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EFFECT OF HEAVY METALS CONTAMINATION ON PERFORMANCE, BLOOD PROFILE OF BROILER CHICKS FED CORN-SOYA MEAL DIET

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ABSTRACT
Five hundred, day old Arbo Acre broiler chicks were used in an experiment to evaluate the effect of heavy metals contamination on the performance and blood profile of broiler chicks fed corn-soya meal diet. The birds were randomly assigned to five treatment diets in a Completely Randomized Design (CRD) and each treatment group was further sub-divided into four replicates containing twenty birds each. The chicks were feed *ad-libitum* with formulated broiler starter diet supplemented with battery waste at levels of 0%, 5%, 10%, 15%, and 20% representing treatments 1,2,3,4 and 5 respectively. Feed intake, daily weight were the performance criteria measured while the blood profile includes heamatology and serum analysis. Results showed that broilers fed battery waste above 5% were significantly affected (P<0.05) in terms of their body weight, feed intake, haematological and biochemical analysis were affected. The result of this experiment demonstrated that inclusion of battery waste in broiler chicks above 5% have a deleterious effect on growth, performance and health status of birds.

KEY WORDS: broiler chicks, battery waste, performance, growth rate, health status.

INTRODUCTION

Poultry production has experienced a remarkable growth within the past Thirty five years. However, there are still a lot of problems facing the industry in its bid to attain viable level of production; some of these problems include disease, poor quality raw materials and most recently feed contamination during feed processing. Deposition of heavy metals in birds is due to the feeding of contaminated feed and water as well as exposure to different manufacturing processes of factories and industries (Gabol *et al.*, 2014). Animal Management involves a combination of good housing, health and feeding. Therefore the quality and safety of feed given to animals determines their performance. Heavy metals contamination originates from many sources such as industrial, anthropogenic and exogenous means. Hazardous wastes are generated by nearly all manufacturing industries (Hutton and Symon, 1986; Nriagu, 1989). Disposal of such wastes results in a serious pollution to the environment. Among the potential feed contaminant is the battery waste which is found to be loaded with heavy metals (Mc Cluggage, 1991).

According to Lenntech (2004) Heavy metals refers to any metallic element that has a relatively high density and is toxic or poisonous even at low concentration. Heavy metals include cadmium (Cd), arsenic (As), zinc (Zn), chromium (Cr), silver (Ag), copper (Cu), iron (Fe), mercury (Hg) and lead (Pb). Heavy metals are specifically regarded with their toxicological as well as carcinogenic effects (Shahneen and Akhtar, 2012). Some of these elements play an essential role in physiological and biochemical processes in living cells when present in required amount (Nolan, 2003; Young, 2005). For instance copper and iron are essential in the formation of hemoglobin in animal. When these elements are present in lethal dose, it results in deleterious effect such as reduced growth rate, malfunction of vital organs such as liver, kidney, spleen and consequently death of the animal. Supplementation of heavy metals with a large safety margin in broilers has resulted into higher mineral excretion and ends up in the environment (Abdul-Jameel *et al*., 2012; Demirezen Uruς́, 2006). Although extensive studies have been done on the effect of heavy metals on animals, yet there is a dearth of information on battery waste contamination in broilers. A timely evaluation of the effects of exposure to animals will provide useful information relating to the disposal and management of the waste. The objective of the studies, to evaluate the effect of battery waste on the performance of broiler chicks, determine the effect of heavy metals on the hematological and biochemical parameters in broiler chicks and to determine the tolerable level of heavy metals in diets of broiler chicks.

Justification of the study

Heavy metals are of toxic properties which can easily penetrate and accumulate into internal organs like the kidney, lungs, liver, and adrenal glands and in some cases causes' damage in the blood. The circulation or migration of heavy metals can be natural via rock decay and soil formation processes or anthropogenic through different branches of agricultural industries (Feed mill), waste management, waste dumping sites, fertilizers to plants which in turns get into the animal body. The heavy metals from these sources are dispersed in the environment and they contaminate soil, water and air causing biotoxic effects when consumed above their bio-recommended limits.

MATERIALS & METHODS

Location of experiment

The experiment is carried out at the Poultry unit of the Teaching and Research farm of Ladoke Akintola University of Technology (LAUTECH), Ogbomosho, Nigeria. The area is located within the derived savannah zone of Nigeria.

Animals and their management

A total of Five hundred day old Arbo acre broilers obtained from a Commercial hatchery in Ibadan were used for the experiment. The birds were divided into five dietary groups and each group subdivided into four replicates of twenty broilers per group in a complete randomized experimental design. They were housed in battery cages designed at the centre. Wood shavings were used as litter material in the cages. The diets and water were offered ad libitum during the three week experimental period. Feed intake was recorded daily body weight and feed conversion ratios were recorded weekly.

Proximate analysis of diets and test ingredients were determined according to AOAC, (2000). Data obtained were subjected to analysis of variance (Steel and Torrie, 1980) and treatment means where significant were separated using Duncan Multiple Range Test (Duncan, 1955).

Blood Analysis

At the 3rd week blood of the experiment, blood samples were collected from the veins of sixteen randomly selected birds per treatment. The blood samples were analyzed for some haematological and biochemical indices. All blood samples for haematological indices were collected into bottles containing EDTA, as anti – coagulant. The haematological indices determined included pack cell volume (PCV), Haemoglobin concentration (Hb), White blood cell count, Red blood cell count (RBC), Mean corpuscular volume (MCV), Mean corpuscular haemoglobin concentration (MCHC) and White blood cells differential counts (*i.e*. Neutrophils, Lymphocyte, Monocytes, and Eosinophils). Packed cell volume (PCV) was determined by the micro- haematocrit method (Dacie and Lewis, 1991), the haemoglobin concentration was determined by the colometry-cyanomethacmoglobin method, Red blood cell counts (RBC) were determined by the improved Neubauer haemocytometer method (Kelly, 1979). While mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentrations (MCHC) were computed according to the method of Jain (1986). The differential white blood cell (WBC) counts were obtained by making a differential smear stained with Wright’s stain and the percentage counts taken for segmented neutrophils and lymphocytes (Dacie and Lewis, 1991). Blood samples that were meant for serum chemistry were collected into bottles free of any anti-coagulant. Parameters like albumin and globulin were colorimetrically analyzed using diagnostic reagent kits (Doumas and Briggs, 1972). Activities of serum glutamic oxaloacetate transaminase (SGOT), serum glutamic pyruvate transaminase (SGPT), and alkaline phosphatase (ALP) were determined colorimetrically (Reitman and Frankel, 1957).

Chemical Analysis

The proximate analysis of the experimental diet was determined by the method of AOAC (2000).

Statistical Analysis

Data collected were subjected to statistical analysis appropriate for the Completely Randomized Design (Snedecor and Cochran, 1980) and significant means separated by Duncan multiple range tests (Duncan, 1955).

RESULTS

The concentration of heavy metals in battery waste is shown in Table 1; Lead had the highest concentration with 540.13mg/l followed by copper 9.46mg/l. Nickel had the least concentration of 0.10mg/l.

TABLE 1. Concentration of Heavy metals in battery waste

Mineral	Concentration (mg/l)
Lead	540.13
Cadmium	0.43
Magnesium	7.17
Copper	9.46
Zinc	6.50
Manganese	3.11
Nickel	0.10

TABLE 2. Composition of experimental diets (%)

Ingredients	Diets				
	T1 (0%)	T2 (5%)	T3 (10%)	T4 (15%)	T5 (20%)
Maize	53.0	53.0	53.0	53.0	53.0
Soya Meal	32.0	32.0	32.0	32.0	32.0
Wheat offal	6.80	6.80	6.80	6.80	6.80
Fish meal	3.75	3.75	3.75	3.75	3.75
Bone meal	2.50	2.50	2.50	2.50	2.50
Oyster shell	1.00	1.00	1.00	1.00	1.00
1Premix	0.25	0.25	0.25	0.25	0.25
Methionine	0.25	0.25	0.25	0.25	0.25
Lysine	0.25	0.25	0.25	0.25	0.25
Salt	0.25	0.25	0.25	0.25	0.25
Total	100.00	100.00	100.00	100.00	100.00
Battery waste	0%	5%	10%	15%	20%

Calculated Analysis					
Dry matter	88.9	88.9	88.9	88.9	88.9
Crude protein	23.2	23.2	23.2	23.2	23.2
Ether extract	3.91	3.91	3.91	3.91	3.91
Crude fibre	4.16	4.16	4.16	4.16	4.16
Ash	3.63	3.63	3.63	3.63	3.63
Nitrogen free extract	56.6	56.6	56.6	56.6	56.6
ME kcal/kg	3005	3005	3005	3005	3005

Premix supplied per kg diet :- Vit A, 15,000 I.U; Vit E, 5mg; Vit D3, 3000I.U, Vit K, 3mg; Vit B2, 5.5mg; Niacin, 25mg ; Vit B12, 16mg ; Choline chloride, 120mg ; Mn, 5.2mg ; Zn, 25mg ; Cu, 2.6g ; Folic acid, 2mg ; Fe, 5g ; Pantothenic acid, 10mg ; Biotin, 30.5g ; Antioxidant, 56mg. 2ME: Calculated metabolizable energy

Table 2 shows the proximate composition of the experimental diets used for the experiment; it contains 88.9% dry matter, 23.2% crude protein, 3.91% ether extract, 4.16% crude fibre, 3.63% ash content and 3005 Kcal/kg of energy.

TABLE 3: Performance and Hematological traits of Broilers Fed varying levels of Battery waste

			Diets				
Parameters	T1	T2	T3	T4	T5	SEM	S/L
Av. Initial body weight (g)	39.0	40.0	42.0	39.7	41.2	0.49	*
Av.Final Live weight (g)	510	409	340	321	309	33.3	*
Av. Feed intake	1.23	1.15	1.4	1.20	1.20	0.16	*
Hemoglobin (g/dl)	3.56	2.31	2.78	1.90	1.40	0.27	*
Packed cell volume (%)	33.17	31.83	31.17	29.20	28.20	1.79	*
Red blood cells (×106mm^3)	5.05	3.06	2.02	1.70	1.40	0.67	*
MCV (FL)	59.9	57.4	46.0	38.3	27.9	0.76	*
MCH(Pg)	29.7	22.6	19.7	12.6	10.9	0.45	*
MCHC (g/dl)	42.33	40.67	38.67	22.80	20.78	0.32	*
WBC(×103/mm3)	5.36	5.66	5.85	7.67	9.56	0.21	*
Neutrophils (%)	35.6	31.0	34.0	35.3	34.7	0.65	*
Lymphocytes (%)	54.0	57.6	54.6	58.6	59.8	0.34	*
Monocytes (%)	0.67	1.33	1.80	1.17	1.50	0.65	*
Eosinophils (%)	6.30	5.78	5.90	5.78	5.67	0.43	*

NS – No significant difference (P>0.05)
*= significant difference (P<0.05)
MCHC – Mean corpuscular haemoglobin concentration
MCH – Mean corpuscular haemoglobin
MCV - Mean corpuscular volume
WBC – White blood cell
RBC- Red blood cell

Table 3 shows the growth performance and the various haematological indices investigated. The final live weight ranges between 510g and 400g. There was a significant difference (P<0.05) among the treatment in term of final weight.

TABLE 4: Serum biochemical parameters of broiler fed varying levels of battery waste

			Treatments			
Parameters	1	2	3	4	5	SEM
Albumin (g/dl)	9.62	7.46	3.90	1.23	0.78	0.44
Globulin (g/dl)	7.67	4.40	2.56	1.32	1.30	0.67
Total protein (g/dl)	6.56	6.30	1.67	1.34	0.70	0.97
SGPT (U/L)	11.23	4.55	3.12	2.67	2.01	0.45
SGOT (U/L)	19.89	15.60	4.09	4.34	1.69	0.53
ALP (U/L)	41.33	41.00	30.78	27.50	25.56	1.70
Cholesterol (mg/dl)	39.0	30.56	24.56	19.45	15.57	0.86

NS: No significant difference (P>0.05)
*= Significantly different (P<0.05)
SGPT: Serum glutamic pyruvic Transaminase
SGOT: Serum glutamic Oxaloacetate Transaminase
ALP: Alkaline Phosphatase

Hemoglobin values obtained are 3.56, 2.31, 2.78, 1.90 and 1.40 for diets 1, 2, 3, 4 and 5 respectively. The values obtained for Red blood count (RBC) are 5.05, 3.06, 2.02, 1.70 and 1.40 (×106/mm3) for diets 1, 2, 3, 4 and 5 respectively. Heamoglobin (Hb), Pack cell volume (PCV), Red blood cells (RBC), Mean corpuscular volume (MCV), Mean corpuscular heamoglobin (MCH) and Mean corpuscular heamoglobin concentration (MCHC) were significantly affected (P<0.05) by the dietary inclusion of battery waste. WBC values marginally increased from treatment 1 to 5 there values within the treatment were significantly affected (P<0.05), neutrophils, lymphocytes, monocytes and eosinophils were not significantly (P>0.05) influenced by different inclusion level of battery waste. The serum biochemical indices as influenced by the diets in Table 4. Total protein values are 6.56, 6.30, 1.67, 1.34 and 0.70 g/dl for treatments 1, 2, 3, 4 and 5 respectively. While albumin values are 9.62, 7.46, 3.90, 1.23 and 0.78g/dl for treatments 1,2,3,4 and 5 respectively. The values for cholesterol in mg/dl are 39.00, 30.56, 24.56, 19.45 and 15.57 for treatments 1, 2, 3 4 and 5 respectively. Albumin, globulin, total protein, cholesterol, Alkaline phosphatase (ALP), Serum glutamic oxaloacetate (SGOT), Serum glutamic pyruvate transaminase (SGPT) were significantly affected (P<0.05) with an increase in battery waste across the treatment.

TABLE 5: Recommended maximum level of heavy metals in drinking water by World Health Organization.

Heavy metals	Max. Acceptable Concentration (WHO)
Zinc	5mg/l
Arsenic	0.01mg/l
Magnesium	50mg/l
Cadmium	0.003mg/l
Lead	0.01mg/l
Silver	0.0mg/l
Mercury	0.001mg/l

DISCUSSION

There were significant differences (P<0.05) in values obtained in the final weight of broiler chicks across the treatments, this could be due to biotoxic effect of heavy metal to the body when consumed above the bio-recommended limits illustrated in table 5, thus having a negative effect on growth of birds, this agrees with the findings reported by (Nolan, 2003; Ferner 2001; Young, 2005) that heavy metals like Cadmium (Cd), Arsenic (As), Lead (Pb) and Mercury (Hg) have no known bio-importance to animal biochemistry and physiology and consumption even at a low concentration.

From the analysis of battery waste in Table 1, it reveals that lead and cadmium contains 540.13mg/l and 0.43mg/l respectively this exceeds the maximum recommended value by World health Organization in Table 6. Decreased in body weight was more pronounced in those group with higher dose of battery waste.

The values of pack cell volume (PCV), haemoglobin (Hb), Red blood count (RBC), Mean corpuscular haemoglobin (MCH), Mean corpuscular haemoglobin concentration (MCHC) were significantly (P<0.05) different. According to a research carried out by Ketavolos *et al*. (2007) he reported that lead causes hemolysis in swan birds, which can lead to anaemia and osmotic fragility. This clearly shows why the RBC values decrease with an increase in battery waste across the treatment. Johri *et al.* (2010) and Gerspacher, (2009) also reported that cadmium affects the cells, neuron, neurotransmitters and hemotologic values of poultry birds.

The effect of battery waste on the RBC agrees with the findings of Lagget (1993) who reported that high dose of heavy metals concentration showed high haemolysis as well as distruction of red blood cells leading to anaemia. This could also affect other blood parameters like the PCV, MCH and MCHC as the inclusion of battery waste increased across the treatments. White blood cell (WBC) values increased with an increase in battery waste across the treatment a significant difference was observed (P<0.05), it could be as a result of immune system disorder, a disease in the bone marrow or the an infection in the body system which triggers an increase in the WBC level across the treatment which agrees with the findings of Ogwuebgu and Muhanga, 2005 who reported that lead poisoning causes the inhibition of the synthesis of haemoglobin; dysfunctions of the kidneys, joints and reproductive systems, cardiovascular systems and acute and chronic damage to the central nervous system and the peripheral nervous system.

Mc Cluggage, 1991 also reported Zinc could also mimic lead poisoning leading to illness in animals and could also trigger the production of WBC. Biochemically, albumin, globulin, total protein, alkaline phosphatase values were significantly affected (P<0.05), these could be due to the presence of arsenic in the feed, their values decrease with an increase in battery waste in the feed which agrees with a report from (INECAR, 2000) that Arsenic acts to coagulate proteins, it forms coenzymes and inhibits the production of adenosine triphosphate (ATP) during respiration.

The reduction in albumin may be as a result of poorer liver functions as well as proteinuria due to kidney damage due to the presence of heavy metals like lead in the battery waste fed to the birds at various inclusion level (Patra *et al*., 2011). The values obtained for SGPT and SGOT pattern decrease with an increase in battery waste which shows a significant difference (P<0.05) across the treatment this result affirm the earlier observations of Romoser,1960 that lead and arsenic appears to exert its toxic effect through inhibition of enzymes and cell damage. Ogwuegbu and Ijioma, 2003 also reported that arsenic toxicity can affect the activity of some important enzymes and could also lead to death of an animal.

CONCLUSION

Most of the parameters tested *viz* growth performance, haematological and biochemical indices showed significant differences. It could therefore be concluded that the inclusion of heavy metals above 5% have deleterious effect on broiler chicks which could affect the growth, performance and health status of the birds.

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