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Genetic and phenotypic correlations among coccidiosis-tolerant traits in Nigerian indigenous chickens

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***Target Audience:** Animal scientists; Farmers; Geneticists; Animal breeders*

Abstract

Poultry meat is mostly consumed by Nigerians because of its low fat content. However, coccidiosis Eimeria poses a serious threat to the livestock industry. The identification and understanding of the association of genes encoding traits that can affect the outcome of the disease can then be applied to increase their frequency in a population through selective breeding. A total of 143 birds of three genotypes of Nigerian indigenous chickens which included Normal feather, Naked neck and Frizzle feather were used for the experiment. Body weight gain (WG), lesion score (LS), feacal oocysts count (FOC) and some haematological parameters which included RBC (Red Blood Cell), WBC (White Blood Cell), LYMPH (Lymphocytes), NEUT (Neutrophils), BAS (Basophils), MON (Monocytes) and EOS (Eosinophil) of control and inoculated birds (with Eimeria tenella) were estimated by nested analysis of variance procedure of SAS to derive paternal half-sib estimate of variance and covariance components used to estimate heritability, genetic and phenotypic correlations (h^2, r_g and r_p respectively) between all pairs of traits. The h^2 increased moderately in haematological traits with EOS having the highest value (0.88 ± 0.03) followed by WBC (0.75 ± 0.15). The highest h^2 was observed in WG (0.95 ± 0.21). The r_g was high for WBC pairs of traits and the highest r_g was observed in WBC/LS (0.96 ± 0.31) suggesting that the level of WBC can affect the severity of the infection as indicated by the LS level. The r_p was high for WBC- correlated traits which are in agreement with their high r_g with the highest being WBC/LYMPH (0.91 ± 0.30), which suggested that the susceptibility of birds changes in relation to the prevalence of these cells.

***Keywords:** coccidiosis, tolerant traits, heritability, genetic correlation, Nigeria.*

Description of Problem

Genotype and phenotype are markedly different in that genotypes represent the genetic make-up of organisms (1). An individual's genotype, because it includes all of the various alleles carried, determines the range of traits possible (e.g. an individual's potential to be afflicted with a particular disease). In contrast to the possibilities contained within the genotype, the phenotype reflects the manifest expression of those possibilities (potentialities). A trait is a distinct variant of a phenotypic character of an organism that may be inherited, be environmentally determined or be a combination of the two (2).

Genetic correlation is the association between sets of genes which influence two or more traits on the same individual or the correlation between the breeding values of

the traits (3). It can be caused by pleiotropism, linkage of genes and different intensities or direction or selection in the non-interbreeding sub-groups of population.

Coccidiosis is a parasitic disease caused by many species of *Eimeria*. About 9 species of *Eimeria* are known to infect poultry (4). The two most important species of *Eimeria* that affect poultry mostly are *Eimeria tenella* which causes caecal feaces and *Eimeria necatrix* which causes bloody intestinal coccidiosis (5). Coccidiosis is one of the most important causes of economic losses in the poultry industry in Nigeria. This is due to the increasing parasite resistant to anti-coccidial drugs currently used in commercial poultry production. Nigerian indigenous chickens are well-adapted to the local climate of Nigeria and also have low fat content; hence, there is the need to utilize the natural immune response of Nigerian indigenous chickens as a method of disease control. One way to counteract coccidiosis is the identification of traits that are related to immunity which may affect the outcome of the disease. The understanding of the association and inheritance of these genes can then be applied to increase their frequency in a population through selective breeding. Therefore, the aim of the study was to determine the genetic and phenotypic correlations of coccidiosis-tolerant traits in Nigerian indigenous chickens.

Materials and methods

Experimental site

The research was carried out in a standard farm in Osiele, Abeokuta. Fertile eggs of each genotype of Nigerian indigenous chickens were collected from The Directorate of University Farms (DUFARM), FUNAAB and the pedigrees of the fertile eggs were recorded based on the past records from DUFARM.

Experimental birds

Three genotypes of Nigerian indigenous chickens based on feather type were used for this experiment, namely, the Frizzle feather, the Normal and the Naked neck chickens which were generated through artificial insemination.

Artificial Insemination Procedure

Semen collection

The sires were trained three weeks before the commencement of semen by continuous massaging. Semen was collected from the males of the three genotypes based on feather types using eppendorf tube through the massaging method. Massaging of the cock was done from the caudal end to the pelvic areas of the male chickens through opening the vent of the chicken. The process of artificial insemination lasted throughout the period of egg collection so as to ensure the collection of fertile eggs and was done after two consecutive days routinely.

Insemination

Pressure was applied at the caudal end of the female birds to open the cloaca, and insemination into the left opening (oviduct) of the female's cloaca was done using Pasteur pipette.

Mating design:

Sire	×	**Dam**
Frizzle	×	Frizzle
Normal	×	Normal
Naked neck	×	Naked neck

Egg collection and Hatchery

The eggs were collected twice daily – morning and evening and 100 eggs were collected for each of the genotypes and a total of 300 eggs were collected. They were checked for deformities before being stored. The eggs were stored for a maximum of 5

days in the hatchery before being set and incubated.

A total of 143 birds of three genotypes of Nigerian indigenous chickens which included Normal feather, Naked neck and Frizzle feather were used for the experiment (84 Normal feather, 40 Naked neck and 20 Frizzle).

Feeding and management

The chicks were brooded for three weeks where they were fed a standard diet (State the crude protein and metabolizable energy of the feed). Feed and water were given *ad libitum*

The daily routine management practices were strictly adhered to.

Inoculation of the birds

The *Eimeria tenella* used for inoculation was obtained from the National Veterinary Research institute, Jos, in Plateau State, Nigeria. The birds of each of the genotypes were divided into two equal numbers, which are 42 for Normal feather, 20 for naked neck and 10 for Frizzle feather. Half of the population was inoculated with *Eimeria tenella* at the third week while the remaining half served as control. All the birds were raised in battery cages to avoid contamination with their feaces. The birds were wing-tagged for identification before inoculation. The birds were inoculated by challenging them with *Eimeria tenella* orally using syringes at the third week. A dose of 1×10^5 oocysts per ml of *Eimeria tenella* was given to each of the birds orally. After inoculation, data were collected.

Data collection

The following data were collected:

Body weight gain (WG)

The weights of the control and the inoculated birds were taken before and after inoculation and at the end of two weeks post-challenge in comparison to those of the control. All measurements were taken in the morning before the birds were fed. Body weight was taken using a sensitive weighing balance with sensitivity of 0.01g and body weight gain (WG) was calculated for both the inoculated and the control birds.

Feacal oocysts count (FOC)

The feacal samples of the birds were taken before and after inoculation. The feacal samples were taken at days 0, 3, 6, 9, 12 and 15 and the number of oocysts per gram of feaces was determined using the McMaster method with the aid of a microscope as previously described by (6).

Lesion score (LS)

Lesion score was determined using procedures as previously described by (7). By using the scale developed by (8), the severity of infection was rated.

Blood sample collection

Blood samples were randomly collected from each of the genotypes of the inoculated and control birds before and after inoculation at the 3rd week from the wing vein of the birds using a 2ml disposable syringe and directly the 2ml was transferred into labeled sample bottles containing Ethylene Diamine Tetra-acetic acid (EDTA) as anti-coagulant.

The non-coagulated blood was used to determine heamatological paramters such as Red Blood Cell (RBC) count, White Blood Cell (WBC) count, Packed Cell Volume (PCV), Eosinophil (EOS), Lymphocytes (LYMPH), Neutrophils (NEUT) and Basophils (BAS) using the haematocrit method.

Statistical analysis

The variance components (VARCOMP) used for the estimation of heritability of each

coccidiosis trait were obtained by Variance Components Procedure (PROC VARCOMP) of SAS (9) while the covariance components due to sire and progeny within sire for all pairs of traits obtained by Nested Procedure (PROC NESTED) of SAS (9). The genetic and phenotypic correlations between all pairs of traits based on sire components of variance and covariance using single parent design were computed as previously described by (10). The standard errors for heritability were computed using the formula given by (11) for unequal number of progeny per sire.

Results

Heritability (h^2 ± SE), genetic (r_g ± SE) and phenotypic (r_p ± SE) correlations of coccidiosis- tolerant traits

Table 1 shows the heritability of coccidiosis -tolerant traits. All the traits indicated high to moderately high heritability. WG had the highest heritability (0.95±0.21). Haematological parameters generally showed moderate to high heritability with the exception of LYMPH (0.06). LS had the lowest heritability of 0.09±0.03 among all the traits (Table 1).

Table 1: Heritability (h^2 ± SE) of coccidiosis- tolerant traits

TRAITS	PCV	WG	EOS	NEUT	LYMPH	MON	LS	FOC	WBC
PCV	0.43±0.02	_	_	_	_	_	_	_	_
WG	_	0.95±0.21	_	_	_	_	_	_	_
EOS	_	_	0.88±0.03	_	_	_	_	_	_
NEUT	_	_	_	0.22±0.09	_	_	_	_	_
LYM	_	_	_	_	0.06±0.05	_	_	_	_
MON	_	_	_	_	_	0.12±0.04	_	_	_
LS	_	_	_	_	_	_	0.09±0.03	_	_
FOC	_	_	_	_	_	_	_	0.27±0.03	_
WBC	_	_	_	_	_	_	_	_	0.75±0.15

The genetic correlation (r_g) (Table 2) between EOS/WG, NEUT/WG, LYMPH/WG, LYMPH/EOS, LS/EOS, LS/NEUT, LS/MON, FOC/LS, WBC/LS, WBC/ MON, WBC/LYMPH, WBC/ NEUT, WBC/EOS, WBC/WG and WBC/PCV were very high (Table 2). WBC/LYMPH had the highest genetic correlation value of 0.95±0.02 while low values of genetic correlation were observed in PCV/WG, EOS/WG, NEUT/EOS, LYMPH/ NEUT, MON/NEUT, MON/LYMPH, LS/PCV, LS/WG, FOC/PCV, FOC/WG, FOC/EOS, FOC/MON and WBC/FOC. The lowest genetic correlation value was observed in LS/WG (0.002±0.01).

PCV=Packed Cell Volume, RBC=Red Blood Cell, LYMPH=Lymphocyte, MON=Monocytes, NEUT=Neutrophils, WBC=White Blood Cell, EOS=Eosinophil, FOC= Feacal Oocysts Count, WG=Weight Gain, BAS=Basophil.

Table 3 shows the phenotypic correlations of coccidiosis tolerant- traits. The phenotypic correlations (r_p) between EOS/PCV, PCV/WG, NEUT/PCV, EOS/NEUT, LYMPH/EOS, LYMPH/ NEUT, MON/PCV, LS/NEUT, LS/EOS, FOC/PCV, FOC/EOS, MON/FOC, LS/FOC, WBC/WG, EOS/WBC, WBC/NEUT, WBC/LYMPH, WBC/MON, WBC/LS and WBC/FOC were relatively high (Table 3). (At which level was your correlation, 0.05, 0.01 or 0.001). The highest phenotypic

correlation was observed in WBC/LYMPH (0.91±0.30). Low phenotypic correlations were recorded in WG/EOS, WG/NEUT, WG/LYMPH, WG/MON, EOS/MON, NEUT/MON, LYMPH/MON, PCV/LS, WG/LS, LS/LYMPH, LS/MON, FOC/LYMPH, WG/FOC and WBC/PCV. The lowest phenotypic correlation was observed in WG/FOC with a phenotypic correlation of 0.002±0.01.

Table 2: Genetic correlation ($r_g \pm SE$) of coccidiosis- tolerant traits.

TRAITS	PCV	WG	EOS	NEUT	LYMPH	MON	LS	FOC	WBC
PCV		_	_	_	_	_	_	_	_
WG	0.55±0.02		_	_	_	_	_	_	_
EOS	0.33±0.09	0.01±0.08		_	_	_	_	_	_
NEUT	0.30±0.05	0.016±0.09	0.32±0.07		_	_	_	_	_
LYM	0.41±0.03	0.07±0.35	0.21±0.51	0.28±0.08		_	_	_	_
MON	0.18±0.31	0.03±0.07	0.015±0.04	0.03±0.01	0.04±0.09		_	_	_
LS	0.03±0.08	0.01±0.02	0.44±0.58	0.021±0.02	0.02±0.06	0.09±0.03		_	_
FOC	0.26±0.06	0.002±0.01	0.21±0.03	0.06±0.06	0.04±0.02	0.37±0.31	0.28±0.07		_
WBC	0.04±0.08	0.46±0.31	0.73±0.02	0.22±0.01	0.91±0.30	0.88±0.02	0.64±0.32	0.25±0.05	

Table 3: Phenotypic correlation ($r_p \pm SE$) of coccidiosis- tolerant traits

TRAIT	PCV	WG	EOS	NEUT	LYMPH	MON	LS	FOC	WBC
PCV	_	0.07±0.08	0.13±0.06	0.15±0.08	0.31±0.09	0.02±0.04	0.03±0.35	0.032±0.23	0.24±0.08
WG	_	_	0.02±0.12	0.34±0.12	0.23±0.05	0.02±0.03	0.002±0.23	0.08±0.23	0.23±0.42
EOS	_	_	_	0.02±0.22	0.19+0.01	0.01±0.95	0.29±0.12	0.01±1.21	0.67±0.03
NEUT	_	_	_	_	0.04±0.39	0.02±0.10	0.34±0.01	0.12±0.34	0.60±0.05
LYM	_	_	_	_	_	0.02±0.14	0.35±0.28	0.34±0.23	0.95±0.02
MON	_	_	_	_	_	_	0.17±0.09	0.012±0.66	0.92±0.03
LS	_	_	_	_	_	_	_	0.80±0.06	0.96±0.31
FOC	_	_	_	_	_	_	_	_	0.04±2.63
WBC	_	_	_	_	_	_	_	_	_

Values along the diagonal represent heritability **($h^2 \pm SE$),** values above the diagonal represent genetic correlation **($r_g \pm SE$)** while values below the diagonal represent phenotypic correlation **($r_p \pm SE$).**

Discussion

The heritabilities of haematological parameters (LYMPH, MON, EOS and WBC) obtained in this study were close to those observed by (12). The moderate heritability observed in most of the traits, especially those of the haematological traits could be due to genetic variations of the birds. Blood type is highly heritable from paternal inheritance and blood group of an individual is made up of those of both of its parents. The heritability of WG observed was the highest and closed to that observed by (13) but higher than that recorded by in chickens. Traits related to body weight gain (WG) generally showed high heritability. These obtained results are in range with previously reported heritability estimates. Body WG traits at various ages were reported to have an average heritability of 0.41 in a review of eighteen reports by (15). Similar findings were also reported by Buss (16) who observed heritability in the same range for body WG traits at different ages. This was due to the effect of common environment has been claimed by (15). Similar conclusions were reached by others researchers regarding the effect of common environment on the estimation of heritability (17).

The highest genetic correlation was observed in WBC/LYMPH while body WG correlated traits were also high, confirming the high heritability of body WG and blood parameters. Literature has very little regarding this aspect of correlation to

enhance comparison. The moderate to high correlations observed were similar to those observed by (12). Based on these results, some hypotheses on the pathogenesis of the disease have been proposed to explain the interactions between the parameters under the conditions in which the experiment was carried out. The haematological parameters LYMPH, NEUT, and RBC are generally associated with hydration levels and/or haemorrhage. The high and positive genetic correlations among all haematological traits in this study implies that they are all being controlled by similar genes (pleiotropism) and thus selection for any one of these traits would lead to positive changes in the other. This agrees with the report of (18) and supports the suggestion that both traits are essentially the same measure of growth and are thus under the influence of similar genes. Thus, the two traits can be regarded as the same trait in a selection programme.

The correlation between the lesion scores and FOC indicated indirectly that a larger amount of parasites in the ceacum causes a higher lesion score. (19) suggested that a greater increase of monocytes may be highly associated with the evacuation of a considerable amount of debris and this may also serve to assist in the defence against parasites in the tissue being affected. Based on the information above, it was found that an increased number of monocytes resulted in higher lesion and was more severe. In the (19) study, put an increase of the number of monocyte resulted to more pronounced at the end of the cycle of the parasite, after the peak period of oocysts production.

The moderate phenotypic correlations observed among other haematological correlated traits indicated that the increase in the number of these cells changes in relation to the degree of bird susceptibility and also suggest a partial replacement between lymphocytes and others in the development of immune response against *Eimeria tenella* (12,19). The moderate phenotypic correlations observed in the MON correlated traits like WG and MON suggest that an increase in body weight is associated with large amounts of monocytes (14). The set of factors that reflect in the bird performance is complex, involving many variables in host-parasite relationship. (20) reported that the pro-inflammatory cytokines, produced by several cell types, among which are the monocytes, are responsible for low growth in immunologically challenged animals. Although this result appears to give evidence of a pathogenic effect associated with this type of cell, under conditions in which this experiment was conducted. The phenotypic correlations observed among haematological traits were moderate and this suggests that both are affected by the same physiological pathways. The highest phenotypic correlation was observed among the WBC correlated traits and this suggests that WBC could be a good indicator for selective breeding which is in agreement with the high genetic correlation.

Conclusion and Application

1. Traits that showed high heritability are PCV, WG, WBC, NEUT, EOS and FOC.
2. Among these traits, MON/WBC showed the highest genetic correlation with appreciable phenotypic correlation.
3. Hence, tolerance to coccidiosis is highly heritable and these traits can be prioritized in selective breeding.

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