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Corresponding author: adeomoh@yahoo.com; 08138273174**ABSTRACT**

Haematological investigation provides the opportunity to clinically study the presence of several metabolites and other constituents in the body of animals and it plays a vital role in the physiological, nutritional and pathological status of an organism. A total of one-hundred and fifty (150)- day old Noiler chicken (70 –males and 80 females) were used to study the impacts of sex and age on hematology and serum metabolites in Noiler chicken. Data collected at 6th, 8th, and 10th week on hematological parameters and serum biochemistry were subjected to two-way analysis of variance and Pearson correlation. At 6th week, sex had no significant effect ($p>0.05$) in both hematology parameters and serum metabolites. At 8th week, sex significantly ($p<0.05$) affected Mean corpuscular volume (MCV), Mean corpuscular hemoglobin (MCH), albumin/globulin ratio (A/G) and chloride, with higher values recorded in female except in chloride. Sex effect was observed in white blood cell (WBC), total protein (TP) and AST at 10th week. In female virtually all the parameters considered were significantly ($p<0.05$) affected by age, except red blood cell (RBC), mean corpuscular haemoglobin concentration (MCHC), glucose, chloride, urea and creatinine. While in male, age effect was only observed in urea, MCH, MCV, WBC and platelet. The correlations among the hematology parameters ranged between -0.678 and 0.999 while among the metabolites was -0.958 and 0.586 for both sexes. It could be concluded that hemato-biochemical parameters considered are age-dependent in female more than male while sex effects were limited.

Keywords: noiler chicken, sex, age, serum metabolites, hematology.

INTRODUCTION

Noiler chicken breed was developed in Nigeria by Amo Farm Sieberer Hatchery Limited, popularly known as Amo Hatchery. It is a dual-purpose chicken breed; that is, for meat and egg production. The breed is a crossbred between local Nigerian chickens and commercial broiler breeds. This hybridization aims to harness the local chicken's resilience and adaptability to the African environment while improving its meat and egg production capabilities (Oladejo *et al.*, 2017). Sexual dimorphism is the phenomenon where males and females of the same species exhibit differences in physical and behavioral traits, often related to reproduction. It is a widespread phenomenon in the animal kingdom, occurring in a diverse range of species, including mammals, birds, reptiles, fish, and insects (Cox and John-Alder, 2007). One of the main sources of sexual dimorphism is the sex chromosomes, which determine the sex of an individual, the differential expression of sex-linked genes can result in sex-specific traits and phenotypes (Pallayova *et al.* 2019). Another major factor that contributes to sexual dimorphism is the sex hormones, which are chemical messengers that regulate various biological processes. The main sex hormones are testosterone in males and estrogen and progesterone in females. These hormones are produced by the gonads (testes and ovaries) and act on target tissues throughout the body. Sex hormones can affect the development, growth, maturation, and function of various organs and systems, such as the brain, bones, muscles, fat, skin, hair, and reproductive organs. Sex hormones can also modulate the behavior, cognition, emotion, and personality of individuals (Schulz *et al.*, 2019).

Blood indices are useful indicators of the response of animal to physiological changes due to environmental variations. They are also prognostic for pathological conditions (Ciesla, 2007). Haematological investigation provides the opportunity to clinically study the presence of several metabolites and other constituents in the body of animals and it plays a vital role in the physiological, nutritional and pathological status of an organism (Aderemi, 2004). It also

helps in distinguishing normal state from state of stress (Afolabi *et al.*, 2010). Odunsi *et al.* (1999) posited that dietary constituents are reflected in the blood picture of the livestock. Haematological and serum biochemical parameters are influenced by feed, medication, toxic compounds, infections, age and sex of the birds (Huff *et al.*, 2008). Due to scarcity of information of sex and age effects on haematological and serum biochemistry in Noiler chicken, the objective of this study was to investigate impacts of age and sex in hematological parameters and serum metabolite in Noiler chicken.

MATERIALS AND METHODS

The study was conducted at Poultry Unit, Teaching and Research Farm and Central Laboratory of Olusegun Agagu University of Science and Technology, Okitipupa, Ondo State, Nigeria. Okitipupa lies between latitude 6^o25 and 6^o46 N and Longitude 4^o35 and 4^o50 E within the tropical rainforest zone of Nigeria. A total of 150 day – old Noiler chicks (70 males and 80 females) were purchased from Amo Hatchery. The birds were reared in deep litter brooder pens and given a floor space of about 1.45 sq ft per bird as suggested by Adejoro, (2002). Each chick was wing - tagged and examined physically to ensure fitness and general body soundness. They were raised for a period of 10 weeks and fed *ad libitum* on a broiler starter diet (containing 24%CP and 2900kcal/Kg/ME) from hatching to 4th week of age followed by a finisher diet (21%CP and 2800kcal/Kg/ME) to 10th week of age. Water and feed were available *ad libitum* to the birds. All the necessary vaccinations and medication were administered accordingly. The blood samples were collected randomly from five (5) males and five (5) females at ages 6, 8 and 10 weeks through wing veins into EDTA bottles and plain bottles for hematological and serum biochemical analyses respectively. The samples were placed in a sample container labeled and carried in a flask (cooler) with ice packs and taken to Central laboratory of the University.

Determination of Serum Metabolites and Haematological Parameters

Packed cell volume (PCV), red blood cell (RBC), white blood cell (WBC), haemoglobin (Hb), total protein, albumin, globulin, uric acid, chloride and glucose were determined using methods as described by Dacie and Lewis (1991). The standard ratios of the mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) were calculated according to Jain (1986).

Statistical Analysis

Data collected on the hematological parameters and serum metabolites were subjected to two ways analysis of variance

(ANOVA) using SAS (v. 9.13) (SAS, 2004), where significant differences occurred in the means, the means were separated using Duncan Multiple Range Test. Correlations among the variables were determined by Pearson correlation.

RESULTS AND DISCUSSION

Haematological parameters in birds have been shown to be influenced by several factors among which are sex, age, nutrition season of the year, breeds (strains), body weight (Oyewale, 1990) and diurnal fluctuation and changes in physical and metabolic activities (Piccione *et al.*, 2005). The effect of sex on hematological parameters and serum metabolites of Noiler chicken are shown in Table 1.

Table 1: Sex Effect on Hematological parameters and Serum Biochemistry of Noiler chicken

VARIABLES	6 WEEKS		8 WEEKS		10 WEEKS	
	MALE	FEMALE	MALE	FEMALE	MALE	FEMALE
PCV	40.50 ^a ±0.87	39.50 ^a ±0.87	41.50 ^a ±1.44	43.50 ^a ±0.87	39.50 ^a ±0.87	38.00 ^a ±1.15
HB	13.50 ^a ±0.29	13.20 ^a ±0.29	13.85 ^a ±0.49	14.50 ^a ±0.29	13.20 ^a ±0.29	12.67 ^a ±0.38
RBC	4.45 ^a ±0.20	4.45 ^a ±0.09	4.45 ^a ±0.09	4.60 ^a ±0.29	4.50 ^a ±0.12	4.77 ^a ±0.09
WBC	11.70 ^a ±0.46	10.80 ^a ±0.58	13.25 ^a ±0.32	13.40 ^a ±0.35	12.07 ^b ±0.38	13.50 ^a ±0.23
PLATE	30.00 ^a ±0.23	30.20 ^a ±0.35	32.80 ^a ±0.52	35.00 ^a ±1.39	31.60 ^a ±1.09	30.10 ^a ±0.35
MCV	94.55 ^a ±4.53	85.65 ^a ±1.99	80.10 ^b ±2.71	95.30 ^a ±4.09	81.90 ^a ±0.17	79.97 ^a ±0.95
MCH	31.70 ^a ±1.62	28.50 ^a ±0.72	26.70 ^b ±0.92	31.80 ^a ±1.39	28.20 ^a ±0.52	26.60 ^a ±0.29
MCHC	33.30 ^a ±0.00	33.35 ^a ±0.03	33.30 ^a ±0.00	32.80 ^a ±0.29	33.27 ^a ±0.03	33.10 ^a ±0.06
TP	75.00 ^a ±0.00	72.50 ^a ±2.02	75.50 ^a ±2.02	74.50 ^a ±2.02	74.00 ^a ±0.58	68.50 ^b ±0.29
ACB	39.00 ^a ±0.58	39.50 ^a ±0.29	36.00 ^a ±1.15	37.50 ^a ±0.29	38.50 ^a ±2.02	40.00 ^a ±0.00
GLOB	36.00 ^a ±0.58	33.00 ^a ±2.31	39.50 ^a ±0.87	37.00 ^a ±1.73	35.50 ^a ±2.59	28.50 ^a ±0.29
AG	1.10 ^a ±0.00	1.25 ^a ±0.09	0.90 ^b ±0.00	1.05 ^a ±0.03	1.17 ^a ±0.15	1.40 ^a ±0.00
AST	9.00 ^a ±1.15	6.50 ^a ±0.87	9.50 ^a ±0.87	7.00 ^a ±1.15	6.50 ^b ±0.29	11.00 ^a ±0.00
ALT	8.00 ^a ±1.15	8.00 ^a ±0.58	9.50 ^a ±2.02	5.50 ^a ±0.87	9.50 ^a ±0.87	11.00 ^a ±0.58
ALP	16.00 ^a ±0.58	14.50 ^a ±0.87	16.50 ^a ±0.29	19.00 ^a ±1.15	17.00 ^a ±1.15	18.50 ^a ±1.44
GLUCOSE	5.50±0.35	5.10 ^a ±0.06	6.05 ^a ±0.61	5.15 ^a ±0.03	5.40 ^a ±0.40	4.77 ^a ±0.20
CHLORIDE	95.00 ^a ±0.00	93.50 ^a ±2.02	99.00 ^a ±0.58	93.35 ^a ±1.53	95.30 ^a ±2.14	96.17 ^a ±0.49
UREA	4.30 ^a ±0.23	4.65 ^a ±0.26	6.05 ^a ±0.61	4.85 ^a ±0.29	3.87 ^a ±0.55	4.47 ^a ±0.09
CREATININE	78.75 ^a ±0.20	75.75 ^a ±4.30	85.85 ^a ±9.15	72.40 ^a ±0.58	70.87 ^a ±11.92	80.27 ^a ±5.69

PCV – Packed Cell Volume; HB - Hemoglobin RBC – Red Blood Cell; WBC – White Blood Cell; plate - platelet;

a, b, means within a row with different superscripts are significantly different ($p < 0.05$).

MCV – Mean Corpuscular Volume; MCH – Mean Corpuscular Haemoglobin; MCHC – Mean Corpuscular Haemoglobin Concentration; TP - Total Protein; ACB - Albumin Cobalt Binding; GLOB - Globulin; AG - Albumin Globulin; AST - Asparate Amino Transferase; ALT- Alanine Amino Transferase; ALP - Alkaline Phosphate

At 6th week, sex had no significant ($p > 0.05$) effect on all the variables, this is contrary to findings from indigenous chickens in Sudan (Elagib & Ahamed, 2011), golden local quails (Muhammad, 2013) and free-range guinea fowl (King *et al.*, 2010). At week 8, sex effect was observed in MCH, MCV, AG and chloride, with higher estimates being observed in females, except in chloride. This is contrary to the reports of Gambo, *et al.*, (2021) who reported no significant effect of sex in local chicken in Maiduguri. At 10th week, significant, higher values of WBC (13.50) was observed in female, this is contrary to the report of Gambo, *et al.*, (2021); and AST (11.00) while lower value observed in TP (68.50). Generally, the discrepancies observed in sex effect on haematological parameters and serum metabolites with previous reseachers in this study could be attributed to differences in age of the birds and the strain of chicken.

Table 2: Age Effect on Hematology and Serum Biochemistry of Noiler chicken

PARAMETERS	FEMALE			MALE		
	6 weeks	8 weeks	10 weeks	6 weeks	8 weeks	10 weeks
PCV	39.50 ^b ±0.87	43.50 ^a ±0.87	38.00 ^b ±1.15	40.50 ^a ±0.87	41.50 ^a ±1.44	39.50 ^a ±0.87
HB	13.20 ^b ±0.29	14.50 ^a ±0.29	12.67 ^b ±0.38	13.50 ^a ±0.29	13.85 ^a ±0.49	13.20 ^a ±0.29
RBC	4.45 ^a ±0.09	4.60 ^a ±0.29	4.77 ^a ±0.09	4.45 ^a ±0.20	4.45 ^a ±0.09	4.50 ^a ±0.12
WBC	10.80 ^b ±0.58	13.40 ^a ±0.35	13.50 ^a ±0.23	11.70 ^b ±0.46	13.25 ^a ±0.32	12.07 ^{ab} ±0.38
PLATE	30.20 ^b ±0.35	35.00 ^a ±1.39	30.10 ^b ±0.35	30.00 ^b ±0.23	32.80 ^a ±0.52	31.60 ^{ab} ±1.09
MCV	85.65 ^b ±1.99	95.30 ^a ±4.09	79.97 ^b ±0.95	94.55 ^a ±4.53	80.10 ^b ±2.71	81.90 ^b ±0.17
MCH	28.55 ^b ±0.72	31.80 ^a ±1.39	26.60 ^b ±0.29	31.70 ^a ±1.62	26.70 ^b ±0.92	28.20 ^{ab} ±0.52
MCHC	33.35 ^a ±0.03	32.80 ^a ±0.29	33.10 ^a ±0.06	33.30 ^a ±0.00	33.30 ^a ±0.00	33.27 ^a ±0.03
TP	72.50 ^{ab} ±2.02	74.50 ^a ±2.02	68.50 ^b ±0.29	75.00 ^a ±0.00	75.50 ^a ±2.02	74.00 ^a ±0.58
ACB	39.50 ^a ±0.29	37.50 ^b ±0.29	40.00 ^a ±0.00	39.00 ^a ±0.58	36.00 ^a ±1.15	38.50 ^a ±2.02
GLOB	33.00 ^{ab} ±2.31	37.00 ^a ±1.73	28.50 ^b ±0.29	36.00 ^a ±0.58	39.50 ^a ±0.87	35.50 ^a ±2.59
AG	1.25 ^a ±0.09	1.05 ^b ±0.03	1.40 ^a ±0.00	1.10 ^a ±0.00	0.90 ^a ±0.00	1.17 ^a ±0.15
AST	6.50 ^b ±0.87	7.00 ^b ±1.15	11.00 ^a ±0.00	9.00 ^a ±1.15	9.50 ^a ±0.87	6.50 ^a ±0.29
ALT	8.00 ^b ±0.58	5.50 ^c ±0.87	11.00 ^a ±0.58	8.00 ^a ±1.15	9.50 ^a ±2.02	9.50 ^a ±0.87
ALP	14.50 ^b ±0.87	19.00 ^a ±1.15	18.50 ^{ab} ±1.44	16.00 ^a ±0.58	16.50 ^a ±0.29	17.00 ^a ±1.15
GLUCOSE	5.10 ^a ±0.06	5.15 ^a ±0.03	4.77 ^a ±0.20	5.50 ^a ±0.35	6.05 ^a ±0.61	5.40 ^a ±0.40
CHLORIDE	93.50 ^a ±0.2.02	93.35 ^a ±1.53	96.17 ^a ±0.49	95.00 ^a ±0.00	99.00 ^a ±0.58	95.30 ^a ±2.14
UREA	4.65 ^a ±0.26	4.85 ^a ±0.03	4.47 ^a ±0.09	4.30 ^a ±0.23	6.05 ^a ±0.61	3.87 ^b ±0.55
CREATININE	75.75 ^a ±4.30	72.40 ^a ±0.58	80.27 ^a ±5.69	78.75 ^a ±0.20	85.85 ^a ±9.15	70.87 ^a ±11.92

a,b,c means within a row with different superscripts are significantly different ($p < 0.05$).

MCV – Mean Corpuscular Volume; MCH – Mean Corpuscular Haemoglobin; MCHC – Mean Corpuscular Haemoglobin Concentration; TP - Total Protein; ACB - Albumin Cobalt Binding; GLOB - Globulin; AG - Albumin Globulin; AST - Aspartate Amino Transferase; ALT- Alanine Amino Transferase; ALP - Alkaline Phosphate

Age effect on hematology and serum metabolites is shown in Table 2. In female, age effect was significant ($p < 0.05$) in virtually all the parameters except in RBC, MCHC, glucose, chloride, urea and creatinine. This is contrary to the findings of Gambo *et al.*, (2021), who reported no significant difference in hematological parameters between young and adult free range village chickens in Maiduguri, Nigeria. Age effect was not significant ($p > 0.05$) in most of the variables in male except in WBC, platelet, MCV MCH and urea. This is in contrast to findings in broiler hybrids by Talabi *et al.* (2005) who reported that, with increasing age, the erythrocytic parameters (except MCV, MCH and MCHC) and leukocytic parameters (except heterophils and heterophil/lymphocyte ratio) were significantly increased. Similar observation was made by Muhammad (2013) where the haematological parameters increased in direct proportion to age in golden quails. This also agrees with an age related haematological findings in Kori Bustard (*Aedeotiskori*) chicks (Sarmour, 2006) where there was steady increase in their haematological parameters between 1-4 months of age although, this was followed by about 5 months of plateau, where no rise in haematological values were observed.

Table 3 illustrated the phenotypic correlations among the hematological parameters of Noiler. The correlation ranged between -0.45 and 0.99. However, most of the paired variables were positive and not significant except PCV/HB

(0.99), PCV/platelet (0.508), HB/platelet (0.507), WBC/MCHC (-0.515), MCHC/platelet (-0.678) and MCH/MCV (0.983). The strong positive correlation between HB and RBC suggests that as hemoglobin levels increase, the number of red blood cells also tends to increase. This differs with the findings of (Alodan and Mashaly, 1999) who reported negative correlation among hematological parameters. The positive correlation between RBC and Platelet suggests that there may be some relationship between the number of red blood cells and platelet count. However, the negative correlations with MCV and WBC indicate that as red blood cell count increases, mean corpuscular volume (MCV) and white blood cell count decrease. The positive correlation between WBC and HB suggests that there may be some connection between white blood cell count and hemoglobin levels. The negative correlation with MCH suggests that as white blood cell count increases, mean corpuscular hemoglobin (MCH) tends to decrease. Platelet count appears to have positive correlations with PCV, HB, and MCV. This is in agreement with the findings of (Talebi *et al.*, 2005) who reported high positive correlation among PCV, Hb, RBC, MCHC. This suggests that as platelet count increases, there may be an increase in packed cell volume, hemoglobin levels, and mean corpuscular volume. The very strong positive correlation between MCV and MCH (Talebi *et al.*, 2005) suggests that these two parameters tend to increase or decrease together.

The negative correlations with RBC and WBC indicate that as MCV increases, the number of red and white blood cells tends to decrease. MCHC has a positive correlation with

RBC, indicating that as MCHC levels rise, there is a tendency for an increase in the number of red blood cells.

Table 3: Phenotypic Correlation among Hematological indices in Noiler chicken

	PCV	HB	RBC	WBC	PLATE	MCV	MCH	MCHC
PCV								
HB	0.999***							
RBC	0.029 ^{ns}	0.022 ^{ns}						
WBC	0.103 ^{ns}	0.092 ^{ns}	0.307 ^{ns}					
PLATE	0.508*	0.507*	-0.159 ^{ns}	0.449 ^{ns}				
MCV	0.349 ^{ns}	0.376 ^{ns}	-0.348 ^{ns}	-0.062 ^{ns}	0.381 ^{ns}			
MCH	0.396 ^{ns}	0.389 ^{ns}	-0.336 ^{ns}	-0.115 ^{ns}	0.417 ^{ns}	0.983***		
MCHC	-0.097 ^{ns}	-0.090 ^{ns}	0.26 ^{ns}	-0.515*	-0.678**	-0.455 ^{ns}	-0.422 ^{ns}	

PCV – Packed Cell Volume; HB - Hemoglobin RBC – Red Blood Cell; WBC – White Blood Cell; plate - platelet; MCV – Mean Corpuscular Volume; MCH – Mean Corpuscular Haemoglobin; MCHC – Mean Corpuscular Haemoglobin Concentration.

Considerable variation of the correlations values among serum biochemistry parameters were found in Noiler chicken as shown in Table 4. TP positively correlates with other parameters, while ACB (-0.31), AG (-0.76), ALT (-0.49) and ALP (-0.26) negatively correlate with TP. ACB, negatively correlates with other parameters unlike TP while AG (0.82) and AST (0.22) positively correlate with it. Globulin also follow the trend which means it negatively correlate with the parameters except glucose (0.42), chloride (0.27), urea (0.58) and creatinine (0.39) which positively correlate. AG also correlate negatively with the other parameters except AST (0.05) and ALT (0.14) which positively correlate. AST positively correlates with all the parameters. ALT also positively correlate with other parameters except glucose (-0.41) and urea (-0.17). ALP also positively correlated with with all the parameters while glucose (-0.35) correlate negatively. Glucose, chloride, urea and creatinine positively correlate with all the parameters.

Table 4: Phenotypic Correlation among Serum Biochemistry of Noiler chicken

	TP	ACB	GLOB	AG	AST	ALT	ALP	GLUCOSE	CHLORIDE	UREA	CREATININE
TP	1.000										
ACB	-0.316	1.000									
GLOB	0.897	-0.702	1.000								
AG	-0.763	0.829	-0.958	1.000							
AST	0.021	0.229	-0.091	0.054	1.000						
ALT	-0.497	-0.152	-0.303	0.142	0.284	1.000					
ALP	-0.269	-0.210	-0.104	-0.008	0.063	0.019	1.000				
GLUCOSE	0.561	-0.004	0.422	-0.323	0.064	-0.415	-0.357	1.000			
CHLORIDE	0.110	-0.415	0.276	-0.339	0.358	0.223	0.209	0.131	1.000		
UREA	0.445	-0.543	0.586	-0.644	0.253	-0.173	0.096	0.441	0.481	1.000	
CREATININE	0.319	-0.335	0.396	-0.452	0.424	0.118	0.001	0.243	0.487	0.729	1.000

TP - Total Protein; ACB - Albumin Cobalt Binding; GLOB - Globulin; AG - Albumin Globulin; AST - Asparate Amino Transferase; ALT- Alanine Amino Transferase; ALP - Alkaline Phosphate

TP demonstrates a predominantly positive correlation with most other parameters, indicating that it tends to increase or decrease in tandem with them. This suggests that TP levels may be influenced by similar underlying factors. Conversely, TP exhibits negative correlations with ACB, AG, ALT, and ALP, suggesting that these parameters tend to decrease as TP levels increase, and vice versa. ACB shows predominantly negative correlations with other parameters, indicating that it often varies in the opposite direction to them. However, it positively correlates with AG and AST, suggesting that these two parameters tend to increase as ACB increases. Globulin follows a similar pattern to ACB, with negative correlations with most parameters. However, it positively correlates with glucose, chloride, urea, and creatinine. AST positively correlates with all the parameters, implying that it generally fluctuates in the same direction as other parameters. ALP also shows positive correlations with all parameters, suggesting a similar pattern of change. However, it negatively correlates with glucose, indicating that ALP levels may decrease as glucose levels increase.

CONCLUSION

Effect of sex on hematological parameters and serum metabolites in Noiler chicken was not significant at 6th week in all the parameters considered; significant effect on some parameters were observed at 8th week and less significant observed at 10th week. Age effect on most of the parameters was significant in female compared with male.

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