SEMEN QUALITY, BIOCHEMISTRY AND MINERAL CONTENT OF FIVE STRAINS OF COCKS RAISED IN THE HUMID ENVIRONMENT OF NIGERIA

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ABSTRACT

The study was conducted to evaluate the genetic effect on semen quality, biochemistry and mineral content of three strains of Nigerian indigenous and two exotic cocks. One hundred (100) adult local breeding cocks comprising 20 normal, 20 frizzle and 20 naked necks, 20 dominant black and 20 dominant blue feather were selected randomly from the Poultry Breeding Unit of the Teaching and Research farm, Ambrose Alli University Ekpoma. Semen was collected using abdominal massage technique and analyzed. Strain significantly ($P<0.05$) affected the semen characteristics examined. The frizzle strain had the highest semen volume while naked neck had the least with the corresponding mean values of 0.59±0.00, 0.56±0.00 and 0.39±0.00ml respectively. The normal-feathered cocks had the highest semen concentration while the naked necked cocks had the least with corresponding values of 21.18±0.03and 18.07±0.16×10^9ml respectively. Mean values of semen pH was significantly ($P<0.05$) different between the strains. The semen pH for all the strains was slightly alkaline and ranged from 7.49±0.05 to 8.30±0.62. Sperm motility was highest and significant ($P<0.05$) for normal feather cocks with mean value of 97.55±1.16%, while the frizzled cocks had the least value of 60.99±0.05%. Semen biochemistry and minerals was affected by strain. Semen biochemistry and minerals was also affected by strain. The naked neck feathered cocks had the highest semen total protein, followed by dominant blue feather, Normal feather, Dominant black feather, while the frizzle feather cocks had the least with corresponding values of 2.59±0.04, 1.82±0.03, 1.63±0.03, and 1.55±0.03 and 1.28±0.02 g/dl respectively. The normal feather strain had the highest semen Albumin while dominant black feather had the least with the corresponding mean values of 0.65±0.04, 0.55±0.02, 0.53±0.03, 0.53±0.02 and 0.36±0.02 g/dl respectively. Mean values of semen Glucose was significantly ($P<0.05$) different among the strains. The semen Glucose for all the strains ranged from 181.95±0.38 to 112.70±0.39 mg/dl. Semen cholesterol, was highest and significant ($P<0.05$) for naked neck feather cocks with mean value of 101.63±0.22, while the frizzled cocks had the least value of 53.24±0.29 mg/dl. The dominant black feather strain had the highest semen sodium while normal feather had the least with the corresponding mean values of 156.75±0.36, 154.76±0.41, 150.35±0.36, 150.16±0.30 and 137.85±0.41 ml/dl respectively. The normal feather strain had the highest semen Albumin while dominant black feather had the least with the corresponding mean values of 0.65±0.04, 0.55±0.02, 0.53±0.03, 0.53±0.02 and 0.36±0.02 g/dl respectively. The naked neck feathered cocks had the highest semen phosphorus, followed by dominant blue feather, Dominant black feather, and frizzle feather while the Normal feather cocks had the least with corresponding values of 18.90±5.79, 13.76±0.06, 12.90±0.34, 12.12±0.05 and 10.18±0.03 m/dl respectively. A significant ($P<0.05$) difference was obtained in the, chloride and potassium content of the semen mineral samples. It was concluded that large genetic variation existed in semen quantity traits in the cocks used for the study and can therefore be included in genetic improvement as gene contributors.

Key words: Frizzle, Indigenous cocks, Normal Naked neck, Dominant blue/black, Exotic, Quality Semen biochemistry and Semen minerals

INTRODUCTION

The reproductive potential of poultry birds (cocks) is often determined to a large extent by the quality of the semen they produce. Evaluation of semen quality and quantity of domestic fowls had been studied extensively (Schneider, 1992; Bah *et al*., 2001; Tuncer *et al*., 2006; Peters *et al.*, 2008 and Isidahomem *et al*., 2009) in indigenous tropical chicken breeds. While the assessment of semen quality characteristics of poultry birds had been reported to be an excellent indicator of reproductive potentials and a major determinant of fertility and subsequent hatchability of eggs (Peters *et al*., 2004). The genetic effects of breeds (Islam *et al*., 2002) and strains significantly affected semen quality and quantity (Schneider, 1992; Bah *et al*., 2001; Tuncer *et al*., 2006; Peters *et al*., 2008). According to Obioha (1992), the use of the Nigerian indigenous poultry for commercial purposes does not make any economic sense, however the Nigerian indigenous chickens account for about 80 to 90 percent of the local population of chickens in Nigeria (Peters *et al*., 2008) and their contribution to survival and adaptability (Ebozoje and Ikeobi, 1995) of poultry population in the tropics is vital to sustainable genetic improvement and biodiversity security of indigenous chicken populations. Indigenous Nigerian chicken breeds have also been reported to have many advantageous gene complexes or gene marker systems, which could be harnessed in the pursuit of genetic improvement of tropically suitable meat and egg chicken types (Machebe and Ezekwe, 2004). Prominent among these major genes are; the naked neck, frizzle and normal feathers (Ajayi *et al.*, 2011). Their genetic diversity and gene pool play important role in the reproductive adaptability (Ebozoje and Ikeobi, 1995; Machebe and Ezekwe, 2004). The value of a species increases in relation to its adaptation, capacity to make socioeconomic contributions, capacity to fill market opportunities and potential for increasing productivity (Birteeb *et al*. 2012). While semen characteristics studies have been broadly used to determine reproductive potentials of breeding cocks, Ilori *et al.*, (2012) argued that other seminal parameters are equally important in understanding cock viability and reproductive potentials. To better understand the reproductive potentials of breeding cocks, therefore, the biochemical content (Langlais and Roberts 1985; Blesbois *et al*. 2000; Khan 2011) as well as mineral properties (McLean 1997; Alavi 2007) are equally important. Various studies on semen characteristic of the domestic fowl have been reported (Schneider, 1992; Bah *et al*., 2001; Machebe and Ezekwe, 2004; Barber *et al.*, 2005; Tuncer *et al*., 2006;Orunmuyi *et al.*, 2013; Peters *et al*., 2008; Mosenene, 2009; Ajayi *et al.*, 2011; Tarif *et al.*, 2013; Sonseeda *et al.*, 2013; Mohan *et al.*, 2013) for the Nigerian indigenous chickens, but very few have compared the semen characteristics side by side with the biochemical and mineral properties. The aim of this study therefore was to determine semen characteristics, biochemistry and mineral content of three known indigenous chicken and two exotic strains.

MATERIALS AND METHODS

Study area: The experiment was carried out in the Poultry Unit of the Teaching and Research Farm, Ambrose Alli University Ekpoma, Nigerian (Lat 6.44^{o}N and Long 6.8^{o}E).This area lies within the South-South geo-political zone of Nigeria and has a prevailing tropical climate with a mean annual rainfall of about 1556mm. Mean ambient temperature ranges between 26^{0}C in December and 34^{0}C in February with relative humidity ranging between 61% in January and 92% in August and a yearly average of 82%. The vegetation represents an interface between the tropical rainforest and the derived savanna.

Management of the birds: A total of 100 cocks generated from indigenous and exotic normal feather, naked neck, frizzled, dominant blue and dominant black feathered parents were used for this study. These consisted of 20 matured cocks each type.

Semen Collection: Semen collection from the sire was accomplished by abdominal massage technique (Lake, 1962).Ejaculated semen was collected into graduated conical tubes and the volume of the sample was recorded to the nearest 0.1ml. Blood stained and contaminated semen was discarded. All

collection apparatus were sterilized after every collection and kept in a safe dry place. Semen was dispensed after releasing the abdominal pressure and allowing the oviduct to return to its natural position. Variations in between the sire strains with respect to semen quantity and quality traits were examined. These traits were examined using the following parameters:

Semen volume: Semen volume from each of the sire strains was measured with the use of collection tubes graduated in ml.

Sperm motility: A drop of semen with the aid of a micro-pipette was placed on a microscope slide, which was then covered with a glass cover slip and examined at a magnification of ×400. Several fields were examined and an estimate to the nearest 10% of the motile sperm was made. The motility determination was carried out by taking into consideration subjective measurements based on the judgment of individuals making the determination and finding the average motility for each strain of chicken. Motility of semen samples is expressed as the percentage of cells that are motile under their own power (Cheng *et al.*,2002).

Sperm concentration: The sperm concentration was measured by the direct cell count method, using the haemocytomete. Normal saline 0.5 was mixed with 1ml of semen at the dilution rate of 1:250. The diluted semen was then picked up using a micropipette. A drop of the diluted semen was then placed from each of two ends of the haemocytometer using a micro pipette and allowed to settle. The loaded haemocytometer was then placed on the microscope set to a magnification of ×400. The spermatozoa's head that fall within the subdivided smaller squares at the four edges and centre of the haemocytometer were counted and the average per strain of bird was recorded based on the judgment of the individuals making the determination. The concentration of sperm per volume was determined using the formula:

$$C = 50{,}000 \times N \times D.$$

Where C = concentration of semen per volume (ml), N = Number of spermatozoa counted, D = Dilution rate.

Semen pH: This was determined with the aid of a calibrated pH meter to two decimal points.

Chemical analysis: Ejaculate collected was subjected to chemical evaluation. Total protein in the semen was determined using the SP400UV/VIS spectrophotometer at 750mm (Lowry *et al*., 1951), whereas glucose and total cholesterol concentration were determined using the colorimetric producer as described by Lindner and Mann (1960). The atomic absorption spectrophotometer as explained by Quinin,*et al.* (1960) was used to analyze other mineral contents of the samples.

Data analyses: Data obtained for the various semen traits were analyzed using the General Linear Model procedures of SAS (1999) in a complete randomized design and Duncan Multiple Range Test was used to separate significantly different means (Gomez and Gomez, 1984).

The model is as specified below:

$$\mathbf{Y_{ijk} = \mu + S_j + e_{ijk}}$$

where Y_{ijk} = the parameter of interest

μ = overall mean for the parameter of interest

S_j = fixed effect of j^{th} sire genotype (j = 1-5)

e_{ijk} = Random residual error.

RESULTS

The effect of genotype of the three Nigerian indigenous cocks and two exotic strains on semen characteristic, biochemistry and minerals are presented in Tables 1,2 and 3. Average for semen volume,

concentration, pH and sperm motility varied significantly (P<0.05). Semen volume was highest for Frizzle feather, followed by Normal feather, Dominant black, Dominant blue and naked neck (0.59±0.00, 0.56±0.00, 0.47±0.00, 0.43±0.00 and 0.39±0.00ml) respectively. Concentration were significant (P<0.05) and higher in normal feathered cocks (21.18±0.03, 20.56±0.04, 20.31, 20.19±0.09 and 18.07±0.16×10^9/ml) than dominant black, dominant, blue, frizzle feathered cocks and naked neck respectively. Semen pH also varied significantly (p<0.05) between the five strains of cocks examined. Semen Motility was highest in Dominant black, Dominant blue, followed by Dominant black, Dominant black, Normal, Naked neck and frizzle feather(99.20±0.41.99.15±0.42.97.55±1.16 , 80.10±0.43 and 60.99±0.05%) respectively. Semen biochemistry and minerals was significantly affected (p<0.05) between the five strains of cocks. Total protein ranged from 1.28±0.02 g/dl for frizzle to 2.59±0.04 g/dl for naked neck. Albumin value was higher (0.65±0.04 g/dl) in normal feather, followed by dominant blue feather (0.55±0.02 g/dl), though frizzle and naked neck was not significantly different (p>0.05) with the values of 0.53±0.03 and 0.53±0.02 g/dl but differ significantly (p<0.05) from the dominant black strain (0.36±0.02 g/dl) respectively. Glucose was highest (181.95±0.38 mg/dl) in frizzle semen and least in dominant blue strain with the value of 112.70±0.39 mg/dl.

Table 1: Semen characteristics of the three Nigerian indigenous and exotic cocks

Traits	No	Normal	Frizzled	Naked neck	Dominant Black	Dominant Blue
Volume(ml)	20	0.56±0.00^b	0.59±0.00^a	0.39±0.00^e	0.47±0.00^c	0.43±0.00^d
Concentration (ml)	20	21.18±0.03^a	20.19±0.09^c	18.07±0.16^d	20.56±0.04^b	20.31±0.02^c
PH	20	8.30±0.62^a	7.49±0.05^b	7.49±0.05^b	7.52±0.04^b	7.53±0.05^b
Motility (%)	20	97.55±1.16^a	60.99±0.05^c	80.10±0.43^b	99.15±0.42^a	99.20±0.41^a

Mean in the same rows with different superscript differs (p<0.05) significantly

Table 2: Semen biochemistry and minerals of three Nigerian indigenous and two exotic cocks

Traits	No	Normal	Frizzled	Naked neck	Dominant Black	Dominant Blue
Total Protein(g/dl)	20	1.64±0.03^c	1.28±0.02^e	2.59±0.04^a	1.55±0.03^d	1.82±0.03^b
Albumin(g/dl)	20	0.66±0.03^a	0.53±0.03^b	0.53±0.02^b	0.36±0.02^c	0.55±0.02^b
Glucose(mg/dl)	20	179.35±0.48^b	181.95±0.38^a	126.35±0.33^c	122.45±0.36^d	112.70±0.39^e
Cholesterol(mg/dl)	20	59.69±0.38^d	53.24±0.29^e	101.63±0.22^a	72.36±0.31^b	67.02±0.37^c
Sodium(ml/dl)	20	138.10±0.38^d	154.76±0.41^b	159.16±0.30^c	156.75±0.36^a	150.35±0.36^c
Chlorine(ml/dl)	20	92.90±0.32^a	92.90±0.31^a	68.90±0.56^c	92.00±0.45^{ab}	91.25±0.38^b
Phosphorus(m/dl)	20	10.18±0.03^b	12.12±0.05^{ab}	18.90±5.79^a	12.90±0.04^{ab}	13.76±0.06^{ab}
Potassium(ml/dl)	20	15.62±0.15^b	12.27±0.08^d	15.55±0.09^b	16.43±0.05^a	14.64±0.11^c
Calcium(mg/dl)	20	7.66±0.04^a	6.99±0.05^b	5.92±0.03^d	6.82±0.03^c	4.89±0.07^e

Mean in the same rows with different superscript differs (p<0.05) significantly

Semen cholesterol was lowest in Fizzle semen (53.24±0.29 mg/dl) and highest in naked neck (101.63±0.22 mg/dl) followed by the two exotic, then normal feather chicken semen values. Naked neck semen had highest values for both Sodium (154.76±0.14 mg/dl) and the least was recorded in the Normal feather strain (138.10±0.38). Chloride value was highest in Normal feather strain (92.90±0.32 mg/dl) and least in the Naked neck strain (68.90±0.56 mg/dl), while phosphorus was highest in naked neck (18.90±5.79 m/dl) and normal feather (10.18±0.03mg/dl) semen respectively. Potassium was highest in dominant black (16.43±0.05 mg/dl) and frizzle feather showed least semen (12.27±0.08 mg/dl) respectively. Calcium value was highest in Normal feather strain (7.66±0.04 mg/dl) and least in the Dominant black feather strain (4.89±0.07 mg/dl).

DISCUSSION

The mean semen volume from the strains probed in this study compared favorably with that from both exotic and other indigenous cocks of the tropics. Orunmuyi *et al.,* (2013) reported a mean of 0.55 ml for semen volume, Bilcik *et al.* (2005) reported the range of 0.34 -0.59 ml for broiler cocks and 0.40-0.73 ml were obtained by Peters *et al.* (2008) on seven different indigenous chickens. In the same vane, Bah *et al*.(2001) reported 0.28ml for breeding cocks of the Sahel, while Tuncer *et al*.(2006) documented 0.7ml for Denizli cocks. The observed range of 0.39 – 0.56ml compared favorably with the range of 0.37-0.73ml, reported by peters *et al*. (2008) for Nigeria indigenous strains.

Variations arising from semen concentration and sperm motility for normal feathered, naked neck, frizzled, dominant blue and dominant black feathered cocks could be attributed to their different genetic background and their natural tendencies. Average semen motility of 83.50 % was reported by Ornmuuyi *et al.,* (2013), Peters *et al.* (2008) reported a range of 70% to 87.35% while Tabatabaei *et al.* (2010) reported a value of 73.9% for Iranian broiler breeder chickens. Concentration of 3.91 by Orunmuyi *et al.,* (2013) was within the range of 3.40-9.70 billion/cc reported by Bilcik *et al.* (2005) but lower than the 4.3 billion sperm/ml obtained by Moya *et al.* (1996) in broiler cocks and 6.6 billion sperm/ml reported by Tabartabaei *et al.* (2009) on Ross broiler breeders. It was however higher than values of 2.17-3.14 x109 reported by Tabartabaei *et al*. (2010) and 2.26 x 10^9 by Bah *et al*. (2001) in Nigerian local breeder cocks.

Values obtained for semen pH in this study compared well with the range reported by Etches (1998). The mean pH obtained by Orunmuyi *et al.* (2013) was 7.40. Range obtained by Peters *et al.* (2008), Gebriel *et al.* (2009) and Abd El Ghany *et al.* (2011) were also in consonance with the current findings in this study. Differences in values of semen quality characteristics of the roosters used in this study and some of those reported in literatures can been attributed to effects of strain, body weight, age and season. According to Abd El Ghany *et al.* (2011). Strain differences affect semen volume, concentration and motility but not pH. However, Udeh *et al.* (2011) reported a significant difference in volume but not in concentration and motility and attributed this to strain effect. Makhafola *et al.* (2012) also reported strain differences in ejaculate volume, semen pH, sperm concentration and total motility, while Tabartabaei *et al.* (2009) reported significantly higher motility in indigenous roosters than from exotic males.

The differences in mean values arising from semen total protein and cholesterol levels of naked neck, frizzled and normal feathers, could aiso have been due to different genetic background and natural tendencies. However, the superiority of naked neck over the other strains studied could be as a result of differences in gene structure (Isidahomen *et al*. 2009). However, some authors had reported that the higher the level of cholesterol, the lower the fertility (Ansal, 1985). Al-Daraji *et al*. (2011) reviewed the relationship between cholesterol and phosphorus and concluded that higher cholesterol to phospholipids ratio of cells such as spermatozoa, promotes higher degree of membrane cohesion and permeability. He also stated that cholesterol in seminal plasma may inhibit fertilization by inhibiting membrane fusion during acrosome reaction as a result of its incorporation into lipid bilayers.

Comparing the potassium levels in the semen of the five strains the normal feathered strain had a highest value. Nevertheless, the potassium values recorded from the five strains were observed to be adequate to support growth. Sodium and chlorine values favoured the frizzled feathered cocks. This value in relation to gene structure could support growth and aid proper bone formation in all the five strains. Activation of motility depends on both intra and extra cellular Sodium (Na+) and potassium (K+), lower level of these minerals when induced by environmental factor had been attributed to

lowered reproductive performance in cocks (Barna *et al*. 1998; Karaca *et al*. 2001; 2002a and b). Mineral effect on motility, fertility and hatchability had been reported to be poor when semen chloride (Cl-) and glucose which are very crucial for spermatozoa performance were low (Ilori *et al.* 2012). Higher fertility and hatchability had been attributed to better performance in terms of seminal plasma potassium (K+), chloride (Cl-) and albumin (Ilori *et al.* 2012). Severe phosphorus deficiency had also been reported to cause the lower fertility.

CONCLUSION

The semen characteristic of Nigerian indigenous and two exotic cocks were observed in the study to have compared favorably with both exotic and local strains from other regions especially with regards to the semen volume, concentration, motility pH, biochemistry and semen minerals. The higher semen volume and total sperm count is an indication of the superior genetic tendencies of Nigerian indigenous cocks for reproductive ability and higher fertility. These indigenous strains should be further evaluated for genetic merit for including them as valuable gene pool for genetic conservation for immediate and future use in the genetic improvement of local chickens and genetic breeding and conservation policies.

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