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Hematological Responses of Broiler Chickens to Vitamins A, C and E

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Abstract. Ninety day old chicks were used to investigate hematological responses of broiler chickens to vitamins A, C and E. Chicks were brooded and similarly managed for 4 weeks to fully adapt them to their environment. After this, animals were randomly assigned to 3 dietary treatments with 30 birds/treatment and 3 replicates of 10 birds/replicate as: T_1 (control diet, contained the vitamins at basal levels of 30mg/kg of diet), T_2 (diet 2, contained vitamins A and E at 100mg of vitamin A + 100mg of vitamin E/kg of diet) and T_3 (diet 3, contained vitamins C and E at 100mg of vitamin C + 100mg of vitamin E/kg of diet). The animals received their diets for 4 weeks. 9 birds from each treatment group consisting of 3 birds from each replicate were sacrificed and their blood collected for analyses: packed cell volume (PCV), haemoblobin (Hb), red blood cell (RBC), white blood cell (WBC) and their differentials: neutrophil (NEU), lymphocytes (LYM), monocytes (MON), oenophile (EON) and basophile (BAS). PCV of the T_1 animals was significantly ($P < 0.05$) lower than those of T_2 and T_3 groups. Hb concentration of T_1 group was significantly ($P < 0.05$) lower compared with T_2 and T_3 groups. No differences ($P > 0.05$) in the RBC amongst treatment groups. WBC was significantly ($P < 0.05$) higher with the T_3 group than T_1 and T_2. No differences ($P > 0.05$) in the NEU amongst treatments. The LYM of T_2 and T_3 groups had were significantly ($P < 0.05$) higher compared with the control (T_1). T_1 animals had significantly ($P < 0.05$) higher EON compared with animals of T_2 and T_3. The trend observed with the EON was also similar with the BAS. It was concluded that antioxidant vitamins A, C and E can improve the quality of lives for broiler chickens, especially vitamins C and E during their growth process.

Key words: Vitamins A, C and E, Hematological Parameters and Broiler Chickens

Introduction

Blood parameters are often used as one of the major indices of determining physiological, pathological and the nutritional statuses of a living organism, including farm animals such as poultry. To this point therefore, the changes seen in the constituents of blood when compared to the control values could be used to explain in part the metabolic state of an animal as well as the quality of the feed ingested by the animal (Babatunde *et al*., 1992). Furthermore, Ekenyem and Madubuike (2006) showed that haematological and serum chemical assays data could also be used to ascertain the physiological disposition of the animal to its nutrition.

The literature is awash with data demonstrating the impacts of nutrition on blood characteristics of different animal species. The data of Saita (1974) demonstrated that a diet with some levels of benzene when fed to animals induced leukemia, erythropenia, neutrophilia, lymphocytosis and alterations in blood platelets' morphologies. Ovuru and Ekweozor (2004) reported similar observations that the diets fed to rabbits in their studies resulted in decreased erythrocytes, platelets and packed cell volumes.

On the other hand, Johnson *et al.* (2019) showed that dietary vitamins improved PCV levels, Hb, RBC and WBC counts in the pig as well as neutrophils and lymphocytes. Furthermore, Okejim *et al.* (2020) observed that vitamin E ingestion ameliorated the

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negatives indices of the haematological properties of pigs initially fed diets contaminated with crude oil. They further demonstrated that the degree of amelioration was more when dietary vitamin E was given together with selenium. From these observations, it is not a gainsaying to state that nutrition has a great impact on the overall health and wellness of the animal and therefore forms the background to this current study. Therefore, the objectives of this study are to investigate the effects of dietary vitamins A, C and E on the PCV, Hb and RBC counts in broiler chickens and secondly to also study the effects of the vitamins on WBC counts as well as its differentials, namely: neutrophils, lymphocytes, oenophiles, monocytes and basophiles.

Materials and Methods

Animals

Ninety (90) *Agrited* day old chicks were acquired and used in this study. The animals on arrival at the Rivers State University Teaching and Research Farm were similarly reared and managed strictly for 4 weeks to ensure that they were fully adjusted to their new environment. After the four weeks of adaptation to their environment they were randomly assigned into three treatment groups of 30 birds/treatment group with 3 replications of 10 birds/replicate. To ensure that the animals were comfortable and also pathogen-free their pens were properly cleaned and disinfected before the birds' arrival. Their feeders and drinkers were also kept under high sanitary conditions. During the first 4 weeks' period of rearing the birds as earlier mentioned received similar management protocols in terms of the birds' safety, including the necessary medications were similarly provided. Animals were feed similar diets from day one through the end of the 4^{th} week. Water was provided *ad libitum* to all the animals throughout the study period.

Experimental Diets

Corn/soybean-based diet was used in the study. The diets fed to the animals during the last four weeks of the experimental period were isocaloric and isonitrogenous except that animals of group 2 (T_2) contained additional vitamins A and E while animals of group 3 (T_3) diet contained additional vitamins C and E, respectively. The control (T_1) diet contained these vitamins A, C and E at the basal levels only. The T_2 diet contained vitamins A and E at: (vitamin A 100mg + vitamin E 100mg/kg of diet) and T_3 diet contained vitamins C and E at: (vitamin C 100mg + vitamin E 100mg/kg of diet). The animals were fed these vitamin-based diets for 4 weeks (28 days).

Blood Sample Collection

At the end of the study period, nine (9) birds were bled for blood collection. Three birds were randomly collected from each replicate of the three treatment groups, respectively. The blood was collected from each bird into treated tubes with ethylene diamine tetra-acetic acid (EDTA) and immediately snap frozen for later haematological analyses.

Blood Analyses

Blood samples were analyzed by haematology auto-analyzer machine (BC-2300). Blood parameters analyzed for were: PCV, RBC count, Hb concentration, total and differential WBC counts of each treatment group, respectively.

Experimental Design and Statistical Analyses

The study was designed and carried out as a completely randomized design (CRD). Data obtained were subjected to analysis of variance (ANOVA) using general linear model

(GLM) procedure of SAS. Treatment means were compared using Bonferoni's test. The model was: $Y_{ij} = \mu + X_i + E_{ij}$, where Y_{ij} = individual observation of the treatment, μ = population mean, X_i = effect of the i^{th} treatment and E_{ij} = the error term. An *α-level* of *0.05* was used for all statistical comparisons to represent significance.

Results

The results of the PCV, Hb and RBC counts of broiler chickens that were fed different vitamins-based diets are shown in Table 1.

Table 1. Means of PCV, Hb and RBC of broiler chickens fed different vitamins-based diets

Item	Treatments			SEM	*P-value*
	T_1, n = 9	T_2, n = 9	T_3,n = 9		
PCV (%)	32^a	34^b	36^c	0.31	0.043
Hb (g/dl)	10.4^a	12.5^b	12.7^b	0.20	0.05
RBC (ul^3)	3.4	3.6	3.7	0.11	0.09

Means within each row with different superscript differ significantly (*P < 0.05*)

The PCV of the T_1 animals although it was within the normal range was significantly (*P < 0.05*) lower than those of T_2 and T_3 animal groups. The T_3 animal group had the highest PCV value that was significantly (*P < 0.05*) higher than the T_2 animal group. Overall, the T_2 group of animals had an intermediate PCV value amongst the three treatment groups. For the haemoglobin concentration, animals of the T_1 group demonstrated a significantly (*P < 0.05)* lower value compared with animals of the T_2 and T_3 groups that had similar (*P > 0.05*) Hb values. In terms of the RBC counts there were no differences (*P > 0.05*) amongst the three treatment groups. Nevertheless, there was a tendency for T_2 and T_3 having a numerical higher values, respectively.

The results of the WBC count and their differentials are shown in Table 2.

Table 2. WBC count and their differentials of broiler chickens fed different vitamins-based diets

Item	Treatments			SEM	*P-value*
	T_1 n = 9	T_2 n = 9	T_3 n = 9		
WBC (ul^3)	15.2^a	17.3^b	20.2^c	0.28	0.034
Neu (%)	46	44	45	1.88	0.11
Lym (%)	43^a	52^b	52^b	1.12	0.041
Eon (%)	4^b	1^a	1^a	0.10	0.022
Mon (%)	2	2	2	0.22	1.00
Bas (%)	5^b	1^a	0.0	0.10	0.01

Means within each row with different superscript differ significantly *(P < 0.05)*

As shown in Table 2, the WBC count was significantly (*P < 0.05*) highest with the T_3 group and therefore different from animals' of groups T_1 and T_2. However, animals on the T_2 diet had a significantly (*P < 0.05*) higher WBC count than those of T_1 that had significantly (*P < 0.05*) the lowest WBC count. There were no differences (*P > 0.05*) in the Neu between treatment groups. In the Lym, animals in the T_2 and T_3 groups had similar values that were significantly (*P < 0.05*) higher compared with the control (T_1) that had a significantly (*P < 0.05*) lower value. Nevertheless, the T_1 animals had significantly (*P < 0.05*) higher value of

EON compared with animals of T_2 and T_3 that had similar values. The trend observed with the EON was also similar with the BAS.

Discussion

The wholesomeness and thus lack of deviations from normal blood morphologies and levels are indicators of the effectiveness of the dietary antioxidant vitamins in the synthesis of antioxidant molecules for maintaining good health of the animals that ingest them. Blood is always used to assess the animal in terms of performance and profitability. Vitamins especially the antioxidant vitamins particularly are implicated in the overall wellness of the animal and hence increased performance and other economic indices, such as profit margins.

Therefore, as earlier eluded to blood parameters are one of the major indices of measuring or determining the physiological, pathological and nutritional status of any living organism, including poultry. Accordingly, therefore, changes observed in the blood constituents when compared to the control, for instance the T_1 group values in this current study could be used to explain at least in part the metabolic state of the animal as well as the quality of the feed of the animal (Babatunde *et al*., 1992; Ekenyem & Madubuike, 2006). To this extent, Ekenyem and Madubuike, 2006 demonstrated that haematological parameters can be used to gain more insights about the physiological disposition of the animal and consequently to their nutrition. This assertion becomes more dependable as it has been further shown that haematological parameters are affected by factors like nutrition, environment and health condition of the animal (Menzel, 1992; NRC, 2012).

One major take away from this current study was the fact that animals that received extra vitamins or the vitamins-based diets (T_2 and T_3) had higher values of PCV, Hb, RBC, WBC and LYM compared to the control (T_1) group. Furthermore, the animals in the control group demonstrated numerical higher value for NEU. This observation in this study supports the fact that antioxidant vitamins are capable of enhancing the quality of lives of poultry. This finding further demonstrates that antioxidant vitamins can stimulate the immune system of the animal as clearly evidenced by the significant higher values of LYM in the animals that ingested the vitamins-based diets compared to the control. This thus resulted in increasing the ratio of LYM to NEU compared to the control.

When this finding is further extrapolated it simply demonstrates the fact that antioxidant vitamins were capable of stimulating a protective immune response that can be adequate to induce resistance to pathogens and possible other environmental factors that can cause ill-health. Therefore, this finding is in tandem with previous workers, such as those of De La Fuente and Victor (2000) and Fragou *et al*. (2004).

Conclusion

Dietary ingestion of antioxidant vitamins improved haematological parameters and also improved the immune functions of broiler chickens; indicating that the ingestions of the vitamins improved the quality of lives of the animals. Therefore, this strategy can be used in enhancing the health status of broilers for improved productivity.

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