

## Research Article

### Morphological Study of the Testis of Adult Sudanese Duck (*Anas platyrhynchos*)

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**Abstract:** The aim of the work to show the morphology of the testis of Adult Sudanese duck (*Anas platyrhynchos*). The adult duck testes were two bean-shaped, large and soft, the left testis was usually higher in position and larger in size than the right one. The testis was active during cold weather with the mean diameter of the seminiferous tubules being 124  $\mu\text{m}$  in the duck. It was less active during the hot season with the mean diameter of the seminiferous tubules being 134  $\mu\text{m}$  in the duck. The non-breeding season seemed to be characterized by a decline in the spermatogenic activity only and not by complete spermatogenesis. The mean volume of left testis in the adult duck was 9.12  $\text{cm}^3$ . They exceeded those of the right testes by 8%; the relative volume of the components remained essentially the same in both testes. The seminiferous tubules formed 90% of the volume of the testes in duck.

**Keywords:** *Anas platyrhynchos*, duck, morphological, sudanese, testis

#### INTRODUCTION

The morphology of the reproductive tract of many avian species, including the domestic fowl, has been extensively studied (Aire, 1982; Patricia *et al.*, 2007). Considerable attention was given to the structure of the epidermal region and the excurrent duct system of the testes (Hess and Thurston, 1977; Obidi *et al.*, 2008).

Search in the literature has shown no morphological data on testes of any of the avian species in Sudan.

The aim of the work to show the morphology of the testis of Adult Sudanese duck (*Anas platyrhynchos*).

#### MATERIALS AND METHODS

In Sudan (Khartoum state), the material was collected during different seasons (Winter and Summer) from forty mature male Hisex white and brown chickens. The birds were killed by dislocation of the neck vertebrae and the abdomen was immediately opened. The location of testes was examined prior to their removal from the abdominal cavity.

Tissue samples for histological study were obtained from the testis fixed in different fixatives. Bouin's fixative was found to be more satisfactory than others. Tissues were routinely treated and embedded in paraffin wax and sectioned at 5-7  $\mu\text{m}$  in a rotary microtome.

The sections were stained with haematoxylin and eosin (H&E) for general histology, Masson's trichrome

for collagenous fibres, Verhoff's and Gomori's aldehyde fuchsin method for elastic fibres, Gordon and Sweets silver impregnation method for reticulin fibres, and Gless and Marsland modification of Bielschowsky's method for neurons (Culling, 1974).

For the estimation of seminiferous tubules diameter, the measurement were carried out using an Olympus Tokyo microscope fitted with a micrometer  $\times$  6 eye piece at  $\times$ 10 magnification.

#### RESULTS

**Gross features:** The testes lie ventral to the anterior lobes of the kidney. They were bean-shaped, white and richly vasculatured. The left testis was usually higher in position and large in size than the right one (Fig. 1). The mean weights of the left and right testes were 9.60 and 8.88 gm, respectively.

**Histological observations:** A connective tissue capsule consisting mainly of collagenous fibres and reticular fibres enclosed the testis (Fig. 2 and 3). No mediastinum testis was observed as the rete testis was found outside the testicular parenchyma (Fig. 4). The latter was constituted of convoluted seminiferous tubules and interstitial tissue. A thin fibrous basement membrane, collagenous fibres and reticular fibres surrounded and supported the tubules (Fig. 5 and 6).

The large cells lying next to the basement membrane were the spermatogonia (Fig. 7). Both types of spermatogonia A and B were observed (Fig.8).

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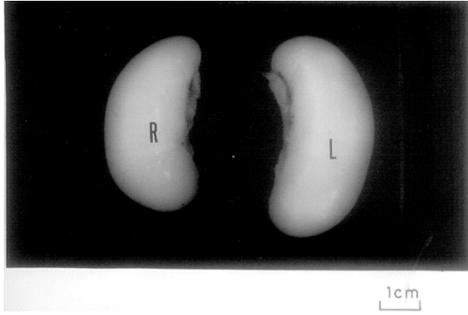


Fig. 1: Photograph of the testis of the duck. R, right testis. L, left testis

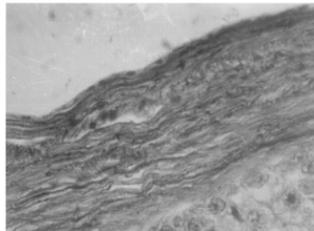


Fig. 2: Photomicrograph of the testis showing the collagenous fibre in the capsule of duck testis. Masson's trichrome. X 500

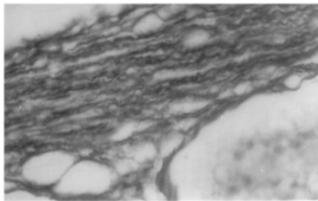


Fig. 3: Photomicrograph of the testis showing the reticular fibre in the capsule of duck testis. Gordin and Sweets. X 500

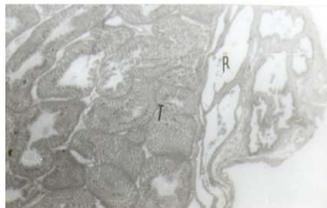


Fig. 4: Low power photomicrograph of duck testis showing no mediastinum testis, the rete testis (R) is found outside the testis (T). H. & E. X 40

Spermatogonia type B were more numerous in number than the spermatogonia type A.

The largest of the spermatogenic cells were the primary spermatocytes which lie adjacent to the spermatogonia (Fig. 7). More than one generation was found. Their nuclei were darkly stained with the chromatin showing a net-like appearance.

The secondary spermatocyte were occasionally seen in some tubules. They were smaller than the

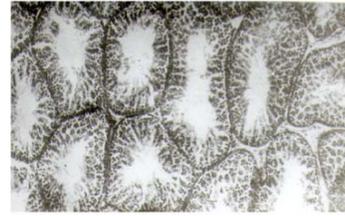


Fig. 5: Photomicrograph of the testis showing the PAS-reactive substance resistant to diastase digestion in the boundary tissue of seminiferous tubules of duck. Counterstain with haematoxylin. X 100



Fig. 6: Photomicrograph of duck testicular parenchyma showing the framework of reticular connective tissue. Gordin and Sweets. X 100



Fig. 7: Photomicrograph of the duck testis showing spermatogonium (A), primary spermatocyte (P), secondary spermatocyte (C), spermatid (M), spermatozoa (S), sertoli cell (O) and the interstitial tissue with abundant leydig cells (E). H. & E. X 500

primary spermatocytes and possessed darkly stained nuclei (Fig. 7).

The secondary spermatocyte were occasionally seen in some tubules. They were smaller than the primary spermatocytes and possessed darkly stained nuclei (Fig. 7).

Close to the lumen of the seminiferous tubules, the spermatids were round in shape and arranged in small groups (Fig. 7). They become closely associated with the sustentacular cells of sertoli.

Spermatozoa possessed small deeply staining head; their tails extend freely into the lumina of the seminiferous tubules (Fig. 7). The sperm content of the latter was variable and some of the tubules especially in the summer samples were completely empty (Fig. 9).

The sustentacular cells of sertoli were slender, elongate with irregular outline. They extended from the basement membrane to the lumen of the seminiferous tubules.

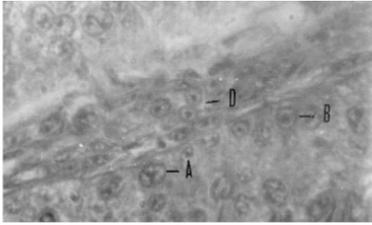


Fig. 8: Photomicrograph of the duck testis showing the type A and type B spermatogonia and Leyding cells (D) in the interstitial tissue of duck. H. & E. X 1000

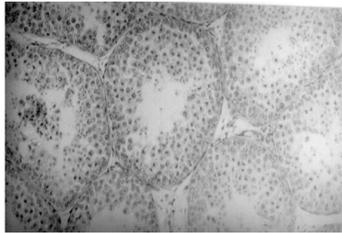


Fig. 9: Photomicrograph of the duck testis collected in the summer. Some of seminiferous tubules contained no spermatozoa. H. & E. X 200

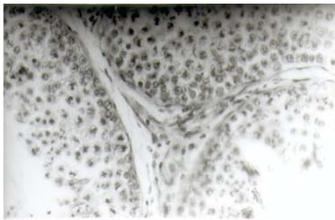


Fig. 10: Photomicrograph of the interstitial tissue of duck with scattered Leyding cells as seen in a summer sample. H. & E. X 400



Fig. 11: Photomicrograph of normal structure of the duck seminiferous tubules showing numerous spermatozoa (winter sample). H&E. X100

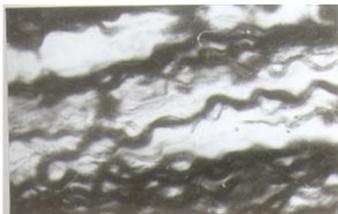


Fig. 12: Photomicrograph of nerve fibres in the capsule of duck testis. Glees. X1000



Fig. 13: Photomicrograph of nerve fibres in the interstitial tissue of duck testis. Glees. X1000

Their nucleus was large, light staining and irregular in shape with distinct nucleolus (Fig. 7). The nuclear shape varies from round, oval, pear-shaped to elongate.

Leyding cells, fibroblast, collagenous fibres and reticular fibres, lymphatic vessels and blood capillaries constituted the testicular interstitial tissue. Some of the Leyding cells appeared in small or large groups (Fig. 7 and 8). The Leyding cells appeared scattered in the summer season (Fig. 10). They were polygonal in shape with a large spherical nucleus and a distinct nucleolus (Fig. 7, 8 and 10).

**Seasonal variations:** During the Winter season (November to March), the specimen collected showed normal histological pictures with a mean diameter of the seminiferous tubules of about 124 $\mu$ m (Fig. 11).

During the hot season (May to October), the specimen collected showed less number of primary spermatocytes and spermatids and absence of spermatozoa in the seminiferous tubules. The mean diameter of the seminiferous tubules during the hot season was about 134 $\mu$ m (Fig. 9).

**Intrinsic innervation:** Large nerve bundle were seen in the capsule (Fig. 12 and 13). Fine nerve fibres were seen also amongst the Leyding cells and in association with the blood vessels of the interstitial tissue

## DISCUSSION

As described by El Jack (1970), the testes of adult duck are two large bean-shaped, white and soft. In accord with King (1975), the left testis is usually higher in position and large in size than the right one. Also, the testes are situated intra-abdominal and not within the scrotal compartment. In all large species so far studied, the testes are situated extra-abdominally and within the scrotum, with the exception of the African elephant in which they are intra-abdominal (Hay and Deane, 1966). In hippopotamus, the testis is sub pubic, but is not contained in a scrotum is usually higher in position and larger in size than the right one (El Jack, 1970).

In the adult chicken and duck, the mean diameter of the seminiferous tubules in the winter season about 126 and 124  $\mu$ m and in the hot season they were 135 and 134 $\mu$ m, respectively. In the winter season, the

testes were more active than in the hot season. In the camel however, the mean diameter of the seminiferous tubules during the cold and hot season were about 162.6 and 176.4  $\mu\text{m}$ , respectively (Tingari *et al.*, 1984). In fact, the camel has been described as being more active sexually during the winter season than the hot season (Osman, 1975). This may hold true of chicken and the activity seems to be inversely proportional to diameter of seminiferous tubules.

The tunica albuginea of the chicken like that of the camel (Osman, 1975), consists of elastic and collagenous fibres and no smooth muscle fibres were present. In that respect, it differs from that of the horse where the tunica albuginea, septa and trabeculae are formed of collagenous and smooth muscle fibres (Julian and Tyler, 1959). As described by Tingari (1971), there is no distinct mediastium testis; the rete network lies out side the testis.

The testicular interstitial tissue has been described as being loose in structure and consist of connective tissue elements and isolated polygonal leyding cells (Bloom, 1962).

Hay and Deane (1966) reported that the amount of interstitial tissue in the ram testis was scanty and contain rather few cells, most of which appeared undifferentiated, while the leyding cells could not be distinguished in routine preparations. The clustered interstitial cells were very large and abundant in the boar (Hay and Deane, 1966). The interstitial tissue of camel is constituted of leyding cells, some fibroblasts, collagenous fibres, lymphatic vessels and blood capillaries (Tingari *et al.*, 1984). The leyding cells are large and their shape is polyhedral in dogs (Creed, 1960), but round to oval or elongate in elephants (Johnson and Buss, 1967). Trautmann and Fiebier (1957) stated that the leyding cells of domestic animal were oval in shape with large spherical nuclei and distinct nucleoli. leyding cells, either singly or in the form of groups are found in the spaces between the seminiferous tubules (Julian and Tyler, 1959). In the one humped camel leyding cells have been described as polygonal in shape and occur either singly or in small group (Tingari *et al.*, 1984). During November and December, i.e., Winter there was a remarkable increase in the amount of these cells. In the present study, the leyding cells appeared in the form of small or large groups during Winter, but in the summer season, they appeared somewhat scattered. they were generally polygonal in shape and had a large spherical nucleus with a distinct nucleolus.

In dog, the seminiferous tubules are surrounded by collagenous fibres, elastic fibres and a thin basement membrane (Creed, 1960). They presence of contractile cells in the boundary tissue of rat seminiferous tubules was shown by Korman and Niemi (1966) and Dym and Fawcett (1970). In domestic fowl the seminiferous tubules were bounded by collagenous fibres and cellular components; the cellular elements were inner fibroblast and outer myoid cells (Rothwell and Tingari, 1973). The boundary tissue of the seminiferous tubules

in the adult chicken and duck examined in this study and as revealed by light microscope, consists of collagenous and reticular fibres. This is consistent with the finding of Rothwell and Tingari (1973).

The intratubular cells of all animal studied so far have many features in common. Sertoli cells, for example, have indistinct and irregular boundaries and a vesicular nucleus with a distinct nucleolus (Trautmann and Fiebier, 1957). In bulls, different types of spermatogonia are seen, but the main ones are types A and B. Type A is large with a round or oval nucleus and one nucleolus. Type B is smaller and has a coarse chromatin in the spherical nucleus (Ortavant, 1959). Primary spermatocytes have large darkly staining nuclei, the chromatin of which shows a net-like appearance. secondary spermatocytes are smaller with a light nucleus and no nucleolus (Trautmann and Fiebier, 1957). In the elephant, secondary spermatocytes have nuclei resembling these of type B spermatogonia (Johnson and Buss, 1967). The spermatids are small cells with deeply staining nuclei in the dog (Creed, 1960), but in the elephant the nuclei of spermatids do not stain darkly except during elongation (Johnson and Buss, 1967). In the camel the structure of the intratubular elements fits well with what was reported in other animals (Osman, 1975). In this study, the spermatogonia were of two types, A and B. The primary spermatocytes were the largest of the spermatogenic cells, whereas the secondary spermatocytes were small. The spermatids were round and small lying usually in group close to the lumen of the seminiferous tubules.

The adult chicken spermatozoa are similar to other avian spermatozoa (Tingari, 1973). They were almost bilaterally symmetrical consisting of the usual segments, head, neck and tail.

Nerve fibres have been observed in the interstitial tissue of both fowl and duck testes. In the human testis, most of the nerve fibres have been reported in company of blood vessels which they also supplied (Risley and Skrepetos, 1964). Innervated vessels are found to penetrate more deeply into the interior of the testis in cat (Norberg *et al.*, 1967), man and the rhesus monkey (Baumgarten and Holstein, 1967). There is a general agreement that nerves do not penetrate the boundary tissue to enter the cellular portion of the seminiferous tubules (Shioda and Nishida, 1966; Risely and Skrepetos, 1964). However, a few fibres have been observed to pass near the contractile cells of the seminiferous tubules in bull (Shioda and Nishida, 1966) and man (Baumgar and Holstein, 1967). In cat, rats and guinea pigs, cell groups of leyding are associated only with vascular fibres (Norberg *et al.*, 1967), while in man and rhesus monkey, terminal fibres from arterial plexuses have been observed at times to come into close relationship with individual leyding cells (Baumgarten and Holstein, 1967).

In the human testis nerve fibres were found to penetrate the groups of leyding cells and then break up to encircle the interstitial cells completely (Gray, 1947).

Tingari and Lake (1972) reported that single or aggregated nerve fibres were located in the interstitial tissue of domestic fowl, some branches, appeared to be associated with the seminiferous tubules, while others were associated with blood vessels of the interstitium. The intrinsic nerves in this study were found in the interstitial tissue either free or in associated with blood vessels and seminiferous tubules, an observation which is essentially similar to that of Tingari and Lake (1972). The close association of these nerve fibres to contractile cells of cells of the boundary tissue and leyding cells may indicate a functional significance relating to neural stimulation of contractility of myoid cells or/and secretory activity of leyding cells.

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