Variation in helminth susceptibility of indigenous chickens in Kano and Jigawa States of Nigeria

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Abstract

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This study was carried out to identify the helminth parasites and burden associated with five indigenous chicken strains namely normal feathered Fulani ecotypes, naked neck, dwarf and frizzle kept under free-range system in six local government areas of Kano and Jigawa States. A total of 540 fresh faecal samples were collected from the gastrointestinal tract. Faecal egg count (quantitative examination) was carried out using the modified McMaster technique with flotation solution. All the helminths (cestodes and nematodes) were examined under light microscope and identified on the basis of helminthological keys. Worm count, EPG, Packed cell volume (PCV), total serum proteins and albumin were used to assess chicken susceptibility to worm infection. Significant differences were observed among the chicken strains for egg per gramme (EPG), cestodes and nematodes. The highest EPG (119.80) was observed in naked neck chickens, followed by frizzled (114.18), dwarf (62.86), normal feathered (30.80) and Fulani ecotypes (0.71). Frizzled chickens had higher mean cestode (7.96) count than the other strains. Dwarf chickens were significantly ($P < 0.05$) more susceptible to nematode infection. Higher EPG (60.55) and worm burden (cestode, 6.08) was found in males than females. The high gene flow and low differentiation between Fulani ecotype and normal feathered and, naked neck and frizzled buttressed the assertion that the similarity in EPG between them were because of genetic closeness. Susceptibility to worm infection in the study measured by the prevalence and EPG was therefore genetically controlled. There were generally negative and low correlations between egg and adult worm counts with haematological indices considered. Thus, blood parameters do vary in response to helminth infection and could be good measures of susceptibility.

Keywords: Helminth parasites, EPG, worm burden, gene flow, susceptibility

Variation de la susceptibilité à Helminthe des poulets autochtones dans les États de Kano et Jigawa du Nigéria

Résumé

Cette étude a été réalisée pour identifier les parasites de Helminthe et le fardeau associé à cinq souches de poulet autochtones, à savoir des écotypes Fulani à plumes normaux, un nuque nu, un nain et une frisée maintenus sous un système de liberté dans six zones gouvernementales locales des États de Kano et Jigawa. Un total de 540 échantillons fécaux frais ont été collectés à partir du tractus gastro-intestinal. Le nombre d'œufs fécaux (examen quantitatif) a été réalisé en utilisant la technique modifiée McMaster avec une solution de flottation. Tous les helminthes (cestodes et nématodes) ont été examinés sous un microscope léger et identifiés sur la base de clés helminthologiques. Le nombre de vers, EPG, volume de cellules emballé (VCE), les protéines sériques totales et l'albumine ont été utilisées pour évaluer la sensibilité au poulet à une infection de vers. Des différences significatives ont été observées parmi les souches de poulet pour l'œuf par gramme (EPG), des cestodes et des

nématodes. Le plus haut CEPG (119,80) a été observé dans des poulets de cou nu, suivis de frisanges (114,18), de nains (62,86), d'écotypes normaux à plumes (30,80) et de Fulani (0,71). Les poulets fronçettes avaient une cestode moyenne moyenne (7,96) comptant que les autres souches. Les poulets nains étaient significativement ($p < 0,05$) plus susceptibles d'infection nématode. L'CEPG (60,55) et la charge de ver supérieur (CESTOD, 6,08) ont été trouvés chez les poulets que les poules. Le flux de gènes élevé et la faible différenciation entre les écotypes Fulani et la normale à plumes normaux et à la nuque nue et à contreforts librizatés l'affirmation selon laquelle la similitude dans CEPG entre elles étaient à cause de la proximité génétique. La susceptibilité à une infection de vers dans l'étude mesurée par la prévalence et l'CEPG a donc été contrôlée génétiquement. Il y avait généralement des corrélations négatives et faibles entre le nombre de ver d'œufs et d'adultes avec des indices hématologiques considérés. Ainsi, les paramètres sanguins varient en réponse à l'infection d'helminthe et pourraient être de bonnes mesures de susceptibilité.

Mots-clés: Parasites helminthe, CEPG, Fardeau de ver, flux de gènes, susceptibilité

Introduction

Poultry production in tropical countries is based on the traditional scavenging system and chickens are the most important species (Lamidi *et al.*, 2014). The system provides meat, eggs, organic manure of high quality and is a reliable source of petty cash. The village chickens also fulfil a number of intangible benefits (Dube *et al.*, 2010). These include pest control, uses in traditional ceremonies and festivals (Muchadeyi *et al.*, 2004) because of few social and religious taboos against them (Mafu and Masika, 2003). Strategic increase in productivity of rural chickens will assist poverty alleviation; improve household – security, and protein intake of the rural communities, and in the long run curb urban migration (Dube *et al.*, 2010). Despite the importance of local chickens to humans, information on their genetic make-up and variability with respect to performance, adaptability, resistance to diseases are scarce (Alipanah *et al.*, 2011). In addition, very little strategies have been adopted geared at conservation. The efficiency of indigenous poultry farming in developing countries, and indeed in Nigeria however is affected by the presence of parasites (and other factors) (Permin and Hansen, 1998). High prevalence of helminth parasites have been reported in

free-range chickens in several developing countries (Abdelqadir *et al.*, 2008; Mukaratirwa and Hove, 2009). Iqbal *et al.* (2004) reported that helminths are frequently found in indigenous and exotic poultry and vary from 1 to 85% depending on geographical location. Parasitic diseases in chickens however, are often neglected because they are rarely lethal (Tesfaheywet *et al.*, 2012). Low gastrointestinal infections nonetheless contribute to sub-clinical conditions (Pinckney *et al.*, 2008), while heavy loads have devastating effect on growth, egg production and overall health (Pandey *et al.*, 1992; Permin *et al.*, 1997). Chickens pick up eggs of parasite by ingesting contaminated feed, water, litter and intermediate hosts such as snails, earth worms, or other insects (Butcher and Miles, 1992). Helminth parasites are commonly divided into three main groups; Nematodes (Roundworms), Cestodes (Tapeworms) and Trematodes (Fluke). Nematodes constitute the most important group in poultry both in number of species and extent of damage cause. The main genera include *Capillaria heterakis*, and *Acaridia* (Jordan and Pattison, 1996). The Cestode genera of importance are *Railletina* and *Hymenolepis* (Matur *et al.*, 2010). Trematode infections however are not very common in domestic chickens as

Prosthogonimus ntowi has been the only species reported from the belt of Ghana (Hodasi, 1978). Therefore, the present research was undertaken to identify the helminth parasites associated with the strains and determine whether worm burden is related to genotype and sex.

Materials and methods

Study area

The study was conducted in six local government areas (LGAs) of Kano and Jigawa States. Geographically, the LGA's are situated between latitude 12° 11'N and 12° 58'N, and longitude 7° 38'E and 8° 25'E (Olofin, 2007), at an average altitude of 485m above sea level. The area is bordered to the north by Niger Republic; by Katsina state, south; Plateau, southeast; Bauchi east and Kaduna southwest

Sample collection

Six markets were purposely selected to represent the different geographical areas of the states. Chicken gastrointestinal tracts were subsequently collected. Samples collection was carried out from December, 2014 to January, 2016. Appropriate sample size was determined using the following expression (Permin and Hansen, 1998):

$$n = \frac{Z^2 PQ}{L^2}$$

Where:

n = sample size,

Z = statistics for a level of confidence, usually stated at 1.96,

P = expected prevalence in the flock/population,

Q = 1 - P

L = required precision, e.g., the largest acceptable difference between the true and the estimated prevalence.

The helminth prevalence was not known; however, to maximize sample size, the expected prevalence was set at 90%; precision was 5%, and the confidence level, 95%. The samples were transferred into 200ml bottles containing 10% formalin

(Plate IX), labeled and transported to the laboratory for examination. All samples were qualitatively and quantitatively analyzed according to the techniques described by Soulsby (1982) and Urquhart *et al.* (1996), where direct and concentration (simple floatation and egg count) methods were carried out using salt as ideal media for cestode and nematode eggs count.

Faecal egg counts

Faecal egg count FEC (quantitative examination) was carried out using the modified McMaster technique with flotation solution as described by Urquhart *et al.* (1996). About 2.0g of faeces was weighed into a 30 mL polythene tube; 5 mL of water was added and thoroughly mixed with the aid of a glass rod to break it up. Water was added to the brim. The tube was covered and thoroughly shaken. The suspension was poured through a fine mesh sieve and the filtrate collected, agitated and centrifuged in a test tube at higher specific gravity for three minutes. It was allowed to bench for five minutes before counting to allow the eggs to rise in the floatation fluid. Both chambers of the slide were examined and numbers of eggs counted and, multiplied by 20 to determine egg per gramme (EPG)

Worm identification and counts

The gastrointestinal tract was opened longitudinally; visible helminths were collected followed by scraping off the mucosa and washing contents through a 63 µ aperture sieve. The remaining contents captured on the sieve were examined for the presence of helminth parasites under a stereo-microscope and observed helminths were collected and preserved in 70 % ethyl alcohol. For identification, the helminths were mounted in drops of lactic acid, and the scoleces and mature proglottids of cestodes were cleared with lactic acid, nematodes were cleared using lacto-phenol. All the helminths were then examined under light microscope and were identified on the

Variation in helminth susceptibility of indigenous chickens

basis of helminthological keys described by Dunn (1978), Soulsby (1982), Urquhart *et al.* (1987) and Permin and Hansen (1998).

Evaluation of resistance/susceptibility to worm infection

Worm count, EPG, packed cell volume (PCV), total serum proteins and albumin were used to assess chicken resistance to worm infection.

The PCV was determined according to the techniques described by Preston and Allonby (1978); Coles (1986). Blood samples (2 mL) were collected from the wing via disposable needle and syringe into tubes. Blood samples were centrifuged at a specific gravity for five minutes and count was determined using a Neubauer hemacytometer. The percentage of packed cells volume was recorded.

Total serum proteins and albumin were measured by colorimetric method using clinical chemistry kits (Human Company-Wiesbaden, Germany, 1985).

Statistical analysis

The statistical package for Social Sciences (SPSS, 2010) version 20.0 statistical software programme) was used for data analysis. The general linear model was employed to carry out analysis of variance on non-categorical measurements – worm count, EPG, PCV, TSP, ALB to test the effects of categorical factors. The model adopted was:

$$Y_{ijklm} = \mu + C_i + B_j + e_{ijklm}$$

Where:

Y_{ijklm} = is the observation on the $ijklm$ th chicken

μ = the overall mean

C_i = effect of i^{th} strain ($i = 1, 2, 3, 4, 5$)

B_j = effect of j^{th} sex ($j = 1, 2$)

e_{ijklm} = random error effects peculiar to each observation.

Investigating the genetic diversity of the Nigerian indigenous chickens using microsatellites markers

Sixty blood samples from the five

populations were taken from the wing vein. The DNA sample were extracted and genotyped at 8 microsatellite loci choosing from those used by the EU funded project “Development of Strategy and Application of Molecular Tools to Assess Biodiversity in Chicken Genetic Resources, EC Contract Number BIO4-CT98-0342” and based on FAO's recommendation as outlined in MoDAD report (FAO, 2004). Allele frequency, mean number of alleles, expected and observed heterozygosity for all loci and populations were estimated to investigate the genetic diversity within strains. Wrights fixation indices (F_{IT} , F_{ST} and F_{IS}) were calculated to define the genetic variance, the differentiation between populations. Pair-wise F_{ST} were estimated to calculate the Nei's genetic distance and draw a dendrogram.

Results

Effect of strain on worm burden

The effect of strain of chickens on egg and adult worm counts (mean intensities) in the study area are as presented (Table 1). There was significant ($p < 0.001$) EPG variation by strain. The highest EPG was observed in naked neck chickens (119.80), followed by frizzled (114.18), dwarf (62.86), normal feathered (30.80) and Fulani ecotypes (0.71). Strain variation with regards to cestode was also significant ($P < 0.001$). Frizzled chickens had higher mean cestode count (7.96) than the normal feathered, naked neck, Fulani ecotype and the dwarf with values of 5.37, 4.51, 3.38 and 2.00 respectively. Dwarf chickens were significantly ($P < 0.05$) more susceptible to nematode infection (3.29) as compared to 1.56, 1.20, 0.32 and 0.31 for normal feathered, Fulani ecotype, naked neck and frizzled birds respectively. Similarly, the effect of strain on mixed infection was significant ($P < 0.05$). Frizzled chickens had the highest counts (8.27), followed by normal feathered, dwarf, naked neck and

Fulani ecotype with values of 6.92, 5.29, 4.83 and 4.57 respectively.

Effect of sex

The effect of sex on worm burden (EPG) is depicted in Table 1. There was higher ($P<0.01$) egg count in males (60.55) than females (24.21). For adult mean worm

count, cestode ($P<0.01$) and mixed infection ($P<0.05$) values were higher in males (6.08 and 7.50) than females (4.27 and 5.65) respectively. However, no significant variation was observed between nematode adult worm burdens by sex of birds.

Table 1: Worm burden by chicken strain and sex

Variable	Number	EPG Mean $\pm$ se	Cestode Mean $\pm$ se	Nematode Mean $\pm$ se	Mixed infection Mean $\pm$ se
Strain		***	**	*	*
Normal feather	377	30.80 $\pm$ 5.40 ^b	5.37 $\pm$ 0.35 ^{ab}	1.56 $\pm$ 0.27 ^{ab}	6.92 $\pm$ 0.44 ^{ab}
Fulani ecotype	56	0.71 $\pm$ 0.70 ^c	3.38 $\pm$ 0.46 ^b	1.20 $\pm$ 0.50 ^b	4.57 $\pm$ 0.66 ^b
Naked neck	41	119.80 $\pm$ 33.82 ^a	4.51 $\pm$ 0.75 ^b	0.32 $\pm$ 0.23 ^b	4.83 $\pm$ 0.76 ^{ab}
Dwarf	21	62.86 $\pm$ 30.18 ^{ab}	2.00 $\pm$ 0.53 ^c	3.29 $\pm$ 1.42 ^a	5.29 $\pm$ 1.46 ^{ab}
Frizzled	45	114.18 $\pm$ 45.32 ^a	7.96 $\pm$ 1.75 ^a	0.31 $\pm$ 0.19 ^b	8.27 $\pm$ 1.83 ^a
Sex		**	**	NS	**
Male	274	60.55 $\pm$ 10.85 ^a	6.08 $\pm$ 0.46 ^a	1.43 $\pm$ 0.28	7.50 $\pm$ 0.55 ^a
Females	266	24.21 $\pm$ 5.48 ^b	4.27 $\pm$ 0.36 ^b	1.37 $\pm$ 0.30	5.65 $\pm$ 0.46 ^b

Mean $\pm$ se with different superscript within column between each parameter differ significantly, * = $P<0.05$,

** = $P<0.01$, *** = $P<0.001$, EPG = egg per gramme

The PCV values did not vary significantly by strain. The RBC values in chickens varied significantly ($P<0.05$) with strain. Highest RBC value was recorded in Fulani ecotype (4.67), followed by naked neck (4.00), dwarf (3.91), normal feathered (3.53) and frizzled (2.89) (Table 2). Values of the leucocyte counts (neutrophyl, monocytes, eosinophyl, basophyl and lymphocytes) are presented in Table 2. Neutrophyl values did not vary significantly with strain. However significant ($P<0.05$) difference was observed between strains in the number of monocytes. The highest counts (1.33%) was in normal feathered chickens followed by Fulani ecotype (0.71%), naked neck

(0.67%), fizzled (0.35%) and dwarf (0.20%). Percentage lymphocytes distribution showed that dwarf had significantly ($P<0.05$) higher lymphocytes count (51.0%) than the normal (47.17%), Fulani ecotype (39.29%), naked neck (44.67%) and frizzled (43.0%) chickens. There was significant ($P<0.05$) difference among strains with respect to plasma albumin. The highest plasma albumin value was recorded in dwarfs (2.13g/dL), followed by frizzled (1.92g/dL), Fulani ecotype (1.61g/dL), normal (1.39g/dL) and naked neck (1.38g/dL). The values of plasma protein differed ($p<0.01$) among the chicken strains from 4.50 g/dL in naked neck to 6.61 g/dL in dwarf chicken.

Table 2: Mean haematological parameters of free-range chickens

Variable	PCV (%)	RBC $\times 10^{12}/L$	Neutrophyl%	Monocytes %	Eosinophyl %	Basophyle %	Lymphocyte %	Plasma Alb (g/dl)	Plasma Prot (g/dl)
Strain	Ns	*	Ns	**	Ns	Ns	*	**	**
Normal	34.44 $\pm$ 1.30	3.53 $\pm$ 0.32 ^{ab}	44.08 $\pm$ 2.04	1.33 $\pm$ 0.19 ^a	4.78 $\pm$ 0.44	1.39 $\pm$ 0.29	47.17 $\pm$ 1.81 ^a ^b	1.39 $\pm$ 0.08 ^c	5.56 $\pm$ 0.18 ^{ab}
feather									
Fulani	36.88 $\pm$ 2.29	4.67 $\pm$ 0.52 ^a	53.12 $\pm$ 3.83	0.71 $\pm$ 0.22 ^{ab}	5.00 $\pm$ 1.33	1.88 $\pm$ 0.52	39.29 $\pm$ 3.78 ^b	1.61 $\pm$ 0.18 ^b	5.05 $\pm$ 0.29 ^b
ecotype									
Naked neck	34.89 $\pm$ 1.74	4.00 $\pm$ 0.37 ^{ab}	49.22 $\pm$ 3.84	0.67 $\pm$ 0.24 ^{ab}	3.22 $\pm$ 0.57	2.22 $\pm$ 0.86	44.67 $\pm$ 3.51 ^{ab}	1.38 $\pm$ 0.22 ^c	4.50 $\pm$ 0.65 ^c
Dwarf	35.50 $\pm$ 1.73	3.91 $\pm$ 0.75 ^{ab}	42.80 $\pm$ 4.42	0.20 $\pm$ 0.13 ^b	3.30 $\pm$ 0.63	2.70 $\pm$ 0.97	51.00 $\pm$ 4.71 ^a	2.13 $\pm$ 0.25 ^a	6.61 $\pm$ 0.54 ^c
Frizzle	33.18 $\pm$ 1.35	2.89 $\pm$ 0.33 ^b	49.82 $\pm$ 3.67	0.35 $\pm$ 0.15 ^b	4.53 $\pm$ 0.49	2.88 $\pm$ 0.83	43.00 $\pm$ 3.24 ^{ab}	1.92 $\pm$ 0.20 ^{ab}	6.05 $\pm$ 0.50 ^{ab}
Sex	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns
Male	34.58 $\pm$ 0.97	3.89 $\pm$ 0.27	47.62 $\pm$ 2.05	0.94 $\pm$ 0.15	4.60 $\pm$ 0.50	1.92 $\pm$ 0.31	46.43 $\pm$ 2.08	1.59 $\pm$ 0.10	5.17 $\pm$ 0.22
Females	35.19 $\pm$ 1.29	3.47 $\pm$ 0.30	46.81 $\pm$ 2.09	0.68 $\pm$ 0.15	4.24 $\pm$ 0.41	2.11 $\pm$ 0.46	45.04 $\pm$ 1.38	1.64 $\pm$ 0.10	6.12 $\pm$ 0.23
Mean $\pm$ se with different superscript within column between each parameter differ significantly, RBC= Red blood cells, PVC= Packed cell volume, alb= albumin, prot= protein									

Relationships among some helminth and haematological parameters

The Pearson correlation coefficients among chickens' helminth and haematological indices are presented in Table 3. There were generally negative and low correlations

between egg and adult worm counts with haematological indices considered. However, most correlation values between egg (EPG) and adult worm counts and, within the latter were generally positive, significant ($P < 0.01$) and fairly high.

Table 3: Correlations between helminth parasites and blood parameters of free-range chickens

	EPG	Nematode count	Cestode count	MI	PCV	RBC	Eosinophyl	Albumin
EPG	-							
Nematode	0.474**	-						
Cestode	0.274**	0.012	-					
MI	0.466**	0.554**	0.797**	-				
PCV	-0.052	0.006	-0.152	-0.063	-			
RBC	-0.158	-0.001	-0.074	0.038	0.109	-		
Eosinophyl	-0.161	-0.127	0.083	-0.015	-0.096	0.077	-	
Albumin	0.060	-0.035	0.131	0.107	-0.095	0.061	0.029	-
Protein	-0.079	-0.203	0.001	-0.095	-0.028	-0.125	0.048	0.455**

EPG = egg per gram, RBC= Red blood cells, PVC= Packed cell volume, MI = Mixed Infection, **= $P < 0.05$, **= $P < 0.01$, others not significant.

Table 4: Genetic Differentiation (F_{ST} below diagonal) and Gene flow (above diagonal) of indigenous chicken strains

Populations	Normal feathered	Fulani ecotype.	Naked neck	Dwarf	Frizzled
Normal feathered	****	5.356	4.670	5.752	5.473
Fulani ecotype	0.047	****	5.312	5.503	3.554
Naked neck	0.053	0.041	****	4.205	8.789
Dwarf	0.049	0.054	0.052	****	3.678
Frizzled	0.042	0.064	0.029	0.058	****

Figure 1. Dendrogram of relationships among 5 strains of chicken using Nei's (1978) genetic distance and Neighbour-joining methods.

Discussions

Effect of strain on worm count

Variation in helminth infection has been established in this study by comparing the 5 strains of chickens. Based on EPG, Fulani ecotype and normal feathered chickens had the lowest worm burden. As already stated in our previous report, these two strains were genetically closer than others. Gradation in EPG for the other strains was also according to the genetic distance study (Dendrogram figure 1). The naked neck and frizzled chickens had the highest EPG. These two strains were also genetically closer at the other end of dendrogram. Furthermore, the high gene flow and low F_{ST} between Fulani ecotype and normal feathered and, naked neck and frizzled (Table 4) has in addition buttressed the assertion that the similarity in EPG between them were because of genetic closeness. Thus it could be firmly stated that susceptibility to worm burden in the study measured by EPG was genetically controlled. This is consistent with the findings of many researchers that immunity and susceptibility to diseases in chickens may be genetically determined (Bumstead and Barrow, 1993; Okoye and Aba-Adu Lugba, 1998; Poulsen *et al.*, 1998). Though, acquired immunity could play a part. Investigation into the immune system has shown that chickens have the ability to expel worms (Permin *et al.*, 1999). Variations in adult worm burden (cestodes, nematodes and mixed infection) by strain though significant, did not readily correspond to the genetic strain relationships. That there was significant strain variation in worm burden in this study agrees with Urquhart *et al.* (1988). It has been established that the Fulani ecotype is heavier than the other Nigerian chicken strains and as in the present study usually exhibits lower worm burden (Sonaiya, 2013). This is in agreement with Ackert *et al.* (1935) that heavy chicken breeds are less

susceptible than light ones. It however contradicts the report that larger host harbour more gastrointestinal helminths because they consume more food and therefore are more likely to ingest higher quantity of infective stages (Nalubambe *et al.*, 2015). In view of this contradiction, it can be stated that the mechanism of difference in strain susceptibility is not yet well known.

Furthermore, understanding host immune response to helminths is also essential in functional characterization of strains (Dominik *et al.*, 2010; Matika *et al.*, 2011; Silva *et al.*, 2012).

Effect of sex

Overall, males had higher worm burden than females. This corroborates the findings of Urquhart *et al.* (1988) and Klein (2000a, b), and that it may be due to sex steroid which modulate several aspect of host immunity. More so, Todd and Hollingwilt (1951) reported that female chickens had lower worm burden and concluded that sex hormones might be an important factor in the development of parasite population.

Packed cell volume

The haematocrit or packed cell volume is a component of the blood parameters. Its evaluation is essential for determining the state of health of a bird. The pattern of PCV distribution in the present study was similar among chicken strains and fall within the normal physiological range reported by Jain (1993).

The PCV was slightly higher in Fulani ecotype chickens which exhibited the least helminth infection. It has been reported that PCV is somewhat higher in resistant than susceptible ecotypes, but not in all the cases (Gray, 1995). In a study with rams, Gruner *et al.* (2002) found that resistant rams had higher burden than susceptible ones and stated that if faecal egg counts were ambiguous, then adult worm count and PCV could be helpful in evaluating host susceptibility to helminth parasite.

Red blood cells

Red blood cells (erythrocytes) serve as carrier of haemoglobin. Haemoglobin reacts with blood to form oxyhaemoglobin (Johnston and Morris, 1996). The variation in the RBC values in the present study is in agreement with the report of Duke, (1955) and Islam *et al.* (2004) that haematological values are influenced by breed. The values recorded in the present study are within the physiological range of $2.2 - 4.0 \times 10^{12}/L$ (Schalm, 1975).

In a study conducted by Peters *et al.* (2011) on variation in haematological parameters of Nigerian native chickens; normal-feathered birds had higher mean values compared to frizzle and naked neck strain. This differs from the present study where Fulani ecotype had the highest mean value compared with the other strains. Similarly, Agaie and Uko (1998); Islam *et al.* (2004); Chineke *et al.* (2006) and Peters *et al.* (2011) observed some strain differences. However, the observed low values of RBC count in some strains of chicken in the present study could be as a result of higher worm burden associated with them. Aade *et al.* (2011) found in chickens infected with cestode showed lower RBC values and related immune status. Similarly, Misra *et al.* (1996) also observed decreased haemoglobin and RBC counts in lambs infected with nematode.

Differential leucocytes count

The major function of the WBC and other differentials counts is to defend the body by phagocytosis against invasion by foreign organisms and to produce, transport and distribute antibodies in immune response (Etim *et al.*, 2014). Animals with high WBC and leucocytes counts are capable of generating high volume of antibodies during phagocytosis and have high degree of resistance to disease (Soeten *et al.*, 2013) and enhance adaptability to local environment (Kabir *et al.*, 2011; Isaac *et al.*, 2013). The differential leucocytes reported

in the present study are lower than the values reported by Islam *et al.* (2004). However, they are similar to the values reported by Oyewale (1987) in Nigerian local fowls.

Higher monocytes counts might have been recorded in normal feathered chickens in this study due to more infection by nematodes. Dhanalakshmi *et al.* (2002) reported that sheep infected with *Strongyles* (nematode) had significantly higher monocytes counts. Monocytes (which are agranulocytes of WBC) values were within the normal range, from 0.06 to 5.0% for chickens (Riddell, 2011). Islam *et al.* (2004) reported monocyte value of 3.42% for local chickens of Bangladesh, 6% in Kashmir, Bank, (1974) and 1 to 2% in Nigeria (Afolabi *et al.*, 2011).

There was no significant difference among the five strains of chickens relative to eosinophyl. Different reports however exist on level of eosinophils in resistant and susceptible breeds. Pernthaner *et al.* (1996) reported that there was higher circulating eosinophyl in resistant than susceptible animals. But, Buddle *et al.* (1992) reported that decrease level of eosinophil is associated with susceptibility to nematode infection. Eosinophils are multifunctional cells. Their primary function is considered to be defence against organisms that are too large to be phagocytosed, particularly parasitic helminths. Butterworth (1984), hypothesized that the primary function of eosinophils in defending hosts against infection by relatively large organisms is based on the accumulation of observations that eosinophils degranulate and can kill helminths *in vitro* in the presence of antibody and/or complement; they move from the blood and aggregate in the locality of helminths *in vivo*; large numbers of eosinophils are often observed in close association with both intact and damaged helminths *in vivo*; and they degranulate in the vicinity of, or on to the surfaces of,

helminths *in vivo*. Eosinophils are involved in animal pathological response and are inevitably present in large numbers in inflammatory lesions associated with helminth infections or allergic conditions. However, Woolaston *et al.* (1996) stated that the role of eosinophyl is unclear and may vary in host parasite system (Meusen and Barlie, 2000). Following infection, eosinophyl move from the bone marrow through blood to the tissue and only a quantity goes into gastrointestinal tract and instant in regulating growth of parasites (Henderson and Stear, 2006). They further reported that aallergic or parasitic conditions may show increased numbers of eosinophils. Lymphocytes are usually the most numerous and frequent WBC type followed by heterophiles, eosinophils and monocytes. They are the key elements in the production of immunity. Dwarf chickens had significantly higher lymphocytes than the other four groups. This contradicts the reports of Dhanalakshmi *et al.* (2002) that animals infected with nematode had lower lymphocyte counts, since in the present study, dwarf chickens had higher nematode burden than others. The lymphocyte values in this study are within the normal physiological range of 20 – 50% for healthy chickens (Duke, 1955). However, the relatively high values of lymphocytes and neutrophils counts in this study are consistent with the findings of Sturkie (1965) and Afolabi *et al.* (2011). However, available information indicates that haematological values of avian species are significantly influenced by poultry diseases including parasites (Juranova *et al.*, 2001).

Plasma albumin and protein

Albumin is the largest single protein fraction in healthy birds synthesized in the liver and serves as the major reservoir (Sahu, 2014). In this study, plasma albumin variations were similar to that of total plasma protein and were low. They are lower than the normal values 6.0 - 8.5 and

2.9 - 3.9 g/dl respectively (Cole, 1986). Gadre *et al.* (2008) reported that low albumin and total proteins are usually due to parasites and haemorrhage that may lead to anaemia. In particular, albumin is a negative acute phase protein and its concentration decreases during inflammation and parasite infection (Majabi, 2000; Stockham and Scott, 2002). In humans, children with helminth infection were found to have low blood albumin and total proteins (Bashir *et al.*, 2012). Northop (1987) showed that albumin concentration increased significantly after anthelmintics treatments in children infected with parasites. More so, increase in albumin concentration is associated with dehydration and decrease occurs with lower synthesis (in chronic liver diseases, dietary protein deficiency, renal disease, and gastrointestinal disease (Margaret and Wissan, 2001). Although the mechanism of reduction in the albumin and protein concentration in the blood is not well known, it has been suggested that intestinal parasitism increases albumin catabolism (Tanwar and Mishra, 2001). This is probably due to reduced energy intake during parasitism thus proteins are mobilized to supply the needed amount (Crompton and Nesheim, 2002). Plasma proteins are essential components of the blood and play a crucial role in maintaining homeostasis (Okwonkwo *et al.*, 2011). Changes in plasma protein are common clinical symptoms of gastrointestinal parasitism (Harvey, 1975). That total plasma protein in dwarf and frizzle chickens were highest, but Fulani ecotype and naked neck had least values is contrary to Ladokun *et al.* (2008) and Ibrahim *et al.* (2012). They reported higher protein levels in normal feathered than other strains of local chickens. This according to them may be due to infection differences. Yon Brand (1973) reported that changes in the serum protein levels are characteristic features of parasitic infections. He also observed that

variations in serum protein depended on stages or severity of an infection and the host species. Plasma protein loss had also been associated with helminth infection in sheep and goat (Yacob *et al.*, 2008). A slight decline in the serum protein level during ascariasis in children had also been reported (Venkatachalam and Patwardhan, 1953). Sadun and Willams (1986) also reported decreased serum protein level in albino rats during experimental schistosomiasis. On the other hand, increased serum protein level was observed by Khadoon and Ansari (1979) in buffalo calves with *Setaria cervi* infection.

Relationships among some helminth and haematological parameters

Haematological parameters have been used to evaluate worm infection (Nalumbamba *et al.*, 2015). There were few positive correlations between worm burden and haematological parameters eg plasma albumin and cestode, or total worm count. This shows that there might have been increases in their haematological parameters in response to infection probably to combat the situation. On the other hand, negative correlations e.g. between EPG and PCV, RBC, eosinophyl, plasma protein and, albumin might have been due to loss of blood parameters due to the combating situations. The negative correlations between PCV, RBC and eosinophyl with EPG have been documented (Albers *et al.*, 1990; Baker *et al.*, 2003). They further concluded that it is ideal to select for higher PCV and decreased EPG. More so, Yadav *et al.* (1993) observed that PCV level was significantly lower in relation to higher faecal egg count and worm burden of *H. contortus* in sheep. They also reported that damage of gastrointestinal mucosa caused by gastrointestinal nematodes resulted in leakage of plasma protein causing hypoproteinemia and loss of body weight. Similarly, significant decline in PCV and

increase in total neutrophil and eosinophil count were observed in sheep experimentally infected with *H. contortus* (Saddiqi, 2010). Furthermore, Dominik (2005) reported that gastro-intestinal helminth infection in sheep was incriminated as the cause of declined level of total erythrocyte count, packed cell volume (PCV) and lymphocyte count. They further reported that gastro-intestinal parasites caused development of immature cells which led to formation of weak cell junctions in the host body. Some studies have however shown that helminth burden did not have much effect on plasma albumin probably due to the ability of the host to balance the relationship between their nutritional requirement and the losses through helminth infestation (Moore and Hopkins, 2009; Cripps *et al.*, 2014).

A prominent feature of this study was the complete absence of trematodes. This is in conformity with the works of Fabiyi, (1972) and Yoriyo *et al.* (2005) which did not find any trematodes among birds examined in different parts of northern Nigerian. It is concluded that chicken strains used responded differently to the helminth infection, which may be attributed to their differences in their natural or acquired resistance. The Fulani ecotype was the least susceptible to helminth parasites followed by normal feathered chickens. Therefore, the Normal feathered and Fulani ecotype have been showed to be less susceptible to worm and should be utilized in any effort to improved resistance to helminth infection in chickens.

Conclusion

It is concluded that chicken strains used responded differently to the helminth infection, which may be attributed to their differences in their natural or acquired resistance. The Fulani ecotype was the least susceptible to helminth parasites followed by normal feathered chickens. The normal

feathered and Fulani ecotypes were the most closely related strains, though the five strains had common ancestry.

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Variation in helminth susceptibility of indigenous chickens

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