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Genetic parameter and sexual dimorphism on Immune response traits of Nigerian Indigenous chickens at different ages

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Abstract

The study evaluated correlations and sex and age effect on Immune Response traits in . Blood samples from 100 birds (50 per genotype) were collected for haematology at ages 4, 8, 12 from a population of 200 birds. Haematological parameters were: Pack cell volume (Pcv), White blood cell (Wbc), Red Blood Cell (Rbc), Eosinophils (Eos), Monocytes (Mono), Haemoglobin (Hb), Lymphocytes (Lym), Heterophils (Heter), Basophils (Bas). The project lasted for 12 weeks. CRD Factorial design was used. Data for haematology was subjected to ANOVA using Statistical Software Package (SAS ver, 9.2) for the statistical analysis. The result showed that there was significant ($p<0.05$) difference in some of the immune response traits measured at different ages in this study. The result showed that Pvc, Wbc, Lym, Heter, Eos, Mono were significantly ($p<0.05$) higher in mean values for males of Nigerian Indigenous Chickens at weeks 4 and 12 than females counterpart while at week 8. Wbc, Heter and Eos were significantly ($p<0.05$) higher in males than in females counterpart for the same traits measured. This indicated strong immunity in females than Males of Nigerian Indigenous Chickens as studied. The highest and positive association among immune response traits were found between Pcv and Hb, Pcv and Rbc, Hb and Rbc, Bas and Eos, Eos and Mono at different ages, respectively. Pcv could serve as a trait for selection of Nigeria local chicken for improved growth and productive life of birds through improved weight gain and heat-tolerance.

Wbc could serve as a marker indicating immunity and inherent resistance to tropical poultry diseases and parasites. Males of Nigerian Indigenous Chickens should be raised more by farmers for their high resistant antiviral activities regarding the harsh environmental conditions experienced in the tropical rain forest zone.

Keywords: Age; Antiviral Activity; Correlation; Immune Response Traits; Sex

1. Introduction

Nigeria's indigenous chickens, often referred to as "local" or "village" chickens, play a crucial role in the rural economy and culture. These chickens are typically raised in free-range systems, where they forage for food and are often left to roam freely around home steads. Local chickens are still the option for supplying the human population with protein and income. Due to their short generation interval and their products, chickens are preferred by majority of Nigerians, having the potentials of providing proteins and meats that can meet the demand of the citizen globally [1]. About 35% of the total household income in Nigeria particularly the Niger Delta region is from poultry and livestock production [2]. Each of these genes found in Nigerian Indigenous Chickens play significant role in the productive adaptability in this chicken. The hardy nature of the local chicken, as expressed in their resistance to certain diseases, high survival rate and their ability to thrive well under harsh conditions have helped the birds to be ecotype of tropics. Assessing the difference at the genotype level, Ibe [2] discovered that the Frizzled feather and naked neck matured than the Normal feather in the tropics, this could also result in genes being associated with sexual maturity. Adene [3] made it known that this difference may be due to the hormone testosterone in males between the sexes. Also, apart from this sex

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hormone, the stimulation and development of body parts and features that directly link to reproduction may be of important effect. The sexual dimorphism in chicken has been in favour of the males, as the aggressiveness of the males over the females especially when reared together always put the females at a disadvantage when feeding [4]. Hence, there is need to assess the sex effect on the Immune response traits of Nigerian Indigenous Chickens and their relationship among the immune response trait.

2. Materials and methods

2.1. Experimental site

The study was carried out at the Poultry unit, Teaching and Research Farm of the Department of Animal Science, University of Uyo. Akwa Ibom State. Uyo is found between latitude $4^0 3'$N and 45^0 3′S and longitude $7^0 3'$ and 45^0 35′W. The altitude of the area is 38m above the sea level and a mean rainfall of 200mm. The relative humidity is 79% with average Temperature of 28°C [5]. haematological analysis was carried out at the Biochemistry Laboratory University of Uyo, Uyo.

2.2. Experimental birds and management of the birds

A total of two hundred day old chicks of Nigerian indigenous origin which comprises of Normal feathered, frizzle feathered and naked-neck were purchased from Federal University of Agriculture, Abeokuta Poultry's Farm.

Birds were reared under intensive system of management. Chicks from each genetic group were allotted to the prepared brooders and brood for 4 weeks before transferred to rearing pen and terminated at 12 weeks of age. Birds were individually identified by wing tagging according to genotype by sex prior to transfer. The chicks were vaccinated against new castle disease on arrival and other medications were given routinely preventing disease outbreak. Lighting sources: stove and electricity bulbs were provided for heat. Feed and water were given *ad libitum.* The beddings were changed weekly if wet. Starter mash (CP- 23% and ME 3462kcal/kg) was provided *ad libitum* throughout the brooding stage. The standard management practice was strictly adhered to till termination at 12 weeks of the study. At rearing stage of 5 to 6 weeks grower ration (20% CP and 300 kcal/kg ME) from 6 to 12 weeks finisher ration (18% CP and 2850 kcal/kg ME) was fed to the chickens.

2.3. Data collection

2.3.1. Haematology

Data were collected for haematological analyses. Hematological parameters were Packed cell volume (PCV), White blood cell (WBC), Red blood cell (RBC), Haemoglobin (Hb). Blood sample was collected from the wing vein of 100 randomly selected chickens according to their sexes. 50 per sex and stored in the EDTA bottle for the haematological analysis. The blood samples collected were taken to Biochemistry Laboratory where analysis was done using different haematological Procedures according to each trait' technique. Haematological parameters were collected at different ages (4, 8 and 12 weeks) of the birds in the course of the experiment.

2.4. Statistical Analysis

The data for Immune response trait were subjected to General Linear Model (GLM) Analysis of Variance (ANOVA) of statistical software package [6]. Means were separated using (DMRT) Duncan Multiple Range Test at probability of 5% convenient level. The main effect Model:

$Y_{ijk} = \mu + S_i + A_j + R_k + SA_{ij} + \epsilon_{ijk}$
Y_{ijk} = Observed trait of interest in Nigerian local chickens
μ = population mean
S_i = fixed effect of i^{th} sex (male, female)
A_j = fixed effect of j^{th} age of Chicken (4, 8, 12 weeks)
SA_{ij} = interaction effect of i^{th} sex of j^{th} age
ϵ_{ijkl} = random residual error.

3. Results

3.1. Effect of Sex on the immune response traits of Nigerian indigenous chicken at all ages

There was significant ($p<0.05$) difference in sex effect on some immune response traits of Nigerian Indigenous Chickens measured at all ages in this study

3.1.1. Effect of Sex on the immune response traits of Nigerian indigenous chicken's genotype at week 4 of age.

The result in Table 1 indicated that males recorded significantly ($p<0.05$) higher mean values in Wbc (13.565±0.56g/dl), Heter (33.211±1.26 um), Lym (66.477±1.60k/uL) Eos (1.135±0.18 cell/McL) and Mono (1.178±0.26ml) than the females counterpart with 12.598±0.72 g/dl, 30.489±1.62 um, 64.318±2.04 k/uL, 0.893±0.23 cell/McL and 0.869±0.33ml for the same traits measured, respectively at week 4 of age while the other immune response traits showed no significant.

Table 1 Effects of Sex on the immune response traits of Nigerian indigenous chickens at week 4 of age.

Trait	Sex Male	female
Pcv(mL)	29.837±1.15	29.874±1.47
Hb(mL)	9.937±0.38	10.002±0.49
Rbc(m/mm^3)	2.479±0.10	2.582±0.12
Wbc(g/dl)	13.565±0.56^a	12.598±0.72^b
Heter(um)	33.211±1.26^a	30.2489±1.62^b
Lym(k/uL)	66.477±1.60^a	64.318±2.04^b
Eos(cell/McL)	1.135±0.18^a	0.893±0.23^b
Bas(um)	0.718±0.21	0.853±0.27
Mono(ml)	1.178±0.26^a	0.869±0.33^b

Pcv= Packed Cell Volume Mono= Monocytes Heter = Heterophils Wbc= White Blood Cell Hb= Haemoglobin, Bas= Basophils Eos= Eosinophils Lym= Lymphocytes Rbc= Red Blood Cell

3.1.2. Effects of Sex on the immune response traits of Nigerian indigenous chickens at week 8 of age.

The result in Table 2 indicated that males recorded significantly ($p<0.05$) higher mean values in Wbc (14.674±0.88g/dl), Heter (29.295±1.61um), Lym (70.604±1.94 k/uL) than the females counterpart with 12.205±1.12 g/dl, 27.434±2.05 um, 68.924±1.61k/uL for the same traits, respectively measured at 8 week of age while other immune response traits showed no significant ($p>0.05$) difference in their means.

Table 2 Effects of Sex on the immune response traits of Nigerian Indigenous chickens at week 8 of age

Trait	Sex Male	female
Pcv(mL)	29.804±0.93	30.732±1.19
Hb(mL)	9.881±0.31	10.216±0.40
Rbc(m/mm^3)	2.472±0.08	2.544±0.11
Wbc(g/dl)	14.674±0.88^a	12.205±1.12^b
Heter(um)	29.295±1.61^a	27.434±2.05^b
Lym(k/uL)	68.924±1.52^b	70.604±1.94 ^{a}
Eos(cell/McL)	0.511±0.11	0.548±0.15
Bas(um)	0.333±0.17	0.383±0.22
Mono(ml)	0.934±0.22	1.029±0.28

Pcv= Packed Cell Volume Mono= Monocytes Heter = Heterophils Wbc= White Blood Cell Hb= Haemoglobin, Bas= Basophils Eos= Eosinophils Lym= Lymphocytes Rbc= Red Blood Cell

3.1.3. Effects of Sex on the immune response traits of Nigerian indigenous chicken's genotype at week 12 of age.

The result in Table 3 indicated that males recorded significantly ($p<0.05$) higher mean values in Pcv(35.220±1.17ml), Wbc (17.097±0.96 g/dl), Heter (29.922±1.37 um), Lym (69.828±1.58 k/uL), Eos(2.422±0.22 cell/McL) and Mono (1.059±0.26 ml) than the females counterpart with 33.946±1.50 ml, 15.185±1.23 g/dl, 27.876±1.75 um, 67.912±1.24k/uL, 1.506±0.28 cell/McL and 0.830±0.34 ml for the same traits, respectively, measured at week 12 of age while the other immune response traits were not significantly ($p>0.05$) difference as measured in this study.

Table 3 Effects of Sex on the immune response traits of Nigerian indigenous chickens at week 12 of age

Traits	SEX Male	female
Pcv(mL)	35.220±1.17[a]	33.946±1.50[b]
Hb(mL)	11.764± 0.39	11.282±0.50
Rbc(m/mm^3)	2.987±0.11	2.821±0.14
Wbc(g/dl)	17.097±0.96[a]	15.185±1.23[b]
Heter(um)	29.922±1.37[a]	27.876±1.75[b]
Lym(k/uL)	69.828±1.58[a]	67.912±1.24[b]
Eos(cell/McL)	2.422±0.22[a]	1.506±0.28[b]
Bas(um)	0.705±0.22	0.961±0.28
Mono(ml)	1.059±0.26 [a]	0.830±0.34[b]

Pcv= Packed Cell Volume Mono= Monocytes Heter = Heterophils Wbc= White Blood Cell Hb= Haemoglobin, Bas= Basophils Eos= Eosinophils Lym= Lymphocytes Rbc= Red Blood Cell

3.2. Pearson correlation among immune response traits in Nigerian indigenous chickens at all ages.

The Correlation coefficients among immune response traits of Nigerian Indigenous chickens showed high positive correlations that range from very low to very high range.

3.2.1. Pearson correlation among immune response traits in Nigerian indigenous chickens at 4 weeks of age

Table 4 Pearson correlation among immune response traits in Nigerian indigenous chickens at 4 weeks of age

Trait	Pcv	Hb	Rbc	Wbc	Heter	Lym	Eos	Bas
Hb	0.992***							
Rbc	0.910***	0.926***						
Wbc	0.005	0.020	0.064					
Heter	0.002	0.013	-0.043	-0.122				
Lym	-0.015	-0.023	-0.043	0.154	0.154			
Eos	0.095	0.096	0.061	-0.080	-0.080	-0.165		
Bas	-0.042	-0.036	0.066	-0.005	-0.005	-0.058	0.289**	
Mono	0.028	0.009	0.006	-0.114	-0.227*	-0.006	0.452***	0.17

PcvV= Packed Cell Volume Mono= Monocytes Heter = Heterophils Wbc= White Blood Cell Hb= Haemoglobin, Bas= Basophils Eos= Eosinophils Lym= Lymphocytes Rbc= Red Blood Cell; ***= Significant($p<0.05$) correlation; **= highly ($p<0.01$) significant correlation; ***= very highly ($p<0.001$) significant correlation**

The Correlation coefficients among immune response traits of Nigerian Indigenous chickens ranged between -0.006 and 0.992 at week 4 which was a very low to very high range. The results showed very highly ($p<0.001$) significant correlation coefficient between Pcv (Hb(0.992) and RBC(0.910)), Rbc and Hb(0.926); and Eos (Bas(0.289) and Mono(0.452)), respectively at 4 weeks of age, and all were positively correlated. Nevertheless, there were positive relationship in most of the correlated traits, while some showed negative relationships among this traits studied. However, Heter recorded a negative significant correlation ($P<0.05$) between heterophils and monocytes (0.227). The

correlation coefficients ranged between -0.006 to 0.992 with the highest correlation coefficient recorded for Pcv and Hb with 0.992. The least relationship was between Mono and Lym (-0.006). The positive relationships were recorded between Pcv and Rbc (0.910), Hb and Pcv (0.992), Wbc and Pcv (0.005), Heter and Pcv(0.002), Eos and Pcv (0.095), and Mono and Pcv(0.028); Hb and Rbc (0.926), Hb and Wbc (0.020), Hb and Heter (0.013), Hb and Eos (0.096), and Hb and Mono (0.009); Rbc and Wbc (0.064), Rbc and Eos (0.061), Rbc and Bas (0.066), and Rbc and Mono (0.006); Heter and Lym (0.154); Eos and Bas (0.289), and Eos and Mono (0.452), Bas and Mono (0.17). However, other immune response traits relationships were negatively correlated at week 4 in Table 4.

3.2.2. Pearson correlation among immune response traits in Nigerian indigenous chickens at 8 weeks of age.

The results of the correlation among the haematological parameters measured showed very high significant ($p<0.001$) correlation coefficients between packed cell volume (haemoglobin, red blood cell) haemoglobin and red blood cell, heterophils and lymphocyte, eosinophils and monocytes respectively. However, Heterophils and monocytes were highly ($P<0.01$) significant but negatively correlation, while heterophils and eosinophils showed were but negatively correlated at week 8 of age. The relationship were positive in most of the correlated traits while some showed negative relationships. The correlation coefficient ranged from -0.006 to 0.972 with the highest correlation coefficient recorded for Pcv and Hb(0.972) while the least relationship was between Pcv and Lym(-0.006). The positive relationships were recorded between Pcv and Hb(0.972), Pcv and Rbc(0.877), Pcv and Wbc(0.062), Pcv and Heter(0.076), Pcv and Mono(0.037); Hb and Rbc(0.903), Hb and Wbc(0.015), Hb and Heter(0.099), Hb and Mono(0.067); Rbc and Wbc(0.032), Rbc and Heter(0.069), Rbc and Mono(0.075); Wbc and Heter(0.131); Wbc and Eos (0.032), Lym and Eos (0.119), Lym and Bas(0.073), Lym and Mono(0.097); Eos and Mono (0.512); Bas and Mono (0.078). However, other immune response traits relationships were negatively correlated at week 8 of the study, Table 5.

Table 5 Pearson correlation among immune response traits in Nigerian indigenous chickens at 8 weeks of age

T	Pcv	Hb	Rbc	Wbc	Heter	Lym	Eos	Bas(um)
Hb	0.972***							
Rbc	0.877***	0.903***						
Wbc	0.062	0.015	0.032					
Heter	0.076	0.099	0.069	0.131				
Lym	-0.006	-0.096	-0.067	-0.113	-0.976***			
Eos	-0.079	-0.047	-0.011	0.032	-0.246*	0.119		
Bas	-0.123	-0.130	-0.137	-0.027	-0.180	0.073	-0.124	
Mono	0.037	0.067	0.075	-0.167	-0.277**	0.097	0.512***	0.078

PcvV= Packed Cell Volume Mono= Monocytes Heter = Heterophils Wbc= White Blood Cell Hb= Haemoglobin, Bas= Basophils Eos= Eosinophils Lym= Lymphocytes Rbc= Red Blood Cell; ***= Significant (p<0.05) correlation; **= highly (p<0.01) significant correlation; ***= very highly (p<0.001) significant correlation**

3.2.3. Pearson correlation among immune response traits in Nigerian indigenous chickens at 12 weeks of age.

The results of correlation coefficient among the immune response traits measured showed very high ($p<0.001$) significant correlation between packed cell volume (haemoglobin, red blood cell) haemoglobin and red blood cell, heterophils and eosinophils; eosinophils and monocytes, respectively at weeks 12 of age.

Lymphocyte and eosinophils; heterophils and monocytes were negatively correlated but significant. Also, there was positively significant ($p<0.05$) correlation between packed cell volume and white blood cell, haemoglobin and white blood cell, and eosinophils and basophils respectively, while heterophils and basophils showed negative significant ($p<0.05$) correlation at week 12 of age in Table 6

There was positive relationship in most of the relationships while some showed negative relationships. The correlation coefficient ranged between 0.015 to 0.989 with the highest correlation coefficient recorded for Pcv and Hb (0.989) while the lowest was between Lym and Mono (0.015).

Positive relationships were recorded between Pcv and Hb (0.989), Pcv and Rbc(0.873), Pcv and Wbc (0.249), Pcv and Heter (0.145); Hb and Rbc(0.898), Hb and Wbc (0.260), Hb and Heter (0.154); Rbc and Wbc(0.188), Rbc and

Heter(0.123); Wbc and Heter(0.146); Lym and Mono (0.015); Eos and Bas (0.243); Eos and Mono (0.536); Bas and Mono (0.190). However, other immune response traits relationships were negatively correlated at week 12 of the study (Table 6)

Table 6 Pearson correlation among immune response traits in Nigerian Indigenous Chickens at 12 weeks of age

Trait	Pcv	Hb	Rbc	Wbc	Heter	Lym	Eos	Bas(um)
Hb	0.989***							
Rbc	0.873***	0.898***						
Wbc	0.249*	0.260*	0.188					
Heter	0.145	0.154	0.123	0.146				
Lym	-0.081	-0.100	-0.058	-0.094	-0.921			
Eos	-0.174	-0.151	-0.162	-0.160	-0.921***	-0.325**		
Bas	-0.115	-0.105	-0.069	-0.101	-0.219*	-0.024	0.243*	
Mono	-0.134	-0.119	-0.178	-0.101	-0.321**	0.015	0.536***	0.190

PcvV= Packed Cell Volume Mono= Monocytes Heter = Heterophils Wbc= White Blood Cell Hb= Haemoglobin, Bas= Basophils Eos= Eosinophils Lym= Lymphocytes Rbc= Red Blood Cell; ***= Significant (p<0.05) correlation; **= highly (p<0.01) significant correlation; ***= very highly (p<0.001) significant correlation**

4. Discussion

The effect of sex on immune response traits of Nigerian indigenous chicken's genotypes at different ages were presented in Tables 1 to .3. Sex significantly (p<0.05)influenced some of the immune response traits measured in this study at week 4, 8 and 12 weeks of ages in this study in Tables 1, 2 and 3, respectively. This is in tandem with Younis and Ghoneim [7] who observed sex differences in their study on the relationship between Sex hormones, Immune Response and Productive Performance of Broiler chicken. Significant effect observed in Eosinophils and Monocyte to show a significant (p<0.05) effect at different ages in this study Tables 4 to 6 is an indication that there was absence of stress reaction, steroid and some allergic reactions. This observation is in line with the report of Onyishi *et al.* [8] who reported a low value of the same parameters with increased in age.

According to Falcones *et al.* [9] lymphocytes which is a fraction of the white blood cells are natural killer cells and are able to kill cells of the body that do not display MHC class 1 molecules, or display stress markers such as MHC class 1 polypeptide-related sequence A (M/C-A). However,

the higher the lymphocytes, the better and stronger the immune system of the animal. These cells play both an immediate and delayed role in response to infection or inflammation. Increased numbers of lymphocytes are associated with most viral infections and some bacterial infections whereas decreased numbers of lymphocytes are characterized by steroid exposure, immunodeficiency and renal failures.

The significant value of heterophils recorded for Naked neck chicken in this study may be an indication that heterophils is part of WBC and have the phagocytic ability regardless of the presence or absence of antibodies. The observation in this work is not in tandem with the report of Opoola *et al*. [10] who reported of no significant difference in the quantity of heterophils produced in Nigerian indigenous chicken.

The test for WBC levels is very important to detect and measure the ability of the body to destroy diseases. A high level of WBC may be due to immunological challenge which may be largely informed by the associated inherent resistance to tropical diseases (Nosike *et al*. [11]. It is also noteworthy that the subsisting ecological condition support several vectors and parasites of economic significance. Nosike *et al.* [11] also observed the effect of genotype on WBC to be highly significant (p<0.05) in a gene of chickens.

The red blood cell of this study decreases as age of the chicken increases though not significant. This might results from the amount of feed consumed as there's effect of dietary feed on chickens, since young chicks consume less amount of feed compared to grown chickens [12]. Normal red blood cell production requires diet containing protein, iron, coppers, vitamin B2, B6, B12 and folic acid [12].

The Packed cell volume (PCV) obtained in this study were showing a rapid increase in values with age, and the highest PCV values significant, were observed at week 12 of age. This is not in line with the research findings of Addass *et al*. [13] and Nosike *et al.* [11] on indigenous chickens. This could be due to age group effect as observed on PCV. Increased PCV shows a better transportation and thus results in an increased primary and secondary polythelia. According to Adedibu *et al.* [14] Pcv value may be useful markers for predicting spermatozoa maturation in testes, sexual maturity and the onset of semen production. PCV is the volume of the red blood cells in the blood. Higher PCV values are usually due to a higher weight gain as well as reduction in heat load. Selection of Nigeria local chicken high PCV would therefore lead to a boost in the growth and productive life. PCV can serve as a trait for selection of Nigeria local chicken for improved growth and productive life of bird through improved weight gain and heat-tolerance [11].

5. Conclusion

It was observed that sex significantly influenced some of the immune response traits of Improved Nigerian indigenous chicken (INC) at different ages in this study. The result showed that Pvc, Wbc, Lym, Heter, Eos, Mono were significantly higher in mean values for females of Nigerian Indigenous Chickens at weeks 4, 8 and 12 than the males counterpart for the same traits measured. Wbc could serve as a marker indicating immunity and inherent resistance to tropical poultry diseases and parasites. The test for Wbc levels is very important to detect and measure the ability of the body to fight against diseases.

This indicated strong immunity in females than males of Nigerian Indigenous Chickens as studied. Correlation coefficient among the immune response traits ranged between -0.123 and 0.992. Indicating very low to very high associations. The highest and positive association among immune response traits were observed between Pcv and Hb, Pcv and Rbc, Hb and Rbc, Bas and Eos, Eos and Mono at different ages, respectively. Pcv can serve as a trait for selection of Nigeria local chicken for improved growth and productive life of birds through improved weight gain and heat-tolerance.

Recommendation

Males of Nigerian Indigenous Chickens should be raised more by farmers due to their high resistant to antiviral activities regarding the harsh environmental conditions experienced in the tropical zone.

Compliance with ethical standards

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Disclosure of conflict of interest

Authors declares that there is no conflict of interest with any financial, personal, or other relationships with other people or organization related to the material discussed in this Article.

Author declaration

All the authors have declared that this article is original article submitted for publication which has not been published anywhere in the world.

Statement of ethical approval

This research was conducted in accordance with ethical guidelines governing the use of animals and genetic materials in scientific research as stipulated by the Ethical Committee of Department of Animal Science, Faculty of Agriculture, University of Uyo, which is in line with Animal health care constitution.

Statement of informed Consent

Authors have given an approval and consent for the article to be submitted for publication.

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