Evaluation of the Effect of Diet Containing Cinnamon and Red Pepper on Serum Biochemistry and Hematological Indices of Broiler

Ugwuowo, L. C.[1*], *Osita, C. O.*[2], *Ezeano,C. I.*[3] *and Nnamani, L.*[1]

[1]*Department of Animal Science, Nnamdi Azikiwe University Awka Nigeria*
[2]*Department of Animal Science, University of Nigeria, Nsukka.*
[3]*Department of Agricultural Economics and Extension, Nnamdi Azikiwe University Awka*

KEYWORDS

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ABSTRACT

This study was conducted to evaluate the effect of diet containing cinnamon and red pepper on serum biochemistry and hematological indices of broiler. One hundred and twenty (120) day old broiler chicks (Ross 308) were randomly divided into 4 treatment groups and 3 replicates of 10 birds per replicate in a completely randomized design. A mixture of cinnamon and red pepper at the ratio of 1:1was put in broiler diets at 0%, 0.5%, 1.0% and 1.5% at starter and finisher stages and were fed to broilers for 8 weeks. At the end of the experiment, 3 birds per replicate were selected and used for analyses of serum biochemistry and haematological indices. Results of serum biochemistry showed significant (P <0.05) effects of dietary cinnamon and red pepper inclusion on Cholesterol, aspartate aminotransferase, creatinine, urea, alkaline phosphatese, alanine aminotransferase, total protein, albumin, glucose, red blood cell count, white blood cell count, hemoglobin, packed cell volume, mean cell hemoglobin, neutrophils, platelet, mean cell volume and mean cell haemoglobin concentration. The results of the experiment showed that cholesterol and urea levels decreased as the level of cinnamon and red pepper increased but treatment three had the normal cholesterol level for broiler. Therefore, cinnamon and red pepper mixture should be included at 1.0% for optimum growth and performance of broiler birds.

*CORRESPONDING AUTHOR

lc.ugwuowo@unizik.edu.ng

INTRODUCTION

Protein from animal is important in human nutrition due to its high level of amino acid content (Tewe, 1997). Animal proteins have higher essential amino acids than plant protein. Malnutrition as a result of deficiency of vital nutrients has been an issue of serious concern in most areas of Africa (Agunbiade *et al*., 2000). Avaliable information indicates that poultry production is the fastest means of bridging the animal protein deficit in Africa (Oluyemi and Roberts, 1988). This is because, poultry has a short production period, a high rate of reproduction, and is distinguished by relatively high efficiency of nutrient modification into quality animal protein (Akinfala *et al*., 1999). Broiler production is considered most profitable in poultry industry. Broilers are bred as mixed-sex flocks in large intensive conditions (Olaifa *et al*., 2019). Farmers can raise broilers severally in batches within a year and in large numbers in order to meet the demand for an ever increasing human population. Phytobiotics are natural bioactive compounds that are got from plants and included in animal feed or water as an extract to promote productivity through the improvement of digestibility, nutrient absorption, and elimination of pathogens found in the alimentary canal of animal. Red pepper is commonly used in diet and traditional medicine. It is a good source of protein and also reduces the cholesterol level for having antioxidant potential. It has antimicrobial peptides which are very efficient in inhibiting growth in human and plant pathogenic bacteria and fungi. It is very rich in vitamin C and pro-vitamin A and B and it is very high in potassium, magnesium, and iron. The substances that give hot pepper the heat are capsaicin and several related chemicals collectively called capsaicinoids. These two phytobiotics with their amazing properties can therefore work in synergy to improve the Serum biochemical and hematological indices of broiler which are reliable indicators of the health status of animals and may have vital roles in prognosis, diagnosis and treatments of poultry diseases.

MATERIALS AND METHODS

Study Location:The study was carried out at the poultry unit of the teaching and research farm of the Department of Animal Science and Technology Nnamdi Azikiwe University, Awka Anambra state.The location lies between latitude 6.24°N and 6.28°N and Longitude 7.00°E and 7.08°E on the southeastern part of Nigeria. The climatic condition is the tropical wet and dry type with a clear season. The mean daily maximum temperature is usually 27°c all-round the year and can be 34°c in March. The mean annual rainfall is about 1600m according to the local meteorological station which has maintained records since 1978 with a relative humidity of 80% at night.

Collection and Processing of Cinnamon and Red Pepper: Air dried cinnamon bark and red pepper were procured from commercial dealers. The air-dried cinnamon and red pepper were milled into fine particles using a local milling machine. The ground samples were stored in an airtight container under room temperature (23.1 – 24.6 ^{0}C).

Experimental Diets*:* According to the Hubbard classic nutrient requirements, two basal diets were formulated and offered as broiler starter diet (0 - 28 days of age) and broiler finisher diets (28 to 56 days of age). Cinnamon and red pepper were mixed at the ratio of 1:1 and the mixture was included at 0, 0.5, 1 and 1.5% levels to represent T1,T2, T3 and T4 respectively in the ration of starter and finisher to form eight different treatment diets. The feed ingredients and composition of starter and finisher diets are presented in Table 1 and 2.

Table 1: Composition of broiler starter diets containing different inclusion levels of cinnamon and red pepper mixture.

INGREDIENTS	**T1 (0%)**	**T2 (0.5%)**	**T3 (1.0%)**	**T4 (1.5%)**
Maize	44	43	42	40.5
Wheat offal	8	8	8	8
Rice bran	6	6	6	6
Soybean meal	20	18.5	17	16
Fish meal	4	4	4	4
Groundnut cake	10	12	14	16
Bone meal	3	3	3	3
Oyster shell	3	3	3	3
Salt	0.5	0.5	0.5	0.5
Cinnamon + red pepper mixture	0	0.5	1.0	1.5
Lysine	0.5	0.5	0.5	0.5
Methionine	0.5	0.5	0.5	0.5
Vit/min premix	0.5	0.5	0.5	0.5
Total	100	100	100	100
Calculated Crude protein (%)	22.16	22.10	22.04	22.00
Calculated ME (kcal/kg)	2700.00	2700.00	2700.00	2700.00
Analysed crude protein (%)	21.38	21.00	21.7	21.2
Analysed ME (kcal/kg)	2677.00	2650.00	2630.00	2690.00

Table 2: Composition of broiler finisher diet containing different inclusion levels of cinnamon and red pepper mixture**.**

INGREDIENTS	**T1 (0%)**	**T2 (0.5%)**	**T3 (1.0%)**	**T4 (1.5%)**
Maize	49	47	45	43
Wheat offal	9	9	9	9
Rice bran	6	6	6	6
Soybean meal	20	20	20	20
Fish meal	4	4	4	4
Groundnut cake	4	5.5	7	8.5
Bone meal	3	3	3	3
Oyster shell	3	3	3	3
Salt	0.5	0.5	0.5	0.5
Cinnamon+red pepper mixture	0	0.5	1.0	1.5
Lysine	0.5	0.5	0.5	0.5
Methionine	0.5	0.5	0.5	0.5
Vit/min premix	0.5	0.5	0.5	0.5
Total	100	100	100	100
Calculated Crude protein(%)	20.20	20.05	20.01	20.00
Calculated ME(Kcal/kg)	3000.00	3000.00	3000.00	3000.00
Analysed crude protein (%)	19.50	19.60	19.00	19.80
Analysed ME (kcal/kg)	2900.00	2900.50	2998.00	2950.00

Experimental Birds: One hundred and twenty (120) unsexed day-old broiler chicks (Ross 308) were used in this study. The broiler chicks were procured from AGRITED hatchery Ibadan, Oyo state through their state distributor.

Experimental Design: Complete randomized design was used in the study. One hundred and twenty (120) broiler chicks were divided into four treatment groups. Each treatment contained 30 chicks and was randomly subdivided into 3 replicates with 10 chicks per replicate. Each treatment group was assigned to each of the treatment diets.

Experimental Procedure: Blood collection: Birds in each replicate pen were reared in individual pen. On the 56th day of study, blood samples were randomly collected from four birds per treatment. Blood samples of 2ml were collected via the wing veins using sterile needles and syringes. The blood samples were put in properly labeled and sterilized anticoagulant (EDTA) tubes and used for hematological analysis. Also, 2ml of blood was collected from each bird and was put into tubes without anticoagulant, for estimation of serum biochemistry. The blood samples inside tubes without coagulant were placed in a slanting position at room temperature for 6 h and incubated overnight in the refrigerator at 4^{o}C to obtain serum. The serum samples were kept at -20 °C before biochemical analysis.

Assessment of serum biochemistry indices: The following biochemistry analyses were estimated: total protein, albumin, creatinine, glucose, cholesterol, urea, AST, ALT, ALP,

Evaluation of hematological indices: The samples were subjected to hematological analysis which included hemoglobin (Hb) concentration, red blood cells (RBC), packed cell volume (PCV), mean cell hemoglobin (MCH), mean cell HB concentration (MCHC), white blood cells (WBC), mean cell volume (MCV), neutrophils, lymphocytes (LY), using Hematological Analyzer Sysmex XP- 100 (Japan).

Statistical Analysis: Data generated were subjected to one way analysis of variance (ANOVA). Differences ($P<0.05$) between treatments means where observed were separated using Duncan's multiple range test (DMRT) as outlined by (Steel *et al*., 1997).

RESULTS AND DISCUSSION

Haematological indices of broiler finisher birds fed diets containing different levels of cinnamon and red pepper mixture.

The effects of different levels of inclusion of cinnamon and red pepper mixture on haematological indices of broiler finisher are shown in Table 3.

Table 3: Haematological indices of broiler finisher birds fed diets containing different levels of cinnamon and red pepper mixture

PARAMETERS	TREATMENTS				
	T (0%)	T2 (0.5%)	T3 (1%)	T4 (1.5%)	P value
Hemoglobin (Hp)	12.03^{b}	13.17^{d}	12.97^{c}	11.03^{a}	0.00
Pack Cell Volum (Pcv)	35.00^{a}	38.43^{b}	39.07^{b}	34.00^{a}	0.00
Red Blood Cell (Rbc)	2.51^{a}	2.49^{a}	2.63^{b}	2.82^{c}	0.00
White Blood Cell (Wbc)	194.17^{c}	192.57^{b}	202.87^{d}	47.43^{a}	0.00
MCH	50.77^{a}	52.67^{c}	$52.1O^{b}$	50.70^{a}	0.00
MCV	146.10^{b}	138.33^{a}	138.37^{a}	147.20^{b}	0.00
MCHC	34.50^{a}	38.10^{c}	37.73^{b}	34.57^{a}	0.00
Platelets	28.00^{b}	17.00^{a}	15.33^{a}	45.67^{c}	0.00
Lymphocyte	2.97^{c}	1.00^{a}	2.00^{b}	1.00^{a}	0.00
Neutrophils	96.20^{b}	97.13^{c}	94.17^{a}	96.23^{b}	0.00

a,b,c,d Means on the same row bearing different superscripts are significantly different ($P < 0.05$)

Hemoglobin, packed cell volume, red blood cell count, White blood cell count, Mean cell hemoglobin concentration, Lymphocyte, platelets and neutrophilis showed significant differences ($p<0.05$) among treatment groups for 8 weeks old broiler birds. The findings in this study agreed with the results of Reiss *et al* (2018), who reported that inclusion of phytochemicals such as cinnamic aldehyde, thymol and carvaacrol in broiler birds significantly increased red blood cell counts and haemoglobin in comparism with the control group. Similar findings in another study were reported by krauze *et al.,* (2020) who studied the dietary effects of probiotic, bacillus subtilis (0.25g/l) enterococcus faecium 0.25g/l), and phytobiotics containing cinnanmon oil (0.25g/l) in broiler birds and found improvements in the immune system and parameters such as red blood cell count and hemoglobin.

The RBC values, WBC, MCH, MCV and MCHC values obtained for the birds in this study were higher than value reported for normal chicken by Mitruka *et al*., (1997) and also higher than the one reported by (Orawan *et al*., 2007).

The number of red blood cells in chicken influences the overall conditions of the birds.

Therefore, the increases in packed cell volume, Hemoglobin and erythrocyte counts of the birds fed the test ingredients are indication that the oxygen carrying capacity of the blood was improved. A decrease in the hemoglobin, mean corpuscular volume, and MCH levels in birds is also an indication that the birds were exposed to stressors (Huff *et al*., 2008).

Dietary additive of cinnamon and red pepper mixture improved the blood cells suggesting better utilization of the dietary nutrients.

Serum biochemistry indices of broiler birds fed diets containing different levels of cinnamon and red pepper mixture

The effects of cinnamon and red pepper on serum biochemistry of broiler at finisher phase are shown in Table 4.

table 4: Serum biochemistry indices of broiler birds fed diets containing different levels of cinnamon and red pepper mixture

PARAMETERS	TREATMENTS				
	T1 (0%)	**T2 (0.5%)**	**T3 (1%)**	**T4 (1.5%)**	**P value**
Total protein	5.54^{d}	5.36^{b}	5.19^{a}	5.48^{a}	0.00
Albumin	3.24^{a}	3.64^{c}	3.71^{d}	3.50^{b}	0.00
Creatinine	1.27^{b}	1.04^{a}	1.93^{c}	1.17^{ab}	0.00
Glucose	72.00^{a}	99.00^{c}	81.00^{b}	81.33^{b}	0.00
Cholesterol	195.00^{d}	99.17^{b}	105.67^{c}	90.43^{a}	0.00
Urea	42.05^{d}	30.23^{c}	18.83^{a}	26.00^{b}	0.00
AST	50.27^{c}	22.00^{a}	55.00^{d}	35.00^{b}	0.00
ALT	50.32^{c}	28.87^{a}	62.33^{d}	32.33^{b}	0.00
ALP	128.30^{a}	156.47^{b}	225.43^{d}	201.00^{c}	0.00

a,b,c,d Means on the same row bearing different superscripts are significantly different ($P < 0.05$)

Total protein, Albumin, creatinine, glucose, cholesterol, urea, AST, ALT, ALP showed significant differences ($p<0.05$) among treatment means for serum indices of 8 weeks old broiler birds. Cholesterol of T1 control group was significantly higher ($p<0.05$) than the values of those in the treatment groups fed diets containing mixture of cinnamon, and red pepper. The findings of the current study showed that increasing dosages of cinnamon and red pepper decreased the levels of cholesterol. The activities of aspartate aminotransferase (AST), alkaline phosphatase (ALP), and alanine aminotransferase (ALT) in the blood are bioindicators of liver damage and function (Yildirim *et al.,* 2011). Increased quantities of these enzymes are associated with liver or muscle damage, resulting from the body reactions to stress. The reduction in AST and ALT due to cinnamon and red pepper treatments can be deduced as an indication of better liver function. Reduced activity of ALP may be an indication of a slowdown of bone growth (Szabo *et al*., 2005). Higher serum levels of alkaline phosphatase are observed when there is improved osteoblastic activity, involving the formation and mineralization of bone associated with increased skeletal growth (Lumeij *et al.,* 2008). Also, the values of ALP in the present study are in agreement with the referenced values of 167ul – 305ul for poultry birds (Oleforuh-Okoleh *et al.,* 2015). Creatinine of T3 fed diet containing 1.0% inclusion level of cinnamon and red pepper showed higher value than the other dietary group and it also shows that T4 was statistically similar to T1 and T2. These findings indicated that mixture of cinnamon and red pepper had no harmful effects on kidney function. Various studies using phytobiotics supplementation in broiler chickens supports this results, including the work by Rubio *et al*, (2019), Ahmad *et al*.,(2018), and Adegoke *et al*.,(2018). Glucose value of T2 fed diet containing 0.5% level of cinnamon and red pepper mixture was significantly higher ($p<0.05$) than the values of other treatment group. The normal reference range of serum glucose in broiler is 200 to 500 mg/dL (Thrall *et al*., 2012). The present study showed that the serum glucose concentrations were influenced by cinnamon and red pepper in the experimental chickens; however, low values were recorded in the control group compared to treatment groups. Total protein of T1 was higher than the ones in the treatment group containing cinnamon and red pepper mixture. Albumin of T3 fed diets containing 1.0 inclusion levels of cinnamon, and red pepper mixture showed higher value than the ones in the other group. The quantity of serum protein in birds is considered an important indicator for the determination of their health status. The results showed that the inclusion of cinnamon and red pepper significantly increased the levels of albumin compared to the control group but the total protein decreased among treatment group and increased in the control group. In addition, albumin showed linear increases with increasing supplementation of cinnamon and red pepper mixture at 1.5%.

CONCLUSION

The research results showed that incorporating cinnamon and red pepper mixture at 0.5 % in broiler diets improved the serum and haematological indices which led to better health.

REFERENCES

Adegoke, A.V.; Abimbola, M.A.; Sanwo, K.A.; Egbeyale, L.T.; Abiona, J.A.; Oso, A.O.; Iposu, S.O. (2018). Performance and blood biochemistry profile of broiler chickens fed dietary turmeric (Curcuma longa) powder and cayenne pepper (Capsicum frutescens) powders as antioxidants. Vet. Anim. Sci. 2018, 6, 95–102.

Agunbiade, J.A., Adeyemi, O.A., Adepoju, O.A. and Lawal, O.A. (2000). The use of wholecassava meal and leaf in broiler diets. Tropical Journal of Animal Science, 5(1): 161 – 173

Ahmad, S.; Khalique, A.; Pasha, T.N.; Mehmood, S.; Hussain, K.; Ahmad, S.; Bhatti, S.A. (2018). Influence of Feeding Moringa oleifera Pods as Phytogenic Feed Additive on Performance, Blood Metabolites, Chemical Composition and Bioactive Compounds of Breast Meat in Broiler. Kafkas Univ. Vet. Fak. Derg. 2018, 24, 195–202. [CrossRef]

Akinfala, O., Kehinde, A.S., Abu, A.O. and Tewe, O. O. (1999). Performance and economy of production of pigs fed whole cassava plant based diets in the tropics. Journal of Animal Production Investigation, 2: 181 – 186.

Huff, G.R., Huff, W.E., Rath, N.C., Anthony, N.B. and Nestor, K.E. (2008). Effects of Escherichia coli challenge and transport stress on hematology and serum chemistry values of three genetic lines of turkeys. Poultry Science. 87:2234–2241

Krauze, M.; Abramowicz, K.; Ognik, K. (2020). The effect of the addition of probiotic bacteria (Bacillus subtilis or Enterococcus faecium) or phytobiotic containing cinnamon oil to drinking water on the health and performance of broiler chickens. Ann. Anim. Sci. 2020, 20, 191–205.

Lumeij, J.T. (2008). Avian clinical biochemistry. Pages 839–872 in Clinical Biochemistry of Domestic Animals. 6th ed. J. J. Kaneko, J. W. Harvey, and M. L. Bruss, ed. Academic Press, Burlington, MA.

Mitruka, B.M. and Rawnsley, H.M. (1997). Clinical Biochemical and Hematological References values in normal Experimental Animals.Masson Publshing USA., Inc. p. 278.

Olaifa R.O., OM Sogunle, LI Obileye, SA Ibitoye, TM Ayodeji, OJ Odutayo (2019). Effect of oral administration of three different phytobiotics on growth performance of locally-adapted turkeys. *EC Vet Sci* 4, 656-62

Oleforuh-okoleh, V.U., Ndofor-foling, H.M., Olorunlekes, S.O. and Uguru, J.O. (2015). Evaluation of growth performance, hematological and serum biochemical response of broiler chickens to aqueous extract of ginger and garlic. Journal of Agricultural Science 7(4):167-173.

Oluyemi, J.A. and Roberts, F.A. (1988). Poultry production in warm wet climate. Macmillan Publishers Limited, London.

Reiss Allison B., Hirra A Arain, Mark M. Stecker (2018). Amyloid toxicity in Alzheimer's disease. Reviews in The Neurosciences (Rev Neurosci)-Vol. 29, Iss: 6, pp 613-627.

Rubio, M.S.; Laurentiz, A.C.; Sobrane, F.; Mello, E.S.; Filardi, R.S.; Silva, M.L.A.; Laurentiz, R.S. (2019). Performance and Serum Biochemical Profile of Broiler Chickens Supplemented with Piper Cubeba Ethanolic Extract. Braz. J. Poult. Sci. 2019, 21, 1–8.

Steel, R.G.D. and Torrie, J.H. (1997). Principles and Procedures of Statistics: A Biometrical Approach. 3rd edition.

Szabo, A., Mezes, M., Horn, P., Suto, Z.,Bazar, G. and Romvari, R. (2005). Developmental dynamics of some blood biochemical parameters in the growing turkey (Meleagris gallopavo). *Acta Veterinary Hungary*, 53:397-409

Tewe, O.O. (1997). Sustainability and development paradigm from Nigeria‘s livestock industry. Inaugural Lecture delivered on behalf of Faculty of Agriculture and Forestry, University of Ibadan, Nigeria. Pp. 50.

Thrall, M.A.; Weiser, G.; Allison, R.; Campbell, T. (2012). Veterinary Hematology and Clinical Chemistry, 2nd ed; John Wiley and Sons: Oxford, UK, 2012; pp. 582–598.