Spermogram and Morphological Characteristics in Testicular and Epididymal Spermatozoa of Large White Boar in Nigeria

Espermiograma y Características Morfológicas de los Espermatozoides en el Testículo y Epidídimo del Verraco Grande Blanco en Nigeria

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OYEYEMI, M. O. & UBIOGORO, O. Spermogram and morphological characteristics in testicular and epididymal spermatozoa of large white boar in Nigeria. *Int. J. Morphol.*, 23(3):235-239, 2005.

SUMMARY: Ten healthy male (boar) of large white pigs aged between 6.0 and 18.0 months, were used for this study. Spermogram and morphological characteristics were studied in testes and in the different parts of the epididymis of the boar. The objective was to study the changes that occur in testes and epididymis during epididymal transit. Progressive motile spermatozoa were first observed in the testes (6-10%) in the caput (20-30%), corpus (40-50%) and highest at the caudal epididymis (85%). Cytoplasmic droplets were lost as the spermatozoa moved up the epididymis. The caput epididymis had a value of 20.05% of abnormal sperm cell, which is different significantly ($p < 0.5$) from 15 – 80 % of the caudal epididymis. The total abnormal sperm cells in corpus are also higher than in the caudal epididymis. Percentage motility was 0.6 ± 2.0 % in the testis, which was lower ($P < 0.5$) than in caput with 23.0 ± 5.0 %. This motility increases from caput to caudal epididymis. The caput epididymis also had 0.20% of spermatozoa with small head, which differed significantly ($p < 0.05$) from corpus (0.05%) and the caudal (0.00%).

The testicular and epididymal parameters were positively correlated (weight of testis and epididymis; weight of testis, scrotal circumference, testicular circumference, weight of epididymis and weight of testis ($p < 0.05$), except the correlation values between testes and epididymis circumference (TEC) and other parameters ($p < 0.01$)

KEY WORDS: Spermogram; Boar; Spermatozoa; Motility.

INTRODUCTION

Reproductive ability in the male comprises the production of semen containing normal spermatozoa (quality) in the adequate number (quantity), together with the desire and ability to mate. The epididymis is an extremely large convoluted structure, which is closely attached to the dorsal part of the lateral surface of the testicle (Setchell, 1977; Oyeyemi *et al.*, 2000). Its functions include storage, maturation and absorption of sperm cells. Changes occurring in sperm during epididymal migration, have been correlated with the function, integrity of the testis and the epididymis (Rao, 1971). This has led to the classification of sperm defects into primary and secondary or major and minor sperm defects (Bloom, 1973) although Moss *et al.*, (1988) classified these defects into primary, secondary and tertiary or miscellaneous.

Coordinated sexual behaviour and first ejaculation in the boar usually occur at 5 - 8 months of age that boars attain puberty. Fertility gradually increase and get to the peak

when the boar reaches about 18 months of age (Leman & Rodefer, 1976; Hurtgen, 1986) and a peak level of sperm production is maintained until a gradual decline sets in at about 5 years of age (Hurtgen). Spermatozoa are carried in a large amount of secreted fluid from the seminiferous epithelium and rete testis into the tortuous efferent ducts or ductuli efferentes, usually, about 13 to 15 in number, located adjacent to the head of the epididymis and opening into the epididymal duct. In the epididymis, peristaltic activity of smooth muscle is largely responsible for sperm transport. In the boars, the epididymal content usually undergo continuous dilution from the caput to the cauda epididymis. The time required in the domestic animals for spermatozoon transport through the epididymis, is about 7 to 15 days. Spermatozoa in the tail of the epididymis are 2 to 10 more fertile than spermatozoa from caput, apparently due to abnormalities in locomotion of the sperm cells taken from the head of the epididymis (Larsen, 1986 and Crabo, 1986).

Most of the work relating to morphological changes of spermatozoa during epididymal transit has been on cattle (Igboeli & Foote, 1968; Amann & Almquist, 1962; Cupps & Briggs, 1965; Akusu *et al.*, 1985), Red Sokoto goat (Awojobi & Oyeyemi, 2000) and epididymal transit in West African buck (Oyeyemi *et al.*, 2000). They have varying reports on the site of commencement of motility, stainability, morphology and position of the protoplasmic droplets.

This study was aimed at studying the changes in epididymal spermatozoa, thus contributing to the information on the epididymal transit of spermatozoa in boars.

MATERIAL AND METHOD

Sample collection: Testes from 10 sexually matured boars, aged between 6.0 and 18 months used for this study were collected from boars slaughtered at the pig unit of the Teaching and Research Farm, University of Ibadan and Bodija Abattoir, in Ibadan located about 2 kilometers from the University of Ibadan campus. The body weight, scrotal circumference, and scrotal length were measured for each animal prior to slaughter. The intrascrotal testes were placed in a well-insulated flask maintained at a warm condition (37°C) immediately the animals were slaughtered. These were transferred to the laboratory.

Semen Collection and Microscopy: The right and left epididymis, were trimmed off the body of the testis and semen samples were collected from the three parts of the epididymis through a 1.0cm incision made (with scalpel blade) on any of the locations (caput, corpus and cauda epididymis). Sperm cells were sucked into a Pasteur pipette from the caudal epididymis. The incisions of the caput and corpus epididymis flushed with 2 - 3 drops of 2.9% buffered sodium citrate kept at body temperature. One half of the collected sperm sample was stained using Wells and Awa stains for morphological studies and Eosin and Negrosin stain for live-dead ratio. The second half was mixed with 0.5ml of 2.9% sodium citrate to study the progressive motility, while undiluted samples were used to study the mass activity. Except for mass activity, others were done under high power magnifications.

Statistical Analysis: Simple correlation coefficient was calculated for some testicular parameters. Paired comparisons were done using "t" test where applicable. Analysis of variance was used where means was significantly differed; separation of means was also done using Duncan's multiple range test (Steel & Torrie, 1980).

RESULTS

The summary of the percentage mean values of spermatozoa characteristics at different parts of the epididymis, regardless of age of the animals, is presented in Table I.

Testicular spermatozoa had a mass activity of zero (0). The mass activity for the caput, corpus and caudal epididymis were estimated as 1, 2, and 3, respectively. The progressive motility increase from caput, corpus and to the caudal epididymis. While only very slight vibratile motility was observed from the testes. Mean values of progressive motility for the different portions of the epididymis were estimated to be between, 20 - 30% for caput, 40 - 50 % for the corpus, 80 - 90% for the caudal epididymis and 6 -10% for the testes. Those values were significant ($p<0.5$) when compared Table 2. Percentage live spermatozoa was generally high ranking between 40 - 60% in the testes, 70 -80% in the caput and 80% and above in the corpus and caudal epididymis.

The caput epididymis has a significant higher mean value of 0.20% ($p<0.05$) of abnormal spermatozoa with small head, compared to the corpus 0.05% and cauda 0.00%. There were more of narrow head in caput and corpus than caudal epididymis, which differed significantly ($p<0.05$).

There were higher values ($p<0.05$) of proximal and distal cytoplasmic droplets in caput and corpus epididymis, when compared with the same values in the caudal epididymis.

Abnormalities in the tail to the spermatozoa in terms of coiled tail, curved tail are higher in the caput and corpus epididymis than in the caudal epididymis. The abnormalities in the middle piece (mid-piece) of the spermatozoa also follow the same trend (Table II).

The correlation coefficient for the testicular and epididymal parameters is presented in Table III. Except for testis and epididymal circumference, which was positively correlated ($p<0.01$), others testicular parameters showed positive correlation at each other at $p<0.05$.

DISCUSSION

The observed morphological changes, which spermatozoa undergo in the caput epididymis, are lower when compared to the reports of Oyeyemi *et al.* (2000) 2 - 20% in buck and Amann & Almquist in the bull.

Table I. Mean percentage values of morphological characteristics of spermatozoa in the epididymis.

PE	TWE	SH	NH	LH	PCD	DCI	CT	CUT	TH	BM	TCA
Caput	20	0.20 ^a	0.15 ^a	0.05 ^b	0.35 ^a	0.15 ^a	3.20	3.65	6.30	3.65 ^a	20.05
Corpus	20	0.05 ^b	0.15 ^a	0.15 ^c	0.40 ^a	0.15 ^a	2.90	3.15	5.85	2.85 ^b	18.00
Cauda	20	0.00 ^c	0.05 ^b	0.00 ^a	0.00 ^b	0.00 ^b	3.05	2.20	6.40	1.95 ^c	15.80s

Means with different letters on vertical column differ significantly ($p < 0.05$).

PE: Parts of epididymis

TWE: Total number of epididymis

SH: Small head

NH: Narrow head

LH: Large head

PCD: Proximal cytoplasmic droplet

DCD: Distal cytoplasmic droplet

CT: Coiled tail

CUT: Curved tail

TH: Tailless head

BM: Bent mid-piece

TCN: Total cell normal

TCA: Total cell abnormal

Table II. Mean percentage motility and livability in the testicles and epididymal spermatozoa.

	Percentage motility	Live/ dead ratio or Percentage live
Testes	0.6 ± 2.0^a	1.5:1 60 $\pm$ 10
Caput	23 ± 5.0^b	2.3:1 70 $\pm$ 5
Corpus	45 ± 7.0^c	4.0:1 80 $\pm$ 8
Caudal	85 ± 10.0^d	7.3:1 88 $\pm$ 10

a, b, c, d = value along the vertical column differently superscripted differ significantly ($p < 0.05$).

Table III. Correlation Table for testicular and epididymal parameters.

	BW	WTE	WT	WE	LT	LE	TC	SC	SL	TEC	TB
BW	1										
WTE	0.985	1									
WT	0.982	1.000	1								
WE	0.986	0.992	0.987	1							
LT	0.845	0.842	0.830	0.888	1						
LE	0.978	0.979	0.977	0.980	0.803	1					
TC	0.987	0.995	0.994	0.986	0.829	0.980	1				
SC	0.951	0.940	0.937	0.941	0.819	0.914	0.918	1			
SL	0.911	0.930	0.929	0.922	0.811	0.888	0.896	0.973	1		
TEC	0.718	0.745	0.743	0.748	0.704	0.738	0.692	0.790	0.812	1	
TB	0.976	0.963	0.962	0.956	0.843	0.941	0.967	0.936	0.892	0.697	1

BW: Body weight

WTE: Weight of testis and epididymis

WT: Weight of testis

LT: Length of testis

LE: Length of epididymis

TC: Testicular circumference

SL: Scrotal length

TEC: Testis epididymis circumference

TB: Testicular breadth

The value of abnormal acrosome or head of the spermatozoa in the caput (0.40%) is higher ($p < 0.05$) than corpus (0.35%) and caudal epididymis (0.05%). These values indicated that the abnormalities are from a defect in spermatogenesis, which is a primary abnormality. This is still lower than 5 -10 % minimum allowed for primary abnormality (Zemjanis, 1977; Rozeboom, 2000).

The levels of immaturity of the sperm cells were indicated by cytoplasmic droplets, which showed a significant difference ($p < 0.05$) between the values of caudal epididymis and other two parts. The distal cytoplasmic droplets in the corpus and caput epididymis were higher ($p < 0.05$) than the values in the caudal epididymis. The ability of the sperm to fertilize is required in the distal half of the body (corpus) of the epididymis. This may be the reason for the lower value observed for proximal cytoplasmic droplet. This agreed with the report of Cole & Cupps, (1977) and Crabo.

The tail abnormalities showed that there was significant difference ($p < 0.05$), in the incidence of curved tail in the caudal part when compared to other parts of epididymis. Other abnormalities (coiled tails, looped tails) in this study may partly reflect tertiary abnormalities that originated from epididymis (secondary) or spermatogenesis (primary). These findings are synonymous with the reports of Oyeyemi & Akusu, (1998) and Lea *et al.*, (1978).

The midpiece abnormalities indicated that bent midpiece was significantly higher ($p < 0.05$) in the caput than in corpus and in turn higher ($p < 0.05$) than values in the caudal epididymis. The same pattern was noticed in the curved midpiece but the values did not differ significantly.

The increase in values of tail and midpiece abnormalities may not be unconnected with high scrotal temperature of the animal prior to slaughter. This is in

agreement with the reports of Oyeyemi *et al.* and Larsen, that testicles are affected in species of animals when testicles are not pendulous, also Cameron & Blackshaw (1980) reported that boars kept outdoors have shown signs of testicular degeneration after exposure to extreme winter temperature.

There was a high correlation ($p < 0.05$) between the testicular weight, body weight and other testicular parameters, like weight of testis and epididymis (WTE), length of testis (LT), testicular and scrotal circumference while there was positive $p < 0.01$ testes and epididymis circumference with other testicular parameters. This analysis was synonymous with the report of Willet & Ohm (1957), Land & Carr (1975) and Rozeboom, that testicular

and epididymal parameters are positively correlated.

The higher percentage motility was recorded in caudal ($86 \pm 10\%$), which differed significantly ($p < 0.05$), when compared to the motility values of corpus ($45 \pm 7\%$), caput ($23 \pm 5\%$) and testes ($06 \pm 2\%$). This indicates that more mature and motile spermatozoa capable of fertilization are found in the caudal epididymis of the boar, which is synonymous with reports on other breeds.

It is therefore concluded that boars raised under semi-intensive conditions can produce quality semen that can be used for either natural breeding or artificial insemination as long as the animals are not in anyway incubating any disease.

OYEYEMI, M. O. & UBIOGORO, O. Espermiograma y características morfológicas de los espermatozoides en el testículo y epidídimo del verraco grande blanco en Nigeria. *Int. J. Morphol.*, 23(3):235-239, 2005.

RESUMEN: Para la investigación se utilizaron 10 verracos de cerdos blancos de gran tamaño, con edades entre 6 y 18 meses. Se realizó el espermiograma y se observaron las características morfológicas, en los testículos y en las diferentes partes del epidídimo. El objetivo fue estudiar los cambios que ocurren en los espermatozoides a nivel de los testículos y del epidídimo durante el tránsito epididimal.

Los espermatozoides móviles progresivos se observaron primero en testículos (6-10%), en la cabeza (20-30%), cuerpo (40-50%) y en la cola de epidídimo (85%). Pierden gotas citoplásmicas, en la medida en que los espermatozoides se trasladan ascendiendo hasta el epidídimo. La cabeza del epidídimo presenta un valor de 20,05% de células anormales de espermatozoides, lo que es significativamente muy diferente ($p < 0.5$) de 15-80% en la cola del epidídimo. El total de espermatozoides anormales en el cuerpo del epidídimo, es también más alto que en la cola del epidídimo. El porcentaje de motilidad fue de $0,6 \pm 2,0\%$ en el testículo, el cual era más bajo ($p < 0.5$) que en la cabeza del epidídimo, con $23.0 \pm 5.0\%$. Esta motilidad aumenta desde la cabeza a la cola del epidídimo. La cabeza del epidídimo presenta 0,20% de espermatozoides con cabeza pequeña, lo que difiere significativamente ($p < 0.05$) del cuerpo (0,05%) y de la cola (0,00%).

Los parámetros testicular y epididimal estaban positivamente correlacionados (peso del testículo y del epidídimo; peso del testículo, circunferencia escrotal, circunferencia testicular (peso del epidídimo y peso del testículo ($p < 0.05$), excepto los valores de correlación entre testículos y circunferencia epididimaria (TEC) y otros parámetros ($p = 0.01$).

PALABRAS CLAVE: Espermiograma; Verraco; Espermatozoides; Motilidad.

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