



Influence of Breed, Season and Feed type on Semen qualities and their Relationships with Testicular size in Bucks

Y.E. Waba, S.T. Mbap¹, D.J.U. Kalla¹ and Y.P. Mancha¹

Department of Animal Health and Production, Federal Polytechnic Bali Taraba State, Nigeria

¹Department of Animal Production, Abubakar Tafawa Balewa, University Bauchi, Nigeria

Abstract

The study evaluated the influence of feed, season and breed on the reproductive parameters of West Africa Dwarf (WAD) and Sahel bucks at National Veterinary Research Institute (N.V.R.I), Vom. A total of eight bucks of the two breeds that service does in a body of feeding experiments were used for this study. The eight bucks each were used for semen collection, using electro-ejaculator and evaluation every two weeks. The study lasted for 12 calendar months (from Nov, 2018- Oct, 2019). The bucks were fed Gmelina and cassava peel meal, Gmelina and cowpea husk, Ficus and cassava peel meal and, Ficus and cowpea husk as treatments (T₁-T₄ respectively). The bucks by breed were allotted to the treatments randomly ie two bucks per treatment. The average scrotal circumference, semen volume, sperm concentration, pH, percentages motility, normal and live sperm cells for WAD goats were 17.70 cm, 424.11x10⁶/ml, 6.65 0.50 ml, 84.88, 93.45 and 92.01% respectively. The corresponding values for Sahel goats were 18.12 cm, 405:73x10⁶/ml, 6.92, 0.44 ml, 81.16 91.18 and 92.01 % (P<0.01). Breed, season and diet significantly (P<0.01) influenced most of the parameters tested except the effects of breed and feed on percentage live sperm cells. The WAD bucks had higher semen volume, concentration, motility and normal sperm cells, while the Sahel was better in the other characteristics (scrotal circumference, semen colour and pH level). Generally the early rainy season had higher values for essential semen qualities such as volume, concentration and normal sperm. Ficus containing diet on the other hand resulted in higher concentration, motility and normal sperm cells.

Key words;-Semen quality, feeds, scrotal size

Introduction

Reproductive efficiency including sperm production is the most limiting constraint to efficient animal production in the tropics (Gbadamosi and Egbunike, 1999). The efficiency of sperm production, tends to remain uniform throughout the reproductive life of an animal, but may be significantly altered by nutrition, environment, age, weight, health status, drugs, photoperiod, and chemicals (Togun and Egbunike, 2006). Among these factors, nutrition is the most prominent and therefore improved feeding can optimize spermatozoa production (Salisbury *et al.*, 1978; Rekwot *et al.*, 1987a; 1988).

Nutrition is related to hormone production and the growth of reproductive organs.

Retarded testicular growth results in decreased size of Leydig cells, low differentiation of the seminiferous tubules and interstitial tissues, delayed appearance of motile sperm in ejaculate as observed in gonads of underfed maturing bull-calves (Almeida *et al.*, 2007). Furthermore, restricted diets significantly decrease the number of cells of the spermatogenic series at all stages and the number of Sertoli cells. Thus, cellularity of the tubular epithelium is significantly lowered in low nutrition animals (Almeida *et al.*, 2007). Breed is also a recognized factor in semen variation (Bearden and Fuquay, 1997).

Another similar factor with major contribution to sperm production variation is the environment (Curtis, 1983; Chandler *et al.*, 1985; Cupps, 1991; Bearden and Fuquay,

1997). The environment could be defined in terms of temperature, nutrition, season and management of the animal (Bonsma, 1980). A suitable environment would result in higher sperm production (Bearden and Fuquay, 1997). Health is also listed as a factor that affects sperm production. Leboeuf *et al.* (2000) reported that healthy animals have higher ability to ejaculate viable sperm. In addition, body weight and scrotal-testicular biometry have been reported to have effect on sperm production in bucks (Gemedu and Workalemahu, 2017).

Materials and Methods

Experimental Site, Animal and Management

Eight breeding bucks weighing 28 kg at average, with average of 3 years old, were used as part of a larger nutritional trial for the study. The bucks were moved to Vom, Jos in the second year of study, for semen collection, using the electro-ejaculator and evaluation every two weeks. The study lasted for 12 calendar months between November, 2018 and October, 2019, i.e. Nov-Jan, as early dry, Feb-Apr, late dry, May-July as early and Aug-Oct as late rainy seasons. At Vom, they were housed in a pen made of concrete blocks, cemented floor; open sided for ventilation and a zinc roof. The bucks were fed Gmelina and cassava peel meal (T1), Gmelina and cowpea husk (T2), Ficus and cassava peel meal (T3) and, cowpea husk (T4) respectively. Water was given *ad-libitum*. Factorial design was used. Routine intensive small ruminant management practices were followed.

Data collection and laboratory analysis

- i. Scrotal circumference measurements and semen were collected from the bucks every two weeks into a graduated tube, the volume was read and immersed in a warm water bath

at 40°C. The other sample semen characteristics were subsequently evaluated in the laboratory as described by Zemjanis (1977).

- ii. Semen Volume: As already stated, it was read from the graduated tube and recorded in millimetres to 0.01ml.
- iii. Semen Colour: This was determined using the universal indicator paper by comparison
- iv. Hydrogen ion concentration: The hydrogen ion concentration (pH) was determined using the Chemocraft pH universal indicator paper. The papers are calibrated by colour from 1 to 14. One centimetre of the paper was inserted into the semen for five seconds, removed and air dried. The colour change was compared to the chart on the pH paper. The corresponding colour/number was recorded as the pH of the sample.
- v. Sperm motility

Sperm motility (movement) was scored as outlined by Melrose and Laing (1970). A drop of semen was placed under a cover slip on a microscope slide. The undiluted sample was observed for swirling motion (mass motility) at low power magnification (x100). Subsequently another drop of semen was mixed with a drop of physiological normal saline (0.9% NaCl) under the cover slip (on a slide) and again viewed at x100. The microscope was then switched to x450 for differential count of 10 moving and non-moving sperms. Several fields of view were observed, the differentials count repeated and motility recorded as the average percentage of moving cells.

vi. Sperm concentration determination

The concentration of the spermatozoa was determined using the Accuread photometer, Edition 07/2008. It measures sperm concentration in a suspension at 595nm. The Accuread photometer was first switched on and it was ensured that it was in concentration mode i.e., the point at which the semen image is displayed, viewed and, assessed on the screen of the photometer. Subsequently, 10 µl of raw semen was diluted with 3990 µl of 0.9% NaCl solution in a cuvette using a micropipette. Another cuvette containing only 0.9% NaCl solution was placed in the sample chamber and the equipment set to zero (zero reading). The cuvette was then replaced with the one containing the diluted semen sample which was properly homogenized. The semen concentration was then read.

vii. Live and dead sperm ratio: The live and dead sperm ratio was estimated in accordance with Hancock (1951). A drop of semen was diluted in a ratio of 1:400 with available diluent. A drop of Eosin-Nigrosine stain was placed on a microscope glass slide one centimetre from the end and a drop of the semen added using a pipette. A second slide was placed on top of the first. The slide was circularly and gently turned to ensure uniform mixture of the semen and stain. Excess fluid was blotted from the edge with a tissue and the two slides gradually and carefully drawn apart to produce a thin smear on the first slide. This was quickly dried and labeled. The slide was subsequently mounted on the microscope for counting the live and dead spermatozoa using x400 magnification. Dead sperm were stained and live ones unstained.

viii. Sperm morphology

The slides prepared for live and dead ratio were used for the morphology studies. Live and abnormal spermatozoa percentages were counted using a tally counter. Some 200 spermatozoa were examined from each sample. The total number of abnormal spermatozoa was counted and recorded. The possible types of abnormality were: detached mid-piece and tail, detached head, mid-piece droplet, coiled and bent tails, and acrosomal abnormality (acrosome membrane detached, acrosome outlines and acrosome cap defect). Acrosomal abnormalities however, were determined using smears stained by Giemsa according to Watson (1975).

Data analysis

The data obtained were subjected to analysis of variance procedure as contained in the statistical package of Statistix 8.0(2009) USA. Significant means were compared, using the Duncan's Multiple Range Test (Duncan, 1955).

Results and Discussion

The average scrotal size and semen quality parameters of bucks fed different feedstuffs are presented in Table 1. There were significant ($P < 0.01$) difference among the breeds, seasons, and treatments for all the parameters considered, except live cells by breed and treatment. The Sahel bucks had wider scrotal circumference (18.12 cm) than the WAD (17.70cm). This breed difference was earlier reported Raji *et al.* (2008) with respect to goat breed variations in scrotal and testicular biometry. They found values of 15.41cm and 28.89cm, for SCR and SL in Red Sokoto adult buck and, 20.86 and 25.54cm for matured Borno white respectively, which are higher in the former and lower in the latter than those reported in this present study.

The WAD had higher values of semen volume (0.500 ml), motility (84.33%), normal sperm (93.49%), and sperm concentration (424×10^6 ml), while Sahel goats were higher for the other parameters. This finding, that WAD was higher in essential semen characteristics (volume, motility, normal sperms and concentration) than Sahel, is an indication that, it is better in sperm qualities at least in this environment. It is well documented that WAD reproduces better/faster than Sahel and gives higher kid crop/annum in the present environment compared to the work of Raji *et al.* (2008). However, Sahel breeds of goats do have a poor performance outside the arid, semiarid and Sahelian vegetation zone.

There is optimum sperm characteristic values required for good fertility in the goat. Sultana *et al.*, (2013) gave a volume range of 0.58-1.04 ml that is required for fertility. For viscosity, pH and motility value of +1 on a scale of +1 to +4, 6.04-6.25 and 88.7-94% respectively have been advocated by (Oyeyemi *et al.*, 2000) and Shamsuddin *et al.* (2000). Values outside these ranges may impair fertility. However the values in the present study were lower, which could be due to environmental and seasonal variation of the study area. Too high viscosity may reduce motility. Adequate motility is required at fertilization site (David *et al.*, 2015). For concentration a range of 1.133 ± 0.44 and $3.54 \pm 0.14 \times 10^9$ /ml is considered adequate, which is in consonance with the present study and idle for reproduction in goats.

Early rainy season had higher scrotal circumference of 18.30 cm while the other seasons had similar values (17.63-17.88 cm). Seasonal variation therefore to some extent affected most of the scrotal/semen parameters. This is in consonance with the finding of Bitto and Egbunike (2006) in WAD. Daudu (1984) made a similar, earlier report on Red Sokoto (RS) bucks.

Furthermore, variations in these parameters with season have been reported by Villar *et al.*, (1993), Moreira *et al.*, (2001) and Campos (2003). That early rainy season had higher values for the reproductive parameters considered in the present study than the other seasons had similarly been reported (Maina *et al.* 2006). Goats tended to have better reproductive parameter in rainy than dry seasons. This may be due to feed and weather conditions (Daudu, 1984). However, Maina *et al.* (2006), and Mellado *et al.* (2011) had reported sperm concentration in Sahel and Granadian bucks to range from 3.73×10^9 /ml in the dry and 2.67×10^9 /ml in the rainy season and, 2210 ± 1139 vs $2055 \pm 656 \times 10^6$ /ml respectively.

Generally treatments containing Ficus had almost the highest values for the parameters considered.

This is an indication that Ficus may be more suitable for increase in these reproductive parameters in goats than Gmelina. To boost these reproductive parameters in goats in the area therefore, farmers should incorporate more Ficus browse in the diet of their goats. However, this should be in addition to other supplement such as cassava peel meal and cowpea husk, especially in the dry season. Toe *et al.* (2000), Ugwu (2009) and Antonio *et al.* (2011) have made similar observations.

Mass and progressive sperm motility was significantly ($P < 0.05$) bit lower during early dry and late rainy seasons in this study, which is consistent with Karagiannidis *et al.* (2000) and Barkawi *et al.* (2006) who reported mass and progressive sperm motility to increase from the non-breeding to the breeding season. The percentage live sperm was significantly ($P < 0.05$) higher during late dry seasons. The individual motility ranged from good (rapid rectilinear) to slow and erratic motion.

Semen pH values in the present study were slightly alkaline and above than reported for Rayini goats (Zamiri and Heidari, 2006), and Zaraibi goats (Barkawi *et al.*, 2006). It was similar to the pH of 7.01 and 7.20 reported for Angora goats in Australia (Mendoza *et al.*, 1989). Lastly the semen quality, as measured by the semen index, was greater in early rainy season except for percentage live cells than in the other seasons ($P < 0.05$). This is also similar to the findings in Zaraibi breed, where the highest semen index has been reported during the summer (Barkawi *et al.*, 2006).

Correlation between Scrotal and Semen Parameters of bucks of different breeds, seasons and feeds

Correlations among the scrotal and semen parameters of bucks are presented in Table 2. The correlation values between 0.439 and 0.701, e.g. between semen concentration and sperm motility (0.701) and between motility and colour (0.539) were positive, high, and significant ($P < 0.001$). The value of -0.684 between semen concentration and volume on the other hand was high, negative and significant ($P < 0.001$). Furthermore, the correlation between semen concentration and abnormal sperm (-0.337) and, between semen motility and scrotal circumference (0.322) were medium. The correlation between scrotal circumference and semen volume (0.315) was also medium in value. The others below ± 0.315 (normal sperm and sperm motility, sperm motility and semen pH) were in the low correlation category.

The relationship between scrotal size and semen qualities were only medium to low. Therefore, scrotal size in the present study

cannot be effectively used to estimate semen quality but only slightly. Ugwu (2009) had reported some relationship between scrotal circumference and gonadal and extra gonadal sperm reserves. He however stated that even when correlation values between scrotal size and quality are high, they may indicate spermatogenic efficiency only. The actual volume and other quality parameters may largely depend on method of collection. The low values of significant correlations between scrotal size and sperm qualities in this study might have been due to method of semen collection as stated above. However, Olurode *et al.* (2018) reported the correlation coefficient amongst, scrotal size and semen concentration, semen volume, motility and sperm morphology values of -0.099, -0.056, -0.117, and 0.634 respectively, which was not in consonance with the present finding. They further reported the relationships of semen parameters with values of -0.15, 0.280, -0.397 for semen volume and motility, semen motility and concentration, and semen concentration and morphology respectively, which also did not agree with present study, except for the results of semen motility and sperm concentration. However, the relationships between semen volume, sperm motility, sperm count, semen morphology and, scrotal size were weak and not significant, which is similar to the finding of the present study.

Normal sperm percentage for adequate fertilization in goat has been reported to be 92-96% (Elsheikh and Elhammali, 2015), while Delgadillo *et al.* (1992, reported 37.8-66.3% and Perez *et al.* (1992) reported a range of 59.4-69.0% for live sperm.

Table 1: Mean scrotal size and semen quality of bucks on different feeds

Variables Factors	N	Scrotal circum. (cm)	Semen vol. (ml)	Sperm conc. (x10 ⁶)	Sperm Ph	Sperm motility(%)	Normal sperm (%)	Live cells (%)
Overall	192	17.91	0.47	414.92	6.78	83.02	92.33	92.083
Breed								
WAD	96	17.70 ^b	0.500 ^a	424.11 ^a	6.65 ^b	84.88 ^a	93.49 ^a	92.013
Sahel	96	18.12 ^a	0.443 ^b	405.73 ^b	6.92 ^a	81.16 ^b	91.18 ^b	92.014
SE±		0.138	0.013	8.057	0.51	0.491	0.499	0.376
Season								
Early dry(Nov-Jan)	48	17.63 ^b	0.430 ^b	415.29 ^b	6.58 ^b	81.21 ^b	91.98 ^b	93.438 ^a
Late dry(Feb-Apr)	48	17.88 ^b	0.450 ^b	412.35 ^b	7.04 ^a	85.17 ^a	90.13 ^c	90.583 ^b
Early rainy(May-Jul)	48	18.30 ^a	0.522 ^a	420.54 ^a	6.90 ^a	84.63 ^a	93.92 ^a	92.021 ^{ab}
Late rainy(Aug-Oct)	48	17.84 ^b	0.477 ^{ab}	411.50 ^b	6.60 ^b	81.06 ^b	93.31 ^a	92.292 ^a
SE±		0.195	0.019	11.395	0.072	0.695	0.706	0.532
Treatment								
Gm+ Cpm	48	18.64 ^a	0.508 ^a	372.46 ^c	7.00 ^b	83.31 ^b	88.96 ^c	90.562
Gm+ Cph	48	16.18 ^c	0.422 ^c	307.42 ^d	6.35 ^d	74.71 ^c	91.23 ^b	91.858
Fs+ Cpm	48	18.14 ^b	0.452 ^{bc}	501.25 ^a	7.27 ^a	87.50 ^a	96.65 ^a	92.875
Fs+ Cph	48	18.69 ^a	0.495 ^{ab}	478.56 ^b	6.54 ^c	86.54 ^a	92.50 ^b	93.042
SE±		0.195	0.019	11.395	0.72	0.699	0.706	0.532

**=P<0.01, a, b and c=means within a subset with the same superscript are statistically similar. WAD=West African Dwarf, Gm=Gmelina arborea, Fs=Ficus sycamores, Cpm=Cassava peel meal, Cph=Cowpea husk, N= sample size, circum. = circumference, conc. =concentration, ns=not significant, SE= Standard Error

Table 2: Correlation matrix of scrotal circumference and semen quality of bucks

	Normal	Abnormal	Scrotal circum.	Semen colour	Semen Vol.	Sperm motility	Semen Conc.
Scrotal circum.	-0.339***	0.351***					
Semen colour	0.181 ns	-0.174ns	0.342***				
Semen Volume	-0.081ns	0.080ns	0.315***	0.082ns			
Sperm motility	0.207**	-0.196**	0.322***	0.439***	0.085ns		
Semen conc.	0.345***	-0.337***	0.194ns	0.539***	-0.684 ***	0.701***	
pH. Level	0.030 ns	-0.027ns	0.133ns	0.165ns	-0.038ns	0.284 **	0.351***

*P<0.05

**P<0.01

***P<0.001

ns- not significant

Conclusion and recommendation

The WAD had overall better semen quality in the environment of the study. Generally semen characteristics were better during the early rains than in the other seasons of the year. Similarly Ficus containing diets had higher semen parameter values than the Gmelina counterpart. The early season and Ficus diet were therefore suitable for the goat reproductive indices.

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