HAEMATOLOGY AND SERUM BIOCHEMISTRY OF BROILER CHICKEN FED WILD COLOCYNTH FRUIT EXTRACT (*Lagenaria breviflora*) IN WATER

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Abstract
A study was carried at the Poultry Unit of the Teaching and Research Farms, Oyo State College of Agriculture and Technology, Igboora with a total number of 96 unsexed day-old broiler chicks which were randomly allotted into 4 groups of 24 birds each in a completely randomized design (CRD). Each group was subdivided into 3 replicates of 8 birds each having an inclusion level of 0ml, 15mls, 30mls and 45mls of wild colocynth fruit extract (WCFE) for a period of 7 weeks. Parameters collected on haematology (Packed cell volume (PCV) (%), red blood cells (RBC) ($x10^6$µl), haemoglobin (Hb) (g/dl), white blood cells (WBC) **(**$x10^3$µl), platelet (µl), eosinophil (%), basophil (%), heterophil (Het) (%), heterophil/lymphocyte ratio (H/L), Monocyte (Mon) (%) and serum (Total protein (Tp) (g/dl)), Albumin (Al) (g/dl), Albumin/Globulin (A/G), Globulin (g/dl), Creatinine (mg/dl), Glucose (mg/dl, Cholesterol (mg/dl), Triglycerides (Trigl) (mg/dl), AST (µl), AST (µl) and ALP (µl)) were subjected to ANOVA using SAS 2012 software, significant means were separated using Duncan's Multiple Range Test. The result obtained showed that WCFE inclusion in drinking water of the birds significantly ($p<0.05$) affected Het, H/L, Mon, Tp, Alb, A/G as well as Trigl while other parameters were similar ($p>0.05$) across the treatment groups. It was concluded from the study that WCFE in drinking water of broiler chicken had no detrimental effects on haematology and serum biochemistry of broiler birds as all the parameters measured were within range of values and can be included up to 45ml/litre to enhance health status of broiler chicken
Keynotes: - Broiler, blood, platelet, serum, total protein

Introduction

Poultry production is unique because it offers the highest turnover and the quickest return on investment than most other livestock enterprise (Saani and Ogundipe, 2005) which has made it become a popular industry for even the small holders that have greatly contributed to the economy. In this regard, poultry industry has witnessed rapid expansion in the past years and the industry has assumed great importance in improving employment opportunity and animal protein in Nigeria. However, the development of resistance to antibiotics and other drugs by pathogens in livestock and consumers of the animal products besides other complications was noticed. This stimulated the interest of many researchers to investigate and explore the use of herbs, spices, plant extracts and their products as botanicals or organic supplements in animal rations (Hernadez *et al.,* 2004 and Afolabi *et al*., 2016). Spices and herbs of various plants extract have appetizing, digestion stimulating property and antimicrobial effects (Alcicek *et al*., 2003). Odoemelam *et al*. (2013) reported that up to one third of all commercial swine and chicken rations in Europe now use mixtures of herbs and spices to accelerate growth and maintain good health. *Lagenaria breviflora* (wild colocynth) is one of those plants with characteristics antibacterial and antiviral herbal remedies in local communities such as Nigeria (El mahmood *et al.,* 2008) and the medicinal values of these plant lie in their component phytochemical mostly alkaloids, tannins, flavonoids, and phenolic compound. The importance of phytogenics in improving blood parameters has necessitated research with the objective of determining the haematological and biochemical indices of broiler chicken fed wild colocynth fruit extract in water.

MATERIALS AND METHODS

Location of the study

The experiment was carried at the Poultry Unit of the Teaching and Research Farm, Oyo State College of Agriculture and Technology, Igboora

Collection and Preparation of Wild colocynth (*Lagenaria breviflora)* fruit extract (WCFE)

Fresh wild colocynth fruits were harvested from Igbo-ora community and environs. The fruits were washed, peeled, cut into pieces, and weighed. 1kg of the fruit was added to 2 Litres of water in a

breathable mesh container, covered tightly and allowed to stay for a period of 7 days. It was then filtered, and the filtrate (extract) was kept at room temperature prior to use.

Experimental animal managements

A total number of ninety (96) unsexed day-old broiler chicks (Marshal ®) were purchased from a reputable hatchery. On arrival, chicks were carefully removed from their box and randomly allotted into groups. Water and feed were supplied *ad libtum*. No vaccination nor other synthetic drugs were administered during the period of study. The wild colocynth fruit extract (WCFE) served as sources of antibiotic, antiviral as well as multivitamin for the treatment groups except the control. The feeding program consisted of the starter diet that was used until 21 days of age and a finisher diet until 49 days of age.

Experimental design

The chicks were randomly allotted into 4 treatment groups of 24 birds each in a completely randomized design (CRD). Each group was subdivided into 3 replicates of 8 birds each placed on a deep litter. The study lasted for a period of 42 days

Treatment was as follows:

T1 = 0 ml of WCFE, T2 = (15 ml of WCFE + 1 litre of water), T3 = (30 ml of WCFE + 1 litre of water)

T4 = (45 ml of WCFE + 1 litre of water)

Data collection

Haematology and serum biochemical characteristic

Blood sample was collected at the 7th week of the experiment, one bird per replicate was randomly selected and blood samples was collected from the jugular veins of each birds using sterilized syringe and emptied into sterilized bottles for serum analysis and another set of bottles containing EDTA for haematological analysis. Packed cell volume (PCV), red blood cell (RBC) and white blood cell (WBC) differential counts. Blood samples for serum analysis after clotting was centrifuged at 3,500 rpm in the laboratory using Gallenkamp laboratory centrifuge in order to obtain the serum. The serum obtained was used to determine the total protein, creatinine, glucose, albumin and cholesterol according to Randox procedure of chemical analysis (2010).

Statistical analysis

All data collected during the study were subjected to analysis of variance (ANOVA) using the Statistical Analysis System (SAS 2012) software. Significantly different means were separated using Duncan's Multiple Range Test (DMRT) of the same software

Table 1: Ingredients and nutrients composition of starter and finisher diet (%)

Ingredient %	**Starter (1-21 days)**	**Finisher (22-49 days)**
Maize	58.00	63.80
Soybean meal	34.50	28.40
Soya oil	3.30	3.70
Di-calcium phosphate	1.70	1.70
Limestone	1.80	1.70
Salt {NaCl)	0.25	0.25
Methionine	0.15	0.15
Premix*	0.30	0.30
Total	**100**	**100**
Calculated composition (%)		
Crude protein %	22.10	19.18

Energy (MEKcal/kg)	3030	3100
Ether extract **%**	5.79	6.34
Crude fibre **%**	2.62	2.51
Calcium **%**	1.04	1.00
Phosphorous **%**	0.46	0.45
Methionine **%**	0.53	0.45

*premix supplied per kg diet: vitamin A 15,000 I.U, vitamin D3 13000 iu, thiamin 2mg, Riboflavin 6mg, pyridoxine 4mg, Niancin 40mg, cobalamine 0.05g, Biotin 0.08mg, choline chloride 0.05g, Manganese 0.096g, Zinc 0.06g, Iron 0.024g, Copper 0.006g, Iodine 0.014g, Selenium 0.24mg, Cobalt 0.024mg and Antioxidant 0.125g. NFE = Nitrogen free extract

RESULTS

The results of the effect of wild colocynth (*Lagenaria breviflora)* fruit extract (WCFE) on haematology of broiler chicken is presented in table 2

The Table revealed wild colocynth (*Lagenaria breviflora)* fruit extract (WCFE) inclusion in the water significantly ($p<0.05$) affected heterophil (Het) (%), heterophil/lymphocyte ratio (H/L) as well as Monocyte (Mon) (%). However, Packed cell volume (PCV) (%), red blood cells (RBC) ($x10^6$µl), haemoglobin (Hb) (g/dl), white blood cells (WBC) **(**$x10^3$µl), platelet (µl), eosinophil (%), basophil (%), mean cell volume (MCV) (fl), mean corpuscular haemoglobin concentration, (MCHC) (%) and mean cell heamoglobin (MCH (pg) were similar ($p>0.05$) across the treatment group. Het showed an increasing trend with increase in WCFE inclusion form T1 – T4 with mean values of 27.67, 34.00, 34.33 and 36.00% respectively. Likewise, mean values H/L also showed an increasing trend with increase in WCFE levels although, there was no difference between H/L of T2 (0.59), T3 (0.69) and T4 (0.64). Mon was observed to be highest at T3 (30mls) (4.00) while T1(0mls) (3.00) and T2 (15mls) (3.00) had comparable means with that of T4 (45mls) (2.00).

Table 2: Effect of wild colocynth (*Lagenaria breviflora)* fruit extract (WCFE) on haematology of broiler chicken

Parameters	**T1 (Control)**	**T2 (15mls) WCFE)**	**T3 (30mls) WCFE**	**T4 (45mls) WCFE**	**SEM ±**	**P-value**
PCV (%)	25.00	28.00	24.67	23.00	1.38	0.230
H.b (g/dl)	8.23	9.07	8.20	7.50	0.94	0.371
RBC ($\times10^6$µl)	2.23	2.50	2.16	2.08	0.15	0.432
WBC ($\times10^3$µl)	13117	14100	12917	12867	463.38	0.405
Platelet (µl)	122,000	114,000	113,000	125,000	4333.25	0.292
Heterophils (%)	34.00^a	27.67^b	34.33^a	36.00^a	1.72	0.045
Lymphocytes (%)	58.33	61.67	58.00	56.67	1.69	0.324
H/L	0.59^a	0.45^b	0.60^a	0.64^a	0.04	0.059
Eosinophils (%)	4.33	4.00	3.33	4.47	0.56	0.452
Basophils (%)	0.00	0.33	0.33	0.67	0.25	0.487
Monocytes (%)	3.00^{ab}	3.00^{ab}	4.00^a	2.00^b	0.39	0.193

WCFE = Wild colocynth fruit extract, PCV = Packed cell volume, H.b = Haemoglobin, RBC = Red blood cell, WBC = White blood cell, MCV = Mean cell volume, MCHC = Mean corpuscular haemoglobin concentration, MCH = mean cell haemoglobin, H/L – Heterophil to lymphocyte ratio

The results of the effect of wild colocynth (*Lagenaria breviflora)* fruit extract (WCFE) on serum biochemistry of broiler chicken is presented in Table 3

The Table revealed wild colocynth (*Lagenaria breviflora)* fruit extract (WCFE) inclusion in the water significantly ($p<0.05$) affected Total protein (Tp) (g/dl)), Albumin (Al) (g/dl), Albumin/ Globulin (A/G) as well as Triglycerides (Trigl) (mg/dl). However, Globulin (g/dl), Creatinine (mg/dl), Glucose (mg/dl, Cholesterol (mg/dl), AST (µl), AST (µl) and ALP (µl) were similar ($p>0.05$) across the treatment group. Tp was observed to be highest at T3 (30mls) (7.93) while T1(0mls) (6.87) and T2 (15mls) (7.07) had comparable means with that of T4 (45mls) (6.47) likewise, the same trend was obtained for Alb, A/G ratio as well as Triglycerides

Table 3: Effect of wild colocynth (*Lagenaria breviflora)* fruit extract (WCFE) on Serum biochemistry of broiler chicken

Parameters	T1 (Control)	T2 (15mls) WCFE)	T3 (30mls) WCFE	T4 (45mls) WCFE	SEM ±	P-value
Total protein (g/dl)	6.87^{ab}	7.07^{ab}	7.93^{a}	6.47^{b}	0.29	0.140
Albumin (g/dl)	1.40^{ab}	1.60^{ab}	2.30^{a}	1.13^{b}	0.25	0.102
Globulin (g/dl)	5.47	5.47	5.63	5.33	0.16	0.685
A/G ratio	0.26^{ab}	0.29^{ab}	0.40^{a}	0.21^{b}	0.04	0.094
AST (µl)	198.00	207.30	222.30	188.67	11.63	0.394
ALT (µl)	28.33	29.33	33.33	24.87	2.89	0.350
ALP (µl)	290.67	272.30	336.00	270.30	25.15	0.345
Creatinine (mg/dl)	0.63	0.57	0.67	0.50	0.05	0.256
Glucose (mg/dl)	182.00	185.67	215.70	175.00	12.87	0.473
Cholesterol (mg/dl)	115.33	121.33	123.00	124.67	5.31	0.653
Triglycerides (mg/dl)	87.67^{ab}	93.67^{ab}	98.67^{a}	82.67^{b}	3.03	0.047

WCFE = Wild colocynth fruit extract, A/G = Albumin /Globulin ratio, AST = Aspartate aminotransferase, ALT = Alanine aminotransferase, ALP = Alkaline phosphatase

DISCUSSION

The non-significant level observed for most haematological parameters suggests an unequal influence of the treatments (i.e. varying levels of WCFE in drinking water) on the haematological indices of the birds (Al-Baghdadi, 2011). However, all the haematological values were within the normal range for healthy chickens reported by Mirtuka and Rawnsley, (1977). Therefore, non-significant influence of the treatments on some of the haematological indices signified their uncompromised physiological relevance in broiler chickens. This was evident in the healthy condition of the experimental birds throughout the duration of the study. WBC differentials are involved in recognising body intruders, killing harmful bacteria and creating antibodies to protect the body against future exposure to some pathogens like bacteria and viruses, the significant level recorded for monocyte (2.00-4.00%) were within the normal range 0-3% reported by (Kahn, 2005) but lower than the 16 % and 8.10-16.10% reported for broiler chickens by Sebastian *et al*. (2012). For the serum metabolites, the birds did not show any liver dysfunction along with others that were virtually similar. This could be as a result of the influence of the antioxidant potential of the extract that impacted beneficially on the physiological functions of the birds especially the ALT. However, the combined high levels of AST and ALT could be detrimental to liver function and consequently compromise the broiler chickens' health. The

albumin values of 1.13 – 2.30 g/dl observed in this study did not fall within the range of 25.00 – 45.00 g/dl and 10.80 – 16.00 g/dl reported by Harr (2002). These deviations could be associated with broiler breed and the treatment levels of WCFE used. Total serum protein and albumin serve as a measure of biosynthetic production of plasma proteins by the liver. Therefore, the level of albumin supported the functionality of the liver; (Oladele and Ayo, 1999).

Conclusion and Recommendation

It can be concluded from the study above that WCFE inclusion in drinking water of broiler chicken had no detrimental effects on blood properties of broiler birds and can be included up to 45ml/litre to enhance health status of broiler chicken.

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