chromatin and copious amounts of cytoplasm. There are several Leydig cells with round to oval nuclei and thin cytoplasmic sheets in between these seminiferous tubules. Little lipid droplets occupy the Leydig cell's little amount of cytoplasm. In the interstitial cell, several sizable lipid droplets also gather (Fig. 5,8,9), this agrees with (26,27).

Sertoli cells have an oval nucleus and a broadly pyramidal shape (Fig. 9,10). The

spermatogonia had round, black nuclei and were shaped like cubes or circles. Primary spermatocytes have granular chromatin aggregates and big spherical nuclei. The elongated spermatids developed from the little spherical spermatids, the spermatids and spermatozoa are present in certain seminiferous tubules. Up to three tubules may be separated by enormous quadrangular interstitial voids, or two seminiferous tubules may be separated by thin strands of interstitial tissue. blood capillaries, a small number of Leydig cells, a few fibroblast cells and blood vessels. Leydig cells were discovered either alone or in clusters within intertubular gaps (Fig10). The thickness of the capsule, diameter of seminiferous, and tubular differential index, were higher in the left testis than the right testis (Table 1). In the direction of the basal lamina were the spermatogonia. Spermatogonia could be categorized as typical spermatogonia type A, typical spermatogonia type B, and "intermediate" types based on the length of the area of contact with the basal lamina, the shape of cell and nucleus, the number of chromatin encrustations in nucleus. Spermatogonia were primarily classified according to their cell and nucleus shapes as well as their area of contact with basal

lamina (Fig. 8). this similar to results of (5) in camel and agrees with (29,30) in rams.

Known as spermatogonia type (A), these cells are large, flat, and feature a broad area that reaches the basement membrane. The Sertoli cells cover surrounding the remaining surface. Different cells have varying cytoplasmic densities. The nucleus is tiny and has an elliptical or circular shape, nuclear cytoplasm has a make up of soft granules and is uniform. One side of the nucleus contains a certain amount of heterochromatin. The cytoplasm will have a dusty appearance, and the nucleus will have a central location. The nuclear membrane of the nucleus is narrow and transparent (Fig 6). In general, the primordial spermatocytes are similar to those in the bull (8,24). (31,32) point out that the Golgi apparatus is highly noticeable in camel spermatocytes, and that proacrosomal granules are found in huge proacrosomal vesicles during the pachytene stage..

Cells of the spermatogonia type (I) are regarded as mediates. It has a varied form and size. Among the differences is the chromatin distribution pattern. Small granules or a few chromatin flakes in the nucleus' center are examples of the dispersed chromatin that can be observed in nuclear cytoplasm (Fig. 6,7). Spermatogonia type (B) is produced by the subsequent

intermediate divisions at mitosis of spermatogonia. Because it has a large elliptical or circular nucleus and more peripheral heterochromatin than spermatogonium (A). it is type comparatively larger than the latter. These chromatin flakes are distributed at various distances and form a crust along the nuclear membrane. The nuclear cytoplasm contains fragments of the chromatin of these big flakes. There is less homogeneity in the chromatin. Near the nuclear membrane, there are just one or two nucleoli (Fig. 7) as result of (32).

Columnar pseudostratified containing stereocilia lined the epididymal ducts. Columnar ciliated cells with signs of apocrine secretion multiplied along the efferent ductulus in the direction of the epididymal duct. There were tall, thin, cells in the epithelium at the epididymal duct junction. As one approached the epididymal duct, frequency of layers increased. The efferent ductulus was surrounded by a smooth muscle. Alkaline phosphatase reactivity was seen in the subepithelial connective tissues. The phosphatase enzymes showed strong granular activity whole throughout the epithelium Mammals' epididymis (Fig.11,12). is believed to be a crucial component of the extra-testicular sperm pathway. Along the lateral side of the testicle, there is a thick mass of a tightly wound tube. It also the sperm acquire motility in addition to being the main site for sperm maturation (6,8,9).

Capsule of testis		Diameter of convoluted		Diameter	of efferent	Diamet	ter of
		Seminfeorus		duct		epididymis	
						duct	
Left	Right	Left	Right	Left	Right	Left	right
272.3±0.	257.1±0.3	180.4±0.3	158.1±0.5B	182.6±0.	177.2±0.3	192.6	178.6±

Table, (1): Measurements each of testis and epididymis of ram, µm Mean ±standard error

2A	В	А		1A	В	±0.1	0.1B
						А	
Values with conital latters in some measure denote to significant differences (P>0.05) between							

Values with capital letters in same measure denote to significant differences (P>0.05) between left and right testis

Table (4):	Thickness	of epithelium	in parts	of epididymis	of camel,	µm Mean	±standard
error							

Measure	epithelium		T-test	Sig.
Part	Left	Right		
Head	863.3±0.5A	857.1±0.1B	12.7	0.000
Body	372.0±0.2B	363.3±0.4D	9.72	0.000
Tail	347.6±1.1A	335.6±1.3C	2.16	0.000

Values with capital letters in same measure denote to significant differences (P>0.05) between right and left epididymis



Fig. (1): Gross section showed the outer observation of testis; A- Right testis, B-Left testis, C- lateral surface, D- medial surface, E- epididymis

Fig. (2): Gross section showed the outer observation of left testis; A- cranial extremity, B- caudal extremity, C- Head of epididymis, D. Body of epididymis, E.Tail of epididymis, F. vas deference



Fig. (3): Microscopic section of the testis: A. tunica vaginalis,B.tunica albuginea , C.tunica vasculosa,D.Interstitial tissue, E.Septa ,F. Blood vessels , G. seminiferous tubules. H&E stain. (X40)



Fig. (4). Microscopic section of the left testis in camel shows the : A.seminiferous tubules , B. basement membrane, C. Lumen of seminiferous tubule, D,Septa ,E. Interstitial tissue . H&E stain. (X40)



Fig. (5): Microscopic section of the Right testis in camel shows the : A. seminiferous tubules, B.basement membaren , C. Interstitial tissue ,D ,Septa , E. Lumen of Seminiferous Tubule Masson stain (X100)

Fig. (6): Microscopic section of the Right testis; A. capsule of testis, B Interstitial Tissue., C. basement membrane D.blood vessels, F. seminiferous tubules, G.Septa, Van Gieson stain (X40)



Fig. (7). section of the testis in camel; A. basement membrane, B, Myoid cell, C, Sertoli cell, D. Secondary sppermatocytes E, First Stage van Gieson stain(X400)





Fig. (8): section of left testis; A. basement membaren, B. Pachytene phase, C. Secondary spermatocytes, D. Four stage, E.Zygotene phase, F. Second stage, G.Leptotene phase, H. Eight stage. I. First stage, Verhoeff,s stain (X400)



Fig.(9): Microscopic section of the right testis in camel shows the: A.Preteptotene B. Third Stage , C. Sertoli cell , D. leydig cell, E.Secondary spermatocytes, F. Second Stage , G.Thirteen stage, H.Six stage, H&E (X400) Fig.(10): Microscopic section of the left testis in camel shows the : A. basement membaren, B. leydig cell , C. Diplotene, D.Nine Stage , E.Eight Stage , Verhoef's stain (X100)



Fig.(11): Microscopic section of the head right epididymis in camel shows the: A. Connective tissue, B. Muscular layer, C.basal cell , D. Blood vessels, E.pseudostratified columnar ep. F,cilia , hematoxylin and eosin stain (X100).



Fig.(12): Microscopic section of the tail left epididymis in camel shows the: A.Elastic fiber, B.Sperm, C.Basal cell, D. Muscular layer, E.lumen, F, Dense connective tissue, G. Blood vessel verhoffs stain (X200)

CONCLUSION:

Although some characteristics unique to camels were noted, spermatogenesis in camels was largely comparable to that of the majority of mammalian species. The camel's testis is situated in the scrotal perineal area. There are six distinct stages that primordial spermatocytes go through. Preleptotene, pachytene, leptotene, zygotene, , diplotene, and diakinesis phases are some of these stages. The spermatids underwent fourteen steps of differentiation before becoming mature sperm. We might draw the conclusion that the investigation demonstrated the notable distinctions

between the histology of the left and right testes. The amount of spermatozoa may have contributed to the left testicle's higher activity in this investigation.

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Availability of data and materials

Data for the current study is available upon an adequate request.

Competing interest

None of the writers have any conflicts of interest.

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Ethical consideration

Study conduct at AL Muthanna University in Iraq had been conducted in accordance with ethical standards.

Author contributions

Every author makes an equal contribution.

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