MORPHOMETRIC STUDY OF THE TESTES OF THE NIGERIAN LOCAL BREED OF CHICKEN

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ABSTRACT

Data from 10 adult male Nigerian local breed of chicken were used to investigate the morphometry of the testis. The mean live weight of the birds as well as testes weights were obtained, and testes samples subjected to histological preparations for light microscopy. The mean live weight of the birds and testes weights were 977g and 6.3g respectively, with a gonadosomatic index of 1.1%. The dimensions of the testes were 5.30cm and 2.70cm for the length and width respectively. The volume densities were 96.6% and 3.4% for the seminiferous tubules and intertubular compartments respectively, out of which Leydig cells occupied 1.4%. The seminiferous tubular diameter was 158.40 µm while tubular lengths per testis and per gram of testis were 308 and 49 meters respectively. The number of Leydig cells per testis and per gram of testis were 2.1 billion and 340 million respectively while the number of sertoli cells per testis and per gram of testis were 2 billion and 300 million respectively. These data obtained, tend to suggest that the Nigerian local breed of chicken has high spermatogenic efficiency.

Keywords: Morphometry, Testis, Nigerian local chicken breed, Seminiferous tubular diameter, Gonadosomatic index

INTRODUCTION

The domestic chicken is an important source of protein in Nigeria. This breed of birds is reared by extensive or semi -intensive methods, thus reducing cost of production. They have also been reported to be more tolerant to many of the endemic poultry diseases, when compared with the exotic breed. At present, there is paucity of information on the general anatomy and physiology of this breed of chicken.

It has been established that morphometric study of the testis of any breed or species is necessary in assessing and estimating quantitative changes in testicular components and spermatogenic functions arising from such factors as age (Johnson and Neaves, 1981; Wang *et al*., 1993), season (Hochereau-de Reviers and Lincoln, 1978), hormone (Varadaraj *et al*., 2001) and drugs (Desouky *et al*., 1991). Quantitative methods have been utilized in assessing testicular structure and function under various physiological and pathological conditions (Clermont and Perey, 1957; Omeke and Igboeli, 2000). Data generated from morphometric studies have been positively correlated with functions of the testis (Mendis-Handagama *et al*., 1988; Hikim *et al*., 1989; Omeke and Igboeli, 2000).

Many studies have been carried out on testes morphometry of different animals like the boar (Lunstra *et al*., 2003), goats (Leal *et al*., 2004), cats (Franca and Godinho, 2003), donkey

and mules (Neves *et al*., 2002) etc. However few data are available on the testis morphometry of birds in general and domestic chicken in particular (Halldin *et al*., 1998; Onu and Ndodo, 2003). The present study may improve on the available information on the testis morphometry of birds in general and Nigerian local breed of chicken in particular.

The study reported herein was carried out to assess the morphormetry of the testis of Nigerian local chicken.

MATERIALS AND METHODS

Ten apparently healthy adult male Nigerian local breed of chicken were purchased from Nsukka market in South Eastern Nigeria. They were first quarantined for two weeks and thereafter stocked in a pen at the veterinary demonstration farm, University of Nigeria, Nuskka. The birds were maintained on a growers mash (vita feed^{R}) and provided with water *ad-libitum.* The birds were weighed and euthanized with an overdose of sodium pentobarbitone (Euthanase ^{R}). The testes were dissected out and weighed on a metler balance. The length and width of the testes were measured, using a vernier caliper.

Tissue preparation for light microscopy: The testes of each bird were cut into thin slices, fixed in Bouin's fluid for 24hours, and then post fixed in 70% ethanol, dehydrated, cleared in xylene, embedded in paraffin wax, sectioned at 5µm, thickness, and stained with heamatoxylin and eosin

Histomorphometric Evaluations

Volume densities: The volume densities of the various components of the testis were determined using a 36 point reticule (grid). 30 fields chosen randomly were scored for each animal at 400x magnification (Okwun *et al.*, 1996).

Volume of testis: The volume of each component of the testis was determined as the product of volume density and testis volume. The volume of testis was determined by water displacement.

Seminiferous Tubule Measurements

Tubular diameter and height of seminiferous epithelium: These were measured at X100 magnification using a digital camera (Moticam 2000, Motic China Group) attached to the ocular of the light microscope and connected to a computer. 30 tubular profiles that were round or nearly round were chosen randomly and measured for each animal (Segatell *et al*., 2004).

Tubular lengths per testis and per gram of testis were calculated using the formula of Varadaraj *et al.* (2001) thus: Tubular length per testis = Volume of seminiferous tubule /πr^2, where r = radius. Tubular length per gram of testis = Tubular length per testis / Testis weight

Sertoli and Leydig cells count: The number of Sertoli cells and Leydig cells per testis and per gram of testis were determined using the formula of Castro *et al.* (2002) thus: Number of cells per testis = Total nuclear volume / Volume of a single nucleus. Number of cells per gram of testis = Number of cells per testis / Testis weight. All data were expressed as means $\pm$ standard error of mean.

RESULTS

The mean body weight of the birds was 977g with a mean testis weight of 6.30g.The dimensions of the testes were 5.30 cm and 2.70 cm for the length and width respectively (Table I). The mean volume densities of the components of the testis are tabulated in Table 2. The tunica propria, seminiferous epithelium, and lumen, occupied 14.6 % 69.9 % and 12.1 % of the seminiferous tubules respectively. Within the inter tubular compartment, the Leydig cells occupied a greater portion (1.4 %) while the connective tissues, blood vessels and lymphatic vessels occupied 0.8 %, 0.2 %, and 1.0 % respectively (Table 3). The mean tubular diameter and height of the seminiferous epithelium were 158.40µm and 56.00µm respectively.

Table 1: Body weight and testes dimensions of Nigerian local breed of chicken

Morphological Parameters	Dimensions
Body weight (g)	977 ± 44
Testis Weight (g)	6.3 ± 0.7
Gonadosomatic index (%)	1.1 ± 0.2
Length of testis (cm)	5.3 ± 0.1
Width of testis (cm)	2.7 ± 0.1

TABLE 2: Volume densities of testes components of Nigerian local breed of chicken

testes components	Dimensions (%)
Tunica Propria	14.6 ± 0.9
Seminiferous epithelium	69.9 ± 0.6
Lumen	12.1 ± 0.8
Leydig cells	1.4 ± 0.4
Connective tissues	0.8 ± 0.2
Blood vessels	0.2 ± 0.1
Lymphatics	1.0 ± 0.1

Table 3: Dimensions of the seminiferous tubules from Nigerian local breed of chicken

Seminiferous Tubules	Dimensions
Tubular diameter (μm)	158.4 ± 7.0
Height of seminiferous epithelium (μm)	54.0 ± 0.9
Tubular length per testis (meters)	308.0 ± 15.4
Tubular length per gram of testis (meters)	49.0 ± 0.2

The total length of the seminiferous tubule was 308 meters while the length per gram of testis was 49 meters (Table 4). The mean nuclear volume density of Leydig cells was 0.6% with a total nuclear volume of 3.80μm^3. The nuclear diameter was 3.20μm while the nuclear volume was 17.30μm$^{3.}$ There were 2.1billion Leydig cells per testis with 340 million per gram of testis (Table 4). The mean nuclear volume density of sertoli cells were 5.8% with a total nuclear volume of 37.10μ m$^{3.}$ The nuclear diameter was 7.30μm, while the nuclear volume was 205.90μm$^{3.}$ There were 2 billion sertoli cells per testis and 300 million sertoli cells per gram of testis (Table 5).

DISCUSSION

The gonadosomatic index of 1.1% recorded in this study is high when compared to what had been reported in mammals. Kenagy and Trombulack (1986), reported 0.1% for bulls, while 0.4% was reported for goats (Lunstra *et al*., 2004), 0.08% for cats (Franca and Godinho, 2003), 0.22% for gerbil (Segatell *et al*., 2004), and 0.04% for buffalos (Franca and Russell, 1998). The high gonadosomatic index suggests high sperm production efficiency. This suggestion was based on earlier reports that there existed positive correlation between testis weight and sperm production (Cameron *et al*., 1948b, Leal *et al*., 2004). The dimensions of 5.30cm and 2.70cm for length and width of the testis respectively were within the range reported by earlier studies in birds (King, 1975; Bezuidenbout *et al*., 1999).

Seminiferous tubular compartment occupied 96.6% of the testis. This proportion is very high compared to what has been reported in mammals where seminiferous tubules occupy 70-90% of the testis (Russell *et al.,* 1990; Franca and Russell, 1998). It has also been demonstrated that there is a positive correlation between volume density of seminiferous tubules and spermatogenic efficiency (Franca and Russell, 1998; Johnson *et al*., 2000). This then further suggests that the Nigeria breed of chicken may have a high reproductive efficiency. The connective tissue compartment was very sparse (0.8%) as noted in the study. This was reminiscent of observation made in the testis of other breed of birds (Aire, 1997). The poor connective tissue content may be attributed to the compact seminiferous tubules which created very little interstitial spaces. The volume densities of 0.2% and 1.0% for blood vessels and lymphatic vessels respectively observed in the study indicate that there are more lymphatic than blood vascularization. This requires clarification and comparative study with other breeds of birds may be very necessary. The tubular diameter of 158.40μm was lower

Table 4: Dimensions of the Leydig cell from Nigerian local breed of chicken

Leydig cell	Dimensions
Nuclear volume density (%)	0.6 ± 0.20
Total nuclear volume (μm^3)	3.8 ± 0.90
Nuclear diameter (μm)	3.2 ± 0.10
Nuclear volume (μm^3)	17.3 ± 1.90
Leydig cells/ testis ($x10^9$)	2.1 ± 0.30
Leydig cells/ g testis ($x10^6$)	340 ± 0.10

Table 5: Dimensions of the sertoli cell from Nigerian local breed of chicken

Sertoli cell	Dimensions
Nuclear volume density (%)	5.8 ± 0.40
Total nuclear volume (μm^3)	37.1 ± 1.80
Nuclear diameter (μm)	37.3 ± 0.50
Nuclear volume (μm^3)	205.9 ± 37.9
Sertoli cells/testis ($x10^9$)	2.0 ± 0.30
Sertoli cells/ g testis ($x10^6$)	300 ± 0.10

than the range (180 – 350µm) reported for most mammals (Setchell *et al*., 1994). However Onu and Ndodo (2003) reported a tubular diameter of 15.60µm for domestic fowls. The striking difference could be as a result of breed, or level of maturity of birds used by these authors.

Generally, 10 to 15 meters of seminiferous tubules were found per gram of testis in mammals (Franca and Russell, 1998; Setchell *et al*., 1994). However 49 meters per gram of testis were observed in this breed. This may have contributed to the high volume density of the seminiferous tubules as well as reduced interstitial space reported for the breed. The lengthy seminiferous tubules may be an adaptation phenomenon

The number of Sertoli cells per gram of testis observed in the study (300 million) was very high compared to mammals already investigated (boar 25million, stallion 28 million, cat 32 million, bull 29 million, rabbit 25 million). The high number of Sertoli cells suggests an improved rate of sperm production. This suggestion is based on earlier reports that each Sertoli cell supports a limited number of germ cells in a species specific manner (Russell and Peterson, 1984; Franca and Russell, 1998). It has also been established that there is a positive correlation between the number of Sertoli cells per gram of testis and spermatogenic efficiency (Johnson, 1991; Johnson *et al*., 2000; Franca *et al*., 2002). Thus sertoli cell establishes the upper limit for sperm production (Steinberger and Steinberger, 1971).

The number of Leydig cells per gram of testis was observed to be higher than those of mammals as well as some exotic breeds of birds (Onuoha and Uchenabo, 1990). The increased number may imply increased secretion of testosterone required for the development of secondary sexual characteristics such as virilism. This is very vital as these local birds are managed under extensive husbandry system requiring the pursuit of hens over wide unenclosed area. This is unlike their exotic counterparts managed intensively.

This study has generated data on testis morphometry of Nigerian local breed of chicken. It is hoped that the data will be valuable in quantitative assessment of testicular structure and function under various physiological as well as pathological conditions.

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