Effect of Ambient Temperature on Reproductive and Physiological Traits of Nigerian Indigenous Chickens

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Abstract

Genetic adaptation and response to selection depend on genetic variation within and between populations, whereas characterization of the population is a first step towards its improvement. This study demonstrates that variation in genotypes observed in reproductive performance observed among the Nigeria indigenous chicken under the tropical ambient temperature could be explained by the difference observed in the seminal plasma biochemical parameters evaluation. A total of Sixty (60) adult local breeding cocks comprising of 20 normal, 20 frizzle and 20 naked neck cocks were selected randomly from the poultry breeding unit of Teaching and Research farm Ambrose Alli University Ekpoma. Semen was collected from them using abdominal massage technique and analyzed for semen characteristics. The normal feathered cocks had the highest values of rectal temperature (RT), respiratory rate (RR), pulse rate (PR) and heat stress index (HSI) while the least was observed among the naked neck cocks. The semen pH for all the strains was slightly alkaline and ranged from 8.30±0.62 for normal-feathered to 7.49±0.05 for naked neck of the Nigerian local cocks. On Semen motility, normal feather cocks had the highest (P<0.05) value with a mean value of 97.55±1.16%, while the frizzled cocks had the least value of 60.99±0.05% respectively. Normal feathered cocks had the highest value of calcium (Ca++) and albumin (Alb), naked neck with the highest level of total protein (TP), phosphorus (P) and total cholesterol (TC) while the frizzled feathered chicken performed better in terms of sodium (Na+) and glucose (Glu). However both naked and frizzled feathered cocks had higher values of seminal plasma potassium (K+) and chlorine (Cl-). Pairwise correlations of the semen characteristics ranged from low to high values. From factor analysis with varimax rotation of the intercorrelated traits, two principal components (PC) which accounted for 84.13% of the variation in motility chicken semen were extracted. The difference observed could be exploited in management, conservation and selection decision under tropical decisions.

Keywords: Cocks, semen physical characteristic, seminal plasma parameters, heat stress index, indigenous chickens
Introduction

Reproduction is the most important requirement of poultry breeding while sperm fertilizing ability is the basis of successful reproduction. The reproductive potentials of poultry birds (cocks) are determined to a large extent by the quality of the semen it produces. Semen volume, sperm concentration and motility are conventionally used as a measure of sperm fertility but some parameters of seminal plasma can provide a better understanding of semen quality (Khan 2011). Sperm motility is essential for optimum fertility in fowl and is influenced by plasma osmolality or factors in seminal plasma and by several parameters as temperature, pH and ions such as Sodium (Na⁺), Potassium (K⁺), Calcium (Ca++) (Alavi 2007; McLean 1997). Research has shown that male broiler breeder contributed more to heat-induced infertility than the female. When the male broiler breeder was exposed to a temperature of 32°C, male fertility declined to 42% and in vivo sperm-egg penetration declined to 52%, compared to values obtained from males that were maintained at 21°C. This shows that heat stress affect sperm viability through qualitative and quantitative depression of semen characteristics such as spermatozoa motility, concentration, volume and consistency (Ayo et al. 2011; Karaca et al. 2002a; McDaniel et al. 1995, 1996). Heat stress may affect osmotic equilibrium and ionic channels that are significant in the interplay between spermatozoa, its environment and the egg, thus distorting spermatozoa homeostasis, its behaviour or its metabolic machinery (Darszon et al. 1999). Ca++, Na⁺, K⁺, and Magnesium (Mg++) are involved in cellular metabolism and influence biochemical activities of spermatozoa (Cummings and Huston 1976; Albert et al. 1994). The activation of sperm motility depends on intracellular and extracellular calcium (Ca++), extracellular sodium (Na⁺) and potassium (K⁺). Ion concentrations increased in whole semen when birds were heat stressed, which may be due to increase in sperm concentration (reviewed by karaca 2002b). Karaca (2001) claimed that change in sperm and plasma Ca++, Chloride (Cl⁻) and Na⁺ concentrations may be related to heat stress infertility while he stated that seminal plasma from semen samples with heat stressed sperm contained lower Ca++, Na⁺, and Cl⁻ concentrations when compared with that from control sperm. Fowl exposed to warm temperatures in vitro releases ca++ with decreased motility and vice versa when incubation temperature is lowered (Karaca et al. 2002b), likewise the movement of cocks from a region of 19°C ambient temperature to a hotter environment of 30°C increases whole semen Na⁺ and K⁺ concentrations but not seminal plasma K⁺ concentrations. It is known that the influx of intracellular Ca++ is important to spermatozoa physiology; flagella motility, sperm respiration, membrane function and metabolism, and to the fusion of acrosome vesicle while any interruption of this critical ionic functions frustrates viability and fertility especially at higher temperature (Ashizawa et al. 1992; Barna et al. 1998; Karaca et al. 2002). Cholesterol has been shown to be involved in the process of capacitation in mammalian spermatozoa and an important role in fluidity and structure of the plasma membrane (Blesbois et al. 2000; Langlais and Roberts 1985). According to Ansah and Buckland (1982), the correlation between seminal plasma cholesterol and fertility of semen were negative. Proteins and serum replacements have also been reported to affect sperm activity. Seminal plasma basic proteins bind to outer surface of spermatozoa membrane while some membrane-bound basic proteins may increase the permeability of biological membrane. Glucose is one of the carbohydrates in the carbohydrate-rich zone on the sperm surface of chicken known as glycocalyx which is essential for immunoprotein in the female tract and early gamete interactions (Pelaez et al. 2011). Parker and McDaniel (2006) reported that increased availability of nutrients such as glucose or fructose as well as oxygen improves sperm motility and fertilizing ability by providing energy as a substrate for the production of ATP.

Several reports on semen characteristic of the domestic fowls have indicated that breed, strain and season significantly affect semen quality and quantity (Bah et al. 2001; Schneider 1992; Peters et al. 2008; Tuncer et al. 2006) especially in the tropics. The tropics are characterized with high ambient temperature, rainfall, high direct and indirect solar radiation and humidity while their interaction with air speed and radiant heat have
deleterious effects on productive performance of different poultry species. Measures of rectal temperature (RT), pulse-rate (PR) and respiratory rate (RR) are some of the most important determinants of the adaptation of poultry to the tropical environment. They also, to a large extent, determine the profitability of the poultry enterprise (Ilori et al. 2012). Animal is considered to be stressed when it has to alter its physiology and behaviour to adapt to adverse environmental and management conditions. This adaptation involves a series of neuroendocrinological, physiological, and behavioural responses which act to equilibrate animal functions. The maintenance of body temperature within physiological limits is necessary for the animal to remain healthy, survive, and maintain its productivity and longevity (Marai et al. 2007). The indigenous Nigerian breed/strain of chickens have been reported to have many advantageous gene complexes or gene markers, that could be harnessed in the development of meat or egg type chickens suitable for used in the tropics (Machebe and Ezekwe 2004). Although, the results of several studies on semen characteristic of the domestic fowl have been reported however, little has been reported on the mineral content of semen from the Nigeria local chicken with particular emphasis on the influence of major genes. (e.g frizzled feathered, naked neck and sex-linked dwarfism). These genes play an important role in the reproductive adaptability of the Nigerian local chickens (Ebozoje and Ikeobi 1995). Basic knowledge of seminal plasma composition, sperm characteristics (sperm motility, concentration, volume), and behavior of sperm relative to our tropical environmental conditions can expand understanding of variation in reproductive performance between these chicken genotypes. Therefore this study was to compare the influence of genotype of the Nigeria indigenous cocks on semen quality and quantity traits.

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°N and Log 6.8°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°C in December and 34°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. The data was taken between January and March, 2011. The average meteorological data during the period was; rainfall (38.95mm), temperature (26.5°C), relative humidity (67.93%), sunshine (4.80) and wind speed (0.50m/s).

Management of the birds

A total of 60 cocks consisting of 20 matured cocks each of normal feather, naked neck, and frizzled feathered were used for this study. The chicks were brooded in deep litter pens according to their genetic groups for four 4 weeks at a brooding temperature of 34°C. These genetic group of chicks generated were reared for 25 weeks. All chicks were wing-tagged for proper identification and subjected to the same management practices throughout the experimental period. Medication and vaccinations were carried out accordingly against stress and diseases. Necessary vaccinations against Newcastle, fowl pox and Gumboro diseases as well prophylactic antibiotics and anticoccidial drugs were also administered to the birds.

Feeds and Feeding: commercial feeds were fed to the birds ad libitum with starter marsh containing 20%CP, 2996Kcal/kg M E from day-old to 4 weeks of age and growers marsh containing 15.86% CP, 2716Kcal/kg M E was offered from 4-15 weeks of age. Breeders marsh containing 16.80% CP, 2823Kcal/kg M E from 15-24 weeks of age. Clean water was provided continuously during the experimental period. The feed is as described in Isidahomen et al. (2012)

Semen physical characteristics

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
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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 pre warmed 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 was expressed as the percentage of motile spermatozoa having moderate to rapid progressive movement and cells that are motile under their own power (Hafez 1978). At least 10 microscopic fields were examined for each semen sample.

Sperm concentration: The sperm concentration was measured by the direct cell count method, using improved Neubauer haemocytometer slide (GmbH & Co., Hamburg, Germany). 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.

**Seminal plasma biochemical parameters**

Seminal plasma was separated from the semen by centrifugation at 3500g x 10 min and stored at -20°C before analysis. The seminal plasma parameters were evaluated as follows.

Total protein (TP) in the seminal plasma was determined using the SP400UV/VIS spectrophotometer at 750mm (Lowry et al. 1951) whereas glucose (Glu) and total cholesterol (TC) concentration were determined using the colorimetric producer as described by Lindner and Mann (1960). The atomic absorption spectrophotometer as explained by Quinn, et al. (1966) was used to analyze other mineral contents of the samples.

**Statistical analyses**

Means, standard errors and coefficients of variation of semen characteristics and seminal plasma ions were calculated. Pearson correlation coefficients (r) were also determined. From the correlation matrix, data were generated for the principal component factor analysis. Anti-image correlations, Kaiser-Meyer-Olkins measures of sampling adequacy and Bartlett’s Test of Sphericity were computed to test the validity of the factor analysis of the data sets. According to Everitt et al. (2001), principal component analysis (PCA), is a method for transforming the variables in a multivariate data set \( x_1, x_2, ---x_p \), into new variables, \( y_1, y_2, ---y_p \) which are uncorrelated with each other and account for decreasing proportions of the total variance of the original variables defined as:

\[ Y_1= a_{11}x_1 + a_{12}x_2 + + + + + + + + + a_{1p}x_p \]

\[ Y_2= a_{21}x_1 + a_{22}x_2 + + + + + + + + a_{2p}x_p \]

\[ Y_p= a_{p1}x_1 + a_{p2}x_2 + + + + + + + + a_{pp}x_p \]

with the coefficients being chosen so that \( y_1, y_2, ---y_p \) which account for decreasing proportions of the total variance of the original variables, \( x_1, x_2, ------, x_p \).

Factors were rotated with variimax rotation of Kaiser in order to minimize the sum of variance of
aij2 quadratic weight. The stepwise variable selection multiple regression procedure was used to obtain models for predicting body weight from body measurements (a) and from established principal component (b).

\[ M = a + B_i x_i + \ldots + B_k x_k \] (a)

\[ M = a + B_i PC_i + \ldots + B_k PC_k \] (b)

where; \( M \) is the semen motility, \( a \) is the regression intercept, \( B_i \) is the i-th partial regression coefficient of the i-th semen measurement, \( X_i \) or the i-th principal component.

The factor programme of SPSS (2010) statistical packages was used for the analysis.

Principal component analysis was performed to minimize overall variables into few meaningful variables that contributed most to variations in the populations.

Results

Effect of genotype on heat tolerance traits

The least squares means of the effects of genotype on heat tolerance traits showed that genotype significantly (p<0.05) affected traits measured (Figures 1-4). The normal feathered cocks had the highest rectal temperature (41.27±0.02) at 24 weeks, followed closely by the naked-neck (41.19±0.03) and the frizzle-feathered genotype (40.1±0.01). Pulse rate and respiratory rate was higher and almost the same in both normal and the frizzle-feathered cocks. Naked-neck chicken had the least respiratory rate. Genotype also had significant (p<0.05) effect on HSI. The normal feathered chicken had the highest HSI followed by the frizzle-feathered and the naked neck cocks.

Fig. 1: Mean RT (°C) of chickens as affected by genotype and age (weeks).

Fig. 2: Mean PR (beats/min) of chickens as affected by genotype and age (weeks).
Effect of genotype on semen physical characteristics

The least squares means of the effect of genetic group on semen physical characteristics revealed significant (p<0.05) differences (Table 1) for all the parameters measured.

Sperm motility and concentration are significantly (P<0.05) higher in normal feathered cocks (97.55% and 21.18×10^9/ml) than frizzle feathered cocks (60.20% and 20.19×10^9/ml) and naked neck (80.10% and 18.07ml) respectively. Semen volume and pH also varied significantly (p<0.05) between the three strains of indigenous cocks.

Effect of genotype on seminal plasma biochemical parameters

The seminal plasma biochemical parameters evaluation were significantly (p<0.05) affected among the three strains of indigenous cocks as shown by the least squares means in Table 2. The seminal TP, TC and P were higher in naked-neck cocks, Glu and Na+ were higher in frizzle-feathered males while Alb and Ca+ were higher in normal feathered males. K+ was higher in both normal and naked-neck cocks while Cl- was higher in both normal and frizzle-feathered chicken.
een seminal plasma characterized by high positive  


e values by the underlying factor which was found to be very high (0.87). The overall significance of the overall matrices tested with Bartlett’s Test of Sphericity for all the semen measurements (chi-square = 1847; P<0.01) validates the factor analysis of the data sets. The communalities, which represent the proportion of the variance in the original variables that is accounted for by the factor solution ranged from 0.084 to 0.995 which further reiterate the appropriateness of the PCA. The analysis revealed two discernable patterns of variation in the chicken semen traits. These coefficients show the relative contribution of each trait to a particular principal component (factor). The first principal component (PC1) accounted for 55.06% of the observed variation and was characterized by high positive loading (factor-variate correlations) for semen volume, concentration, Seminal plasma Glu, Ca++, and Cl-. The second component (PC2) contributed 29% to the observed variation. The variables most associated with PC2 were pH, motility and K+ hence could be termed ‘motility measurement’. PC2 which was mutually orthogonal to PC1, presented patterns of variation independent of PC1 (multicolinearity already avoided). The percentage

**Phenotypic correlation**

Pearson’s coefficient of correlation matrix for semen characteristics and seminal plasma biochemical parameters is shown in Table 3. Low to high significant (P<0.05, 0.01) estimates (positive and negative) were observed between semen characteristics and seminal plasma biochemical parameters except for phosphorus. The highest coefficient (r = 0.99) was observed between semen volume and seminal plasma Glu while the lowest (r = -0.99) was observed between semen volume and seminal plasma TC and between seminal plasma Glu and TC.

**Principal component matrix**

After varimax rotation, principal component weights derived from the correlation matrix of the measurements are presented in Table 4. Anti-image correlations computed showed that partial correlations were low, indicating that true factors existed in the data. This was supported by Kaiser-Meyer-Olkin measure of sampling adequacy from the diagonal of partial correlation, revealing proportion of the variance in the semen measurements caused by the underlying factor

![Image](https://example.com/image-url)

**Table 1**: Means, standard error (SE) and coefficients of variation (CV) for semen characteristics of the three Nigeria local cocks as affected by genotype

<table>
<thead>
<tr>
<th>Genotype</th>
<th>No</th>
<th>Volume (ml)</th>
<th>CV</th>
<th>Concentration (×10⁹/ml)</th>
<th>CV</th>
<th>Motility (%)</th>
<th>CV</th>
<th>pH</th>
<th>CV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal feather</td>
<td>20</td>
<td>0.56±0.01⁰</td>
<td>12.14</td>
<td>21.18±0.03¹</td>
<td>0.64</td>
<td>97.55±1.16²</td>
<td>5.31</td>
<td>8.30±0.62³</td>
<td>3.48</td>
</tr>
<tr>
<td>Frizzle feather</td>
<td>20</td>
<td>0.59±0.02⁴</td>
<td>9.28</td>
<td>20.19±0.09⁴</td>
<td>1.90</td>
<td>69.95±0.25⁵</td>
<td>3.42</td>
<td>7.49±0.15⁵</td>
<td>2.74</td>
</tr>
<tr>
<td>Naked neck</td>
<td>20</td>
<td>0.39±0.02⁶</td>
<td>13.24</td>
<td>18.07±0.16⁶</td>
<td>4.08</td>
<td>80.10±0.43⁷</td>
<td>2.39</td>
<td>7.49±0.35⁷</td>
<td>2.89</td>
</tr>
</tbody>
</table>

Mean ± SEM in the same column with different superscript differs (p<0.05) significantly.

**Table 2**: Means, standard error (SE) and coefficients of variation (CV) for seminal plasma ion concentration of the three Nigeria local cocks as affected by genotypes

<table>
<thead>
<tr>
<th>Parameters</th>
<th>No</th>
<th>Normal feather</th>
<th>CV</th>
<th>Frizzle Feather</th>
<th>CV</th>
<th>Naked neck</th>
<th>CV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total protein (ng/dl)</td>
<td>20</td>
<td>1.63±0.03⁰</td>
<td>6.88</td>
<td>1.28±0.02¹</td>
<td>6.99</td>
<td>2.59±0.04²</td>
<td>6.79</td>
</tr>
<tr>
<td>Albumin (ng/dl)</td>
<td>20</td>
<td>0.65±0.04⁴</td>
<td>25.69</td>
<td>0.53±0.03⁵</td>
<td>2.45</td>
<td>0.53±0.02⁵</td>
<td>16.31</td>
</tr>
<tr>
<td>Glucose (nmol/l)</td>
<td>20</td>
<td>179.65±0.54⁴</td>
<td>1.33</td>
<td>181.95±0.38⁴</td>
<td>0.93</td>
<td>126.35±0.34⁴</td>
<td>1.18</td>
</tr>
<tr>
<td>Cholesterol (nmol/l)</td>
<td>20</td>
<td>59.73±0.37⁵</td>
<td>2.75</td>
<td>53.24±0.29⁵</td>
<td>2.42</td>
<td>101.63±0.22⁵</td>
<td>0.96</td>
</tr>
<tr>
<td>Potassium (nmol/l)</td>
<td>20</td>
<td>15.72±0.16⁶</td>
<td>4.59</td>
<td>12.27±0.08⁶</td>
<td>3.06</td>
<td>15.55±0.08⁶</td>
<td>2.48</td>
</tr>
<tr>
<td>Sodium (nmol/l)</td>
<td>20</td>
<td>137.85±0.41⁶</td>
<td>1.31</td>
<td>154.76±0.14⁶</td>
<td>1.18</td>
<td>150.16±0.30⁶</td>
<td>0.89</td>
</tr>
<tr>
<td>Calcium (nmol/l)</td>
<td>20</td>
<td>7.65±0.18⁷</td>
<td>2.30</td>
<td>6.99±0.22⁷</td>
<td>2.99</td>
<td>5.92±0.14⁷</td>
<td>2.40</td>
</tr>
<tr>
<td>Phosphorus (nmol/l)</td>
<td>20</td>
<td>10.18±0.03⁸</td>
<td>1.27</td>
<td>12.12±0.05⁸</td>
<td>1.81</td>
<td>18.90±5.79⁸</td>
<td>1.37</td>
</tr>
<tr>
<td>Chlorine (nmol/l)</td>
<td>20</td>
<td>92.85±0.32⁹</td>
<td>1.53</td>
<td>92.90±0.31⁹</td>
<td>1.48</td>
<td>68.90±0.56⁹</td>
<td>3.61</td>
</tr>
</tbody>
</table>

Mean ± SEM in the same Rows with different superscript differs (p<0.05) significantly.
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of total variance is used as an index to determine how well the total factor solutions accounted for what variable together represent. The objective of principal component analysis (PCA) is to account for the maximum portion of the variance present in the original set of variables with a minimum number of composite variables. It assumes that the unique variance represents a small portion of the total variance (Parés i Casanova et al. 2012).

Prediction of Motility from interdependent semen characteristics and their independent principal component matrix

The interdependent original semen traits and their independent principal component factor scores were used to predict motility of chickens (Table 5). The result of the stepwise multiple regression analysis revealed that K+ accounted for 20% of the variation in motility. When Ca++ was added to the model, the proportion of explained variance increased to 48%. Further addition of TC and Na+ accounted for 59% of the total variance in semen motility. Both principal component contributed only 20% of the variation in motility.

Table 3: Phenotypic correlation among semen traits in Nigeria local chickens

<table>
<thead>
<tr>
<th></th>
<th>Vol</th>
<th>Con</th>
<th>pH</th>
<th>Mot</th>
<th>Glu</th>
<th>K+</th>
<th>Na+</th>
<th>TC</th>
<th>Alb</th>
<th>TP</th>
<th>Ca++</th>
<th>P</th>
<th>Cl</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vol</td>
<td>-</td>
<td>0.37*</td>
<td>-0.46</td>
<td>0.99</td>
<td>-0.55**</td>
<td>-0.12ns</td>
<td>-0.99**</td>
<td>0.15ns</td>
<td>-0.97**</td>
<td>0.84</td>
<td>-0.23ns</td>
<td>0.98**</td>
<td></td>
</tr>
<tr>
<td>Con</td>
<td>-</td>
<td>-</td>
<td>0.70***</td>
<td>-0.03ns</td>
<td>0.88***</td>
<td>-0.12ns</td>
<td>-0.51**</td>
<td>-0.85**</td>
<td>0.27*</td>
<td>-0.75**</td>
<td>0.92**</td>
<td>-0.23ns</td>
<td>0.88***</td>
</tr>
<tr>
<td>pH</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>0.65***</td>
<td>0.47**</td>
<td>0.51***</td>
<td>-0.94**</td>
<td>-0.39**</td>
<td>0.40**</td>
<td>-0.26*</td>
<td>0.76**</td>
<td>-0.17ns</td>
<td>0.49***</td>
</tr>
<tr>
<td>Mot</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-0.37**</td>
<td>0.95***</td>
<td>-0.80**</td>
<td>0.45**</td>
<td>0.28*</td>
<td>0.55**</td>
<td>0.04ns</td>
<td>0.03ns</td>
<td>-0.35*</td>
</tr>
<tr>
<td>Glu</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-0.47**</td>
<td>-0.21ns</td>
<td>-0.99**</td>
<td>0.18ns</td>
<td>-0.95**</td>
<td>0.89**</td>
<td>-0.25ns</td>
<td>0.99***</td>
<td></td>
</tr>
<tr>
<td>K+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-0.69***</td>
<td>0.54**</td>
<td>0.24ns</td>
<td>0.63***</td>
<td>-0.10ns</td>
<td>0.06ns</td>
<td>-0.43ns</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Na</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-0.35**</td>
<td>0.01ns</td>
<td>-0.56**</td>
<td>-0.11ns</td>
<td>-0.24ns</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TC</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-0.16ns</td>
<td>0.97***</td>
<td>-0.85**</td>
<td>0.24ns</td>
<td>-0.98**</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Alb</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-0.12ns</td>
<td>0.28ns</td>
<td>-0.10ns</td>
<td>0.19ns</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TP</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-0.76**</td>
<td>0.24ns</td>
<td>-0.93**</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ca++</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-0.22ns</td>
<td>0.88***</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>P</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-0.24**</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cl</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-0.24**</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Ns= Not significant; **P<0.01, ***P<0.001
Vol= Volume; Con= Concentration; Mot= Motility; K+= Potassium; Na+= Sodium; TC= Total cholesterol; Alb= Albumin; Ca+= Calcium; P= Phosphorus; Cl= Chlorine

Table 4: Principal components weights for semen traits of chickens

<table>
<thead>
<tr>
<th>Traits</th>
<th>PC1</th>
<th>PC2</th>
<th>Communality</th>
</tr>
</thead>
<tbody>
<tr>
<td>Volume</td>
<td>0.994</td>
<td>-0.85</td>
<td>0.995</td>
</tr>
<tr>
<td>Concentration</td>
<td>0.883</td>
<td>0.357</td>
<td>0.907</td>
</tr>
<tr>
<td>pH</td>
<td>0.450</td>
<td>0.879</td>
<td>0.975</td>
</tr>
<tr>
<td>Glucose</td>
<td>0.995</td>
<td>0.016</td>
<td>0.991</td>
</tr>
<tr>
<td>Potassium</td>
<td>-0.489</td>
<td>0.839</td>
<td>0.944</td>
</tr>
<tr>
<td>Sodium</td>
<td>-0.195</td>
<td>-0.954</td>
<td>0.948</td>
</tr>
<tr>
<td>Total cholesterol</td>
<td>-0.994</td>
<td>0.071</td>
<td>0.993</td>
</tr>
<tr>
<td>Albumin</td>
<td>0.193</td>
<td>0.454</td>
<td>0.243</td>
</tr>
<tr>
<td>Total protein</td>
<td>-0.958</td>
<td>0.201</td>
<td>0.958</td>
</tr>
<tr>
<td>Calcium</td>
<td>0.884</td>
<td>0.404</td>
<td>0.945</td>
</tr>
<tr>
<td>Phosphorus</td>
<td>-0.278</td>
<td>-0.081</td>
<td>0.084</td>
</tr>
<tr>
<td>Chlorine</td>
<td>0.985</td>
<td>0.050</td>
<td>0.973</td>
</tr>
<tr>
<td>Eigenvalues</td>
<td>7.187</td>
<td>3.749</td>
<td></td>
</tr>
<tr>
<td>% of total variance</td>
<td>55.058</td>
<td>29.069</td>
<td></td>
</tr>
</tbody>
</table>
Table 5: Stepwise multiple regression of body weight on original body measurements and on their principal component (PC) factor scores in chickens.

<table>
<thead>
<tr>
<th>Model</th>
<th>Explanatory variables (predictors)</th>
<th>Intercept</th>
<th>Regression coefficient</th>
<th>Standard error</th>
<th>R²</th>
</tr>
</thead>
<tbody>
<tr>
<td>(i)</td>
<td>Original measurements as explanatory variables</td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>1</td>
<td>Potassium</td>
<td>48.89</td>
<td>3.06</td>
<td>2.10</td>
<td>0.20</td>
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<tr>
<td>2</td>
<td>Potassium</td>
<td>256.40</td>
<td>3.43</td>
<td>1.79</td>
<td>0.48</td>
</tr>
<tr>
<td>3</td>
<td>Calcium</td>
<td>-27.67</td>
<td>13.05</td>
<td></td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>Total Cholesterol</td>
<td>-1.69</td>
<td>1.34</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Sodium</td>
<td>0.39</td>
<td>1.11</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(ii)</td>
<td>Orthogonal traits as explanatory variables</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>PC1</td>
<td>96.02</td>
<td>4.07</td>
<td>2.55</td>
<td>0.22</td>
</tr>
<tr>
<td>2</td>
<td>PC1</td>
<td>95.87</td>
<td>4.11</td>
<td>2.71</td>
<td>0.22</td>
</tr>
<tr>
<td></td>
<td>PC2</td>
<td>0.54</td>
<td>3.83</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Discussion

Assessment of semen characteristic is important indicator of the reproductive potential of breeding cocks. 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). This study aims at probing and comparing the reproductive details of Nigerian indigenous cocks through their semen characterization in the humid tropical zone. While variation in semen physical characteristics could be mainly attributed to genetic differences in the present study, heat tolerance traits and seminal plasma biochemical parameters are more likely to be affected by environmental influences. Each breed has a unique major gene that determines how they manage heat load in the tropics and thereby should be expected to have variation in their productivity. However, the frizzle feather and naked neck chicken exhibited better adaptability in terms of heat-tolerant traits when compared with the normal feather chicken and therefore should be expected to have improved performance since the higher the heat stress index, the higher the severity of the heat stress experienced by the bird (Oladimeji et al. 1993). High temperature in addition to changes in RT could lead to disturbance of thermal balance of birds (Lin et al. 2005). According to Dana et al. (2010), adaptation to the production environment was found as an important attribute of village chickens in Ethiopia. Kohler-Rollefson et al. (2009) reported that, in the event of climate change, ability to maintain production and cope with thermal stress could give the indigenous breeds an edge over their exotic counterparts.

Considerable variation was observed in the physical and plasma biochemical parameters of the three genetic groups. 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. In this regards Bah et al. (2001) reported 0.28ml for breeding cocks of the Sahel while 0.7ml was documented by Tuncer et al. (2006) for Denizli cocks. It also compares well with the range of 0.37-0.73ml reported by Peters et al. (2008b) for the same Nigeria indigenous strains in the humid tropics. Variations arising from semen concentration and sperm motility for normal feathered, naked neck and frizzled feathered cocks could be attributed to their different genetic background and their natural tendencies as also noted by Peters et al. (2008b) who reported differences between strain in terms of semen volume, concentration and motility of Nigerian indigenous cocks. Values obtained for semen pH compared well with the range reported by Etches (1998).
Adeleke et al. (2012) ranked them together in terms of reproductive performance. Naked neck chicken consistently had the least reproductive performance. According to the reports, percentage fertility and hatchability was consistently lower among eggs sired by the naked neck genetic group with increased embryonic mortality up to 10-21.2% which resulted in reduced hatchability in pure strain (Adeleke et al., 2012; Crawford 1997; Horst 1980; Rauen 1985; Merat 1986; Peters et al. 2008a). Peters et al. (2005; 2008a) reported that purebred mating involving naked neck birds produce higher number of infertile eggs and high percentage of dead in shell and submitted that naked neck gene is likely to be lethal in homozygote dominant form. Since heat stress may affect osmotic equilibrium and ionic channels that are significant in the interplay between spermatozoa, its environment and the egg (Darazi et al. 1999) we decided to study the effect of our tropical ambient temperature on the seminal biochemical parameters of our indigenous cock semen.

Higher fertility and hatchability from different reports for the Normal feathered chicken from better semen concentration and sperm motility which was also observed from this study could be attributed to their better performance in terms of seminal plasma Ca++, K+, Cl- and albumin. This is in agreement with the report that Ca++ is very important to spermatozoa flagella while its interruption impede viability and fertility. Activation of motility also depends on both intra and extra cellular Ca++, extracellular Na+ and K+.

Heat stressed birds showed lower level of these minerals (Barna et al. 1998; Karaca et al. 2001; 2002ab). Although the Normal feathered cocks showed higher RT and HSI in this study, it is likely that the normal feathered gene has made them to be better adapted to the tropics and give better performance in terms of reproductive performance.

In terms of motility, naked neck followed the normal feathered cocks and as a matter of fact had the lowest values of heat tolerance traits in this study. The cocks should be expected to follow the normal feathered chicken in performance; however, the reverse was the case as the strain has been reported to be low in fertility and hatchability. This could be explained from our result as the strain had the lowest level of Ca++, Cl- and Glu which are very crucial for spermatozoa performance. Although they had the highest level of TP, P and TC, it has been reported that the higher the level of cholesterol, the lower the fertility (Ansal 1985). A-Daraji et al. (2011) reviewed 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 of rabbits may inhibit fertilization by inhibiting membrane fusion during acrosome reaction as a result of its incorporation into lipid bilayers. These could also explain the high level of dead in shell and higher number of infertile eggs reported apart from homozygote dominant form being lethal as reported by Peters et al. (2005).

Frizzle feathered chicken were reported to have positive adaptive gene influence of the frizzle feather trait which significantly affect thermoregulatory ability (Horst 1989). They follow normal feathered chicken in terms of reproductive performance which according to our study could be due to the fact that they follow them in terms of essential seminal plasma ions. The genotype had the same level of K+ and Cl- with the normal feathered chicken and had the highest value of Na+, Glu and with the least value of TC. The frizzled feather strain cocks had higher values for glucose than the other strains examined. This is an indication that the frizzled gene had better glucose to withstand hot or humid environment than others (Obioha 1992). Glucose is also essential for immunoprotein in the female tract and early gamete interactions and by providing energy as a substrate for the production of ATP.
According to Baker (2009), analysis of domestic animal resource is very crucial for management decision making on conservation and for fitness-adaptability traits improvement. Therefore, the present findings open possibility for associating difference in reproductive performance of indigenous chicken of humid tropical Nigeria with difference in the seminal plasma biochemical parameters.

Strong relationship existed between semen characteristics and seminal plasma biochemical parameters. Positive correlations of traits suggest that the traits are under the same gene action (pleiotrophy) and therefore could provide a basis for genetic manipulation and improvement of native stock. The estimates of correlation between motility and seminal plasma ions in the present study are however in contrast with the report of Karaca et al. (2000b) except for K⁺ and Ca++. He however stated that change in sperm and plasma Ca++, Cl⁻, and Na⁺ may be related to heat stress infertility.

The traits (volume, concentration, motility, pH, seminal plasma Glu, Ca++, Cl⁻, K⁺) associated with PC1 and PC2 are by themselves rather good estimator of semen quality. Findings are in accordance with the report of Shahin and Hassain (2000), Kashiwamura et al. (2001) and Yakubu et al. (2009), that the first factor accounted for the largest variance in rabbits, horse and chicken respectively. PC1 and PC2 could be interesting for the purpose of evaluation and comparison of animals for breeding and selection purposes. Since the correlation between principal components is zero, the selection of animals for any principal component will not cause correlated response in terms of other principal components (Pinto et al. 2006). PCA could also be used to identify independent and informative variables thereby eliminating redundant information for the purpose of reducing costs in genetic programmes (Yamaki et al. 2006). The result of prediction of motility from seminal plasma biochemical parameters indicates that motility could be predicted with a fair degree of accuracy from these ions. However, the use of seminal plasma ion measurements should be treated with caution due to multicolinearity which has been shown to be associated with unstable regression estimates (Ibe 1989; Malau-Aduli et al. 2004) which may lead to unreliable predictions and which justifies the use of principal component. However, the result of prediction using PC was low, given a reduction in the amount of variance explained. These findings provide useful insight into the contribution of seminal ions into semen motility but more studies should be conducted with increase in sample size to further validate the results.

Conclusion

The results of this study showed that the variations in the genetic groups of the strains of birds used significantly affected semen characteristics of the chicken. As the cocks were subjected to the same environmental conditions, the difference observed in their reproductive performance could be attributed to their genetic differences which determine their interaction with the environmental conditions with subsequent effect on physical and biochemical semen parameters. These finally affect the reproductive performance of the strains in the humid tropics. The implication of our results is that reproductive ability of the birds does not depend on their ability to adapt to the tropical environmental alone but also on the intrinsic factors of genotype and possibly genetic by environment interaction. The findings with global initiatives could assist in long-term genetic improvement programme for chicken production in the tropics. Selecting the most important traits that explain the major part of the total phenotypic variability between chicken genotypes is a purposeful step towards selection.

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