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Sperm Reserves and its Relationship to Parameters of the Testis, Epididymis and Vas Deferens of Local Cocks in the Sahel Region of Nigeria

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ABSTRACT

The morphometry and sperm reserves of the testis, epididymis and vas deferens of 19 sexually active adult local cocks were studied. The average live weight of the cocks was 1.879±0.316 kg and that of the paired testis, epididymis, vas deferens were 12.17±4.75 g, 0.52±0.19 g and 1.36±0.56 g, respectively. The length of the testis, epididymis and vas deferens were 3.42±0.50 cm, 1.91±0.38 cm and 11.5±1.34 cm, respectively. In majority of cases, the parameters of the left organs were insignificantly greater than those of the right. The width, thickness, circumference and volume of the testis as well as the diameter and volume of the epididymis and vas deferens were recorded. There were strong positive correlation between the weight of the testis and epididymis (r=±0.82) or vas deferens (r=±0.66). The gonadal sperm reserve was 2.11±1.11 x 10^9 sperm (173.7±71.5 x 10^6 sperm g^{-1}) and the extrangonadal sperm, 1.77x 10^9 sperm. The vas deferens with 89.8% of the extragonadal sperm reserved as the main storage organ of sperm for use during threading. Sperm reserve was positively correlated to body weight (r=±0.77, P = 0.000) and to the length of the testis (r=±0.55, P = 0.015). This may suggest that body weight and testicular length are indicators of fertility. The high gonadal sperm and hence reproductive potential may reflect an adaptive change in the local cocks, which have a very high mating frequency per day.

Key Words: Morphometry; Gonads; Sperm; Cocks

INTRODUCTION

The reproductive organs of male chicken are intra-abdominal and located on either side of the mid-line attached to the roof of the coelom. The organs consist of a pair of testis, epididymis and vas deferens which terminates in the local copulatory apparatus (Lake, 1957). In working with adult leghorn cocks, Gray (1937) reported that the right testis was 10% larger than the left meanwhile Latimer (1924) had reported that the left testis of leghorn cockerels below six months of age was 17% larger than the right.

In a sexually active cock, the testis made up 1% of body weight (Burke, 1977). Heavier breeds of cocks have larger testis than lighter breeds (Parker *et al.,* 1942; Kumaran & Turner, 1949). The local varieties of cocks in the Sahel have been described (Ibrahim & Abdu, 1992). The local cocks have small body size and light body weight. The morphometry and sperm reserve of the cocks have not been reported.

The main function of the testis is sperm production via spermatogenesis, the epididymis transport and that of the vas deferens, storage of sperm for maturation and use during threading. In mammals, scrotal circumference has been widely used as an index of fertility. In chicken, there is no such index. This is probably due to the intra abdominal location of cock's testis. With the availability of technologies, which can accurately determine the sizes of internal structures such as ultra-sonography, it is now possible to measure the sizes of the testis of cocks. These studies provide basic data on the morphology and sperm reserve of the reproductive organs of the local cocks in the Sahel region and define the possible relationships between various organs and sperm reserve.

MATERIALS AND METHODS

Nineteen mature cocks purchased from different farmers in Maiduguri were used for the study. The cocks were sacrificed by decapitation one at a time. The carcasses were soaked in water to wet the feathers and the abdomen opened up. The keel was completely removed to expose the thoraco-abdominal cavity. The testes, epididymis, vas deferens and cloacae were carefully dissected out intact and placed in a tray moistened with normal saline.

Morphometry of the reproductive organs. With the help of a thread and a measuring tape, the length of each epididymis and vas deferens together with the circumferences of each testis was measured. The diameter of each vas deferens and epididymis and the length, width and thickness of the testis were measured with a vernier calliper. The thickest and thinnest diameters of the Vas deferens were recorded, after which the various organs were partitioned following the anatomical description of Van Krey (1990).

The testis, epididymis and vas deferens were weighed separately on a sensitive electronic balance (Mettler Toledo B154, ±0.001g) and their volumes determined by the indirect method of water displacement using normal saline (0.85% Nacl). A 10 mL graduated conical centrifuge tube (±0.1 mL) was used to record volumes of the epididymis and the vas deferens while a 100 mL measuring cylinder (±1 mL) was used for the testis. The fluid used in determining the volume of any organ was later on used to homogenize that organ to minimize sperm loss.

Gonadal sperm reserve. The tunica albuginea was carefully peeled off from the testis before the volume was recorded and the testicular tissues sliced into small pieces with a surgical blade and macerated in a mortar with pestle. The crushed tissues were washed into a blander cup with 100 mL formal saline (1%) containing 1% eosin and blend for one minute. The diluent was added to achieve a final dilution of 1:20 and homogenized. The resultant homogenate was transferred into a beaker labeled left or right testis and stored at 4ºC for 24 h.

Extragonadal sperm reserve. Each epididymis and vas deferens was separately crushed in a mortar after slicing into pieces with a surgical blade with 10 mL of diluent. The supernatant and the sediment were rinsed into a blender cup with 1% formal saline so as to achieve a 1:200 dilution. All utensils, which had contact with the crushed organs were rinsed into the blender cup.

The sediments were homogenised for two minutes, before samples were collected for the count.

Counting procedure. The overnight homogenate of the gonads was shaken vigorously for one minute before a drop was introduced into a charged improved Neubauer Hemocytometer counting chamber. The saturated counting chamber was allowed to sediment for 15 min in a moist compartment. Counting was carried out under x 400 magnification of a light microscope only when the distribution of sperm in the chambers observed to be even. Sperm heads in five chambers (four corners and one central) of the E squares were counted. Sperm heads lying on the top and right borders of each counting chamber were included in the count (Hafez, 1987).

Statistical analysis. The total gonadal and extragonadal sperm count was determined by multiplying sperm concentration by the respective volumes of the organs. Sperm concentration is the product of the number of sperm cells counted, the multiplying factor and the dilution factors used.

The data collected were summarized as Mean±SD. Correlation between the variables were calculated and tested for significance at 5 and 1% levels. Student "t" test was used to compare the left and right organs and a one way analysis of variances of sperm reserve between and within organs was carried out.

RESULTS

The mean live body weight of the local cocks was 1.879±0.316 kg. The data on the morphometry and sperm reserve of the reproductive organs are summarized in Table I. The testes made up 0.64±0.22 (range, 0.27 to 1.04% of the live body weight). The left testis, epididymis and vas deferens were 2.0, 8.1 and 6.4% heavier, respectively than their respective right organs. These differences were however, not significant with the exception of the length of both the testes and vas deferens, and the diameters of both the epididymis and vas deferens, the dimensions of the left organs were generally larger than those of the right. There were very high positive correlations ($P<0.05$) between the left and right organs (Table I).

Table I. Mean values of the dimension and sperm reserves of the testis, epididymis and vas deferens and correlation coefficients (r) between the left and right parameters of the local cocks

Organs	Variables	Mean±SD	(r between left and right) r (significance)
Live cock	**Weight (g)**	**1879±316**	
Testis	Sperm reserve (x 10^9 sperm)	2.11±1.11	0.78(P=0.000)
	Paired weight (g)	12.17±4.75	0.82(P=0.000)
	Length (cm)	3.42±0.50	0.63(P=0.012)
	Width (cm)	1.95±0.29	0.56(P=0.013)
	Thickness (cm)	1.71±0.32	0.67(P=0.002)
	Long. Circumference (cm)	8.30±1.21	0.80(P=0.000)
	Equator circumference (cm)	5.46±0.69	0.74(P=0.000)
	Volume (mL)	11.74±4.53	0.88(P=0.000)
Epididymis	Sperm reserve (x 10^9 sperm)	0.18±0.17	0.63(P=0.003)
	Paired weight (g)	0.52±0.19	0.60(P=0.006)
	Paired volume (mL)	0.50±0.15	0.33(P=0.177)
	Length (cm)	1.91±0.38	0.57(P=0.014)
	Diameter (mm)	5.19±0.72	0.59(P=0.010)
Vas	Sperm reserve (x 10^9 sperm)	1.59±0.95	0.58(P=0.011)
deferens	Paired weight (g)	1.36±0.56	0.86(P=0.000)
	Paired volume (mL)	1.25±0.55	0.86(P=0.000)
	Length (cm)	11.59±1.34	0.67(P=0.003)
	Diameter (Thinnest, mm)	1.51±0.31	0.35(P=d.165)
	(bulbous part)mm	4.68±1.48	0.79(P=0.000)

The weights of testis and vas deferens (r=0.66, P=0.002) and their volumes (r=0.64, P=0.004) and the weights of the testis and epididymis (r=0.82, P=0.000) and their volumes (r=0.55, P=0.019) were significantly correlated in the same way as the weights of the Vas deferens and the epididymis (r=0.70, P=0.001). There were positive correlation between sperm reserve and the volume of the testis, epididymis and vas deferens, respectively (Table II).

The sperm reserve in the testis did neither correlate with those in the epididymis nor vas deferens (Table II) but those between the epididymis and the vas deferens correlated (r=0.52, P=0.025). This seems to demarcate the relationships that exist between the gonadal and extragonadal sperm reserve. There was more sperm stored in the testis (2.11 x 10^9 sperm) than in the extragonadal ducts (1.77 x 10^9 sperm). Of the extragonadal sperm, 89.9%

was reserved in the vas deferens which is the main storage of mature spermatozoa. Sperm reserve was positively correlated to the length of the testis, epididymis and vas deferens (Table II) but not to the weight of the testis (r=0.31, P=0.19). There were high positive correlation between body weight and extragonadal sperm reserve of the local cocks (r=0.77, P=0.000).

Table II. Sperm reserves and its correlation coefficients with some biometric parameters of the reproductive organs of the local cocks

	Sperm Reserve(SR)-testis	SR-Epididymis	SR-Vas diferens
Weight	0.31 (P=0.19)	0.65 (P=0.00)	0.80 (P=0.00)
Length	0.55 (P=0.02)	0.50 (P=0.04)	0.37 (P=0.13)
Volume	0.55 (P=0.02)	0.68 (P=0.00)	0.79 (P=0.13)
SR-Testis	0.00	0.02 (P=0.04)	0.43 (P=0.07)
S.R.-Epididymis	0.02 (P=0.14)	0.00	0.52 (P=0.03)
S.R.-Vas deferens	0.43 (P=0.07)	0.52 (P=0.03)	0.00

DISCUSSION

The local breeder cocks in the Sahel region were lighter than those of the Rhode Island Red and white reared in Nigeria (Nwagu *et al.,* 1996) and have smaller testis. Hocking and Bernard (1997) reported that the testis of well fed and healthy cocks were small and attributed it to high protein (17%) content of the diet. The testis made up 0.64% of the body weight. This proportion is less than the 1% reported by Burke (1977).

The mean testicular weight (12.17 g) of the local cocks, was similar to the values (9-13 g) for white Plymouth (Kumaran & Turner, 1949) but lower than the mean values for the light and heavy breeds (16.5-31 g) of white leghorn (Parker *et al.,* 1942). The reproductive organs were arranged symmetrically on either side of the mid-line attached to the roof of the coelom even through there was bilateral morphometric asymmetry of the organs. The weights and dimensions of the left reproductive organs were larger than those of the right. In the sexually active cocks, the length of the bean shaped testis was 2.66 – 4.55 cm long and compared favorably to the 3.25 – 5.60 cm reported by King (1975). The width (1.52 – 2.80 cm) and thickness (1.5 – 2.80 cm) also compare well with the values reported by King (1975).

The epididymis was closely attached along the length of the dorso-medial border of the testis. This was an elongated spindle-shaped enlargement that ran from the cranial to the caudal end of the dorsal surface of the testis. The epididymis was very short (1.1 – 2.6 cm) but coiled. Its diameter varied from 3.2 – 6.4 mm. Gray (1937) reported the thickness of epididymis to vary from 1 to 4 mm. Unlike in mammals, avian epididymis is not sub divided into distinct anatomical regions. Numerous anastomosis is not sub divided into distinct anatomical regions. Numerous anastomosing efferent ductules open into short connecting ductules that run along the length of the epididymis (Van Krey, 1990). Epididymas fluid then progressed into the vas deferens, which served as a storage organ for semen.

The vas deferens left the epididymis as a single duct (thinnest part) and became progressively convoluted towards the bulbous end. It straightened up just prior to entering the urodeum of the cloaca (Van Krey, 1990). At the end of the duct, it appeared bulbous more due to the presence of smooth muscles and connective tissue (Lake, 1957, 1981) than widening of tubule. The length of the undissected vas deferens was 9.4 – 14.70 cm and was similar to the 10.0 cm reported By Parker *et al.,* (1942). The vas deferens entered the copulatory apparatus as paired papillae (King, 1975; Van Krey, 1990).

Spermatozoa were concentrated to the bulbous posterior part of the vas deferens which was the main storage organ of sperm in the cocks. It was observed in this study that the right vas deferens which was wider at the bulbous end contained more spermatozoa than the left, even though the left organ was heavier and more voluminous than the right. This suggested that the distension of the bulbous vas deferens gave a better estimate of fertility than volume or weight of the whole organ.

There were strong correlations between the volumes of the various organs with their respective sperm reserve. With the exception of the vas deferens, the length of the testis and epididymis correlated significantly with the sperm reserve of the respective organs (Table II). These findings suggested that volume and length of the testis and epididymis may be used to estimate the fertility and hence the reproductive potential of the cocks.

More sperm cells were in the testis than in the excurrent ducts of the local cocks. The gonadal sperm reserve of the local cocks (2.11×10^9 sperm) was within the range reported ($1.7 – 2.28 \times 10^9$ sperm) by De Reviers (1975). Developed spermatides are released into the lumen of the seminiferous tubules and the residual bodies are phagocytised by the Sertoli cells (Jones & Lin, 1993). The released testicular spermatozoa were capable of fertilizing ova (Howarth, 1983; Brillard, 1993). This was only when they were inseminated directly into the oviducts or magnumof hens. As spermatozoa migrated through the excurrent ducts, they become structurally mature (Tingari, 1973a) and developed the capacity to move and ascend the female tract to achieve fertilization when inseminated intra-vagina (Howarth, 1983). The removal of sperm surface stays moieties impeded their transvaginal migration (Steele & Wishart, 1996). The sperm surface saitic acid protected it from immunological attack in the vagina. The sciatic acid was added onto spermatozoa during passage through the

collecting ducts. This process together with the thickening of the inner mitochondrial membrane of the spermatozoa while in the collecting ducts constituted the maturation processes of avian spermatozoa (Nicander & Hellstorm, 1967).

De Reviers (1972) reported values which was sufficient for 3.5 days supply of semen. It may be necessary for more work to be done to determine the daily sperm output of the local cocks and evaluate the contribution of testicular sperm reserve to sperm output. The sperm per gram of testicular tissue, 173.7±71.5 x 10^6 sperm/g of the local cocks was higher than values 80 – 120 x 10^6 sperm/g reported by De Reviers and Williams (1984).

CONCLUSIONS

The local cocks in the Sahel region have high reproductive potential. The gonadal sperm reserve was higher than the extragonadal sperm reserve. Body weight, length of testis and wide of Vas deferens were positive indicators of fertility of cocks. The high gonadal sperm may be an adaptive change in the local cocks, which usually have very high mating frequency. There is significant difference in the sperm reserve of the testis, epididymis and vas deferens.

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