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Haematological and Serum Biochemical Indices of Growing Cockerels Fed Varied Levels of Azolla-Turmeric Mixture

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ABSTRACT

This study was conducted to examine the haematological and some serum biochemical indices of growing Isa cockerels fed diets supplemented with Azolla-turmeric mixture. One hundred and twenty (120) four weeks old Isa white cockerels were divided into four groups with three replicates, each of ten birds in a completely randomized design. Treatment 1 was fed basal diet without Azolla – turmeric mixture (ATMP), treatment 2, 3 and 4 were fed basal diets supplemented with ATMP at levels 5, 10 and 15% respectively. Proximate analysis shows that Azolla powder contained 22.51% crude protein, 3.53% ether extracts, 14.13% ash, 18.15% crude fibre and 37.88% nitrogen free extract while turmeric powder contained 9.14% crude protein, 2.40% ether extracts, 8.69% ash, 4.10% crude fibre and 69.10% nitrogen free extracts. The basal diet was formulated to meet the nutritional requirements of birds according to NRC (1994).

All the hematological parameters evaluated : Pack cell volume (PCV), Haemoglobin (Hb), White blood cell (WBC), Red blood cell (RBC), Mean corpuscular haemoglobin concentration (MCHC), Mean corpuscular volume (MCV), Mean corpuscular haemoglobin (MCH) were not significantly(P>0.05) affected by the inclusion of ATMP. However, there were no significant (P>0.05) differences among all treatment groups in the values of total protein, albumin, globulin, alkaline phosphatase, uric acid, creatinine, SGPT and SGOT throughout the experiment which lasted for 90days. It was concluded that Azolla –turmeric mixture could be added up to 15% in the diet growing cockerels without any adverse effects on their blood profile.

INTRODUCTION

Poultry production in Nigeria is faced with a lot of problems, one of which is high feed cost. According to Ekenyem (2001), feed cost represents about 70-80% of the total cost of production. The increase in the prices of conventional feedstuffs like soya meal, fishmeal and maize which are highly used in the production of livestock feed, flour milling, oil industries as well as human consumption is as a result of the competition between humans and animals for grains and upward increase in human population. Profit cannot be maximized unless birds are well fed with formulated diets at reasonable cost to meet up with their nutritional requirement, therefore there is need to look for cheaper alternative sources of feed ingredients to feed livestock. For instance, Azolla is unconventional feed that can be used as a plant protein source in poultry nutrition. According to Alalade and Iyayi (2006) their leaves are small, sensitive with different colours. Azolla is a water fern belonging to the family Azollaceae and order pteridophyta, it is rich in essential amino acids, protein, vitamins and minerals like calcium, phosphorus, potassium, magnesium, copper and iron and can therefore be advantageously used for animal fodder.

Turmeric (*Curculum longa*) is a tropical plant which contains high biologically active compounds such as curcumin, bismethoxycurcumin and dimethoxycurcumin. Turmeric powder is a bright yellow powder made from dry grinding of turmeric rhizomes. According to Chattopadhyay et al (2004), turmeric contains 6.3% protein, 5.1% fat, 13.1% moisture, 69.4% carbohydrates and 3.5% minerals. Its yellow pigment (Curcumin) has a high therapeutic value; several reports have shown that 2-5% turmeric is curcumin (Agarwal et al, 2001) and it's relatively rich in starch (Mangala and Mathew, 1986). Curcumin are excellent source of phenolic compounds, ascorbic and carotenoids which have been reported to show good antioxidant activity (Huda-faujan et al, 2009; M. Kamal et al, 2014).

According to Kurtoglu et al (2005); Ajao et al (2013), nutrition or dietary contents affect the blood profile of healthy animals. Heamato-biochemical analyses are used for nutritional studies for animals (Onyeanusi, 2007); Church et al (1984); Olabanji et al (2007). Hematological values could serve as baseline information for comparisons of nutrient deficiency (Etim et al (2014a); Daramola et al (2005). According to Azzez et al (2009); Yanagita et al (2011); Adeyemo et al (2010) factors such as age, sex, nutrition, environmental condition and management could cause variation in haemato-biochemical parameters. Blood examination gives a clear picture to determine the presence of metabolites and other constituents in the body. Therefore, this experiment was carried out to investigate the haematological and some serum biochemical parameters of Isa cockerels fed different levels of Azolla-turmeric mixture.

MATERIALS AND METHODS

Site of the experiment

The study was carried out at the Poultry unit of Dan-malafia Farms, Ibadan, Nigeria.

Preparation of experimental diets

Fresh Azolla was harvested from the propagation pond within the farm premises and washed thoroughly to remove residues, it was later placed on flat metal trays and sun dried for 5 days until the leaves turns brown and crumbles when squeezed. It was then milled into Azolla powder.

The turmeric rhizome (dry roots) were collected, sun-dried for 8 days and milled to produce turmeric powder. Thereafter, Azolla-Turmeric (ATMP) was mixed together in the ratio of 1:1 to formulate the diet along with other ingredients purchased from a reputable feed mill in Ibadan.

All the experimental diets were formulated to meet the nutritional requirement of birds according to NRC (1994) as presented in Table 1. The proximate composition of Azolla and Turmeric powder is presented in Table 2 and 3 respectively.

Treatment 1- Control 0% Azolla-Turmeric mixture (Basal diet)
Treatment 2- Basal diet + 5% Azolla – Turmeric mixture
Treatment 3- Basal diet + 10% Azolla-Turmeric mixture
Treatment 4- Basal diet + 15% Azolla-Turmeric mixture

Animals and their management

A total of One hundred and twenty, four weeks old Isa white cockerels were randomly distributed into four (4) groups of 30 birds. Each group was further subdivided into 3 replicates with 10 birds per each in a Completely Randomized Design. A deep litter poultry house was used; the house was cleaned and well disinfected before the commencement of the experiment. Vaccines were administered according to the prevailing vaccination schedule in the environment. Feed and water were offered ad-libitum. The light was continuous throughout the experimental period that lasted for 90days.

Blood Analysis

At day 90, three birds were randomly selected from each replicate for hematological and serum analysis. Samples meant for hematology were collected into bottles containing Ethylene diamine tetra acetic acid (EDTA) as anticoagulant, while that of serum analysis was free from anticoagulant. Blood samples were taken from the brachial vein, the hematological parameters analyzed

included Pack cell volume (PCV), hemoglobin concentration (Hb), Red blood cell (RBC), white blood cell (WBC) and its differential counts (Eosinophils, monocytes, lymphocytes and neutrophils), Mean corpuscular volume (MCV), Mean corpuscular haemoglobin (MCH), Mean corpuscular haemoglobin concentration (MCHC). PCV, RBC, WBC and Hb were measured according to Dein (1984). MCV, MCH and MCHC were recorded according to Ritchie et al (1994).

Serum total protein, Albumin, Globulin, Uric acid, Creatinine and Blood glucose were determined using Scott (1965), Alkaline phosphatase, Serum glutamic oxalocaetate transaminase (SGOT) and Glutamic phosphate transaminase (SGPT) were recorded according to Reitman and Frankel (1957).

Statistical Analysis

All data collected were subjected to one way analysis of variance (ANOVA) using SAS 1994 and significant means separated by Duncan multiple range tests. (Duncan, 1955).

Table 1: Composition of experimental diets (%)

Ingredients	Diets			
	1	2	3	4
Maize	55.00	55.00	55.00	55.00
Wheat offal	26.00	26.00	26.00	26.00
Soya meal	10.00	10.00	10.00	10.00
Ground nut cake	5.00	4.80	4.60	4.40
Limestone	1.00	1.00	1.00	1.00
Bone meal	2.00	2.00	2.00	2.00
Lysine	0.25	0.25	0.25	0.25
Methionine	0.20	0.20	0.20	0.20
Premix*	0.25	0.25	0.25	0.25
Salt	0.30	0.30	0.30	0.30
Tridax meal	-	0.20	0.40	0.60
Total	100.00	100.00	100.00	100.00
Chemical composition				
Crude protein	18.18	18.70	18.87	18.97
ME (Kcal/kg)	2804.1	2820.5	2821.4	2821.6

*Premix supplied per kg diet :- Vit A, 10,000 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.

Table 2: Proximate Composition of Turmeric Powder

Parameter	Dry matter (%)
Moisture	9.57
Crude protein	9.14
Ether extracts	2.40
Total ash	8.69
Crude fibre	4.10
NFE	69.10

NFE: Nitrogen Free Extracts

Table 3: Proximate Composition of Azolla Powder

Parameter	Dry matter (%)
Crude protein	22.51
Ether extracts	3.53
Total ash	14.13
Crude fibre	18.15
NFE	37.88

NFE: Nitrogen Free Extracts

Table 4: Hematological parameters of growing Cockerels fed diets supplemented with varying levels of ATMP

	Treatments				
Parameters	1	2	3	4	SEM
Pack cell volume (%)	30.01	31.13	30.77	30.76	1.10
Hb (g/dl)	9.84	10.22	10.36	10.44	0.06
Red blood cell count ($\times10^{6/ml}$)	3.31	3.19	3.10	3.02	0.18
White blood cell count ($\times10^{6/ml}$)	4.14	4.89	4.53	4.94	1.03
MCV (fl)	101.2	118.1	102.1	106.7	2.88
MCH (pg)	34.09	34.90	34.99	34.09	0.51
MCHC (g/dl)	33.19	34.16	34.87	34.89	0.66
Neutrophils (%)	3.11	3.27	3.38	3.41	0.50
Lymphocytes (%)	40.08	41.72	47.01	48.13	1.70
Eosinophils (%)	4.23	4.36	4.40	4.57	0.33
Monocytes (%)	3.26	3.34	3.37	3.41	0.17

Hb: Haemoglobin
MCV: Mean corpuscular volume
MCH: Mean corpuscular haemoglobin
MCHC: Mean corpuscular haemoglobin concentration

Table 5: Serum biochemistry of growing Cockerels fed different diets supplemented with ATMP

	Treatments				
Parameters	1	2	3	4	SEM
Albumin (g/dl)	2.11	2.05	2.10	2.06	0.48
Globulin (g/dl)	1.01	1.10	1.08	1.07	0.13
Total protein (g/dl)	3.12	3.15	3.18	3.13	0.01
Uric acid (mol/dl)	5.01	5.07	5.09	5.02	0.14
Creatinine (mol/dl)	3.07	3.08	3.14	3.03	1.01
ALP (iu/l)	56.01	53.12	50.44	51.66	1.47
SGPT (U/l)	31.07	32.11	31.50	31.53	0.05
SGOT (U/l)	30.23	30.91	31.46	30.41	0.11

SGPT: Serum glutamic phosphatase transaminase
SGOT: Serum glutamic oxaloacetate transaminase
ALP: Alkaline phosphatase

RESULTS AND DISCUSSION

Table 1 shows the percentage composition of experimental diet. The value of the crude protein ranges between 18.18% and 18.97% while the energy level is between 2804.1 and 2821.6 (ME Kcal/kg). The basal diet was formulated to meet the nutrient requirements of birds according to NRC (1994).

The proximate composition of turmeric is shown in Table 2. The proximate components of turmeric are 9.57%, 9.14%, 2.40%, 8.69%, 4.10% and 69.10% for moisture, crude protein, ether extract, ash, crude fibre and nitrogen free extract, the results obtained agrees with the findings of Ikepeama *et al* (2014) and Alagbe, J.O (2016) on the chemical composition of turmeric. Table 3 reveals the proximate composition of Azolla powder, the proximate content of Azolla contains 22.51%, 18.15%, 3.53%, 37.88% and 14.13% for crude protein, crude fibre, ether extract, nitrogen free extract and total ash. The values recorded in this study were within the range reported by Alalade and Iyayi (2006); Alagbe, J.O (2017).

Table 4 shows the Hematological parameters of growing Cockerels fed diets supplemented with varying levels of ATMP. Pack cell volume (PCV) values obtained are 30.01%, 31.13%, 30.77% and 30.76% for treatments 1, 2, 3 and 4 respectively while those of Haemoglobin (Hb) are 9.84, 10.22 ,10.36 and 10.44 (g/dl) for treatments 1, 2, 3 and 4. The values obtained for Red blood cell (RBC) are 3.31, 3.19, 3.10 and 3.02 ($\times10^{6/}$ml) for treatments 1, 2, 3 and 4 respectively while those of MCV are 101.2, 118.1, 102.1 and 106.7 (fl) for treatment 1, 2, 3 and 4 respectively. MCH values obtained are 34.09, 34.90, 34.99 and 34.09 (pg) for treatments 1, 2, 3 and 4 respectively while those of MCHC are 33.19, 34.16, 34.87 and 34.89 (g/dl) for treatments 1, 2, 3 and 4. Pack cell volume (PC V) , Heamoglobin (Hb), red blood cell (RBC), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) were

not significantly (P>0.05) different among the dietary treatments. However, all values fall within the range reported by Talebi *et al* (2005); Kececi and Col (2011); Wiki vet (2013) and Abdi-Hachesoo *et al* (2011) of chicken. According to Togun *et al* (2007) physiological and nutritional status of animals could cause differences in haematological parameters.

Campbell and Lasley (1975) also reported that when the haematological parameters were within the normal range, it is a clear indication of adequate nutritional status. Although, the PCV and Hb values slightly increases as the level of ATMP increased though not at a significant level. High level of Hb and RBC is an index of efficient oxygen and carbon dioxide transportation within the body of the animal, PCV are also involved in the transport of absorbed nutrients. According to Togun *et al* (2007) an increase in PCV and RBC level shows more efficient erythropoiesis in animals. MCV, MCH and MCHC vital parameters used to ascertain anaemic conditions in the experimental animals (Saleh *at al*., 2014).

White blood cells values obtained are 4.14, 4.89, 4.53 and 4.94 ($\times 10^{6}$/ml) for treatments 1, 2, 3 and 4 respectively while the Neutrophils values are 3.11%, 3.27%, 3.38% and 3.41% for treatments 1, 2, 3 and 4 respectively. Lymphocytes values obtained are 40.08%, 41.72%, 47.01% and 48.13% for treatments 1, 2, 3 and 4 respectively. White blood cells (WBC), lymphocytes, neutrophils, monocytes and eosinophils were not significantly (P>0.05) influenced by the different inclusions of ATMP, their values slightly increased from treatment 1 to 4 but fall within the normal range reported by Ibrahim Albokhadaim (2012) on the haematological values of indigenous chicken. According to Ameen *et al* (2007) ; Adenkola and Durotoye (2004) a normal range of WBC, lymphocytes, monocytes, neutrophils and eosinophils shows that the feeding pattern did not affect the immune system of the birds.

The results of the serum biochemical analysis (Table 5) showed that total protein values obtained are 3.12, 3.15, 3.18 and 3.13 (g/dl) for treatments 1, 2, 3 and 4 respectively, while the albumin values are 2.11, 2.05, 2.10 and 2.06 (g/dl) for treatments 1, 2, 3 and 4. The values obtained for globulin in g/dl are 1.01, 1.10, 1.08 and 1.07 for treatments 1, 2, 3 and 4 respectively. Albumin, globulin and total protein were not (P>0.05) significantly different among the dietary treatments. The values for all parameters fall within the normal range established for birds by Swenson (1970); Olorede and Longe (2002) for birds. This observation reveals that the quality of protein in the experimental diet is enough to support normal protein reserves across the treatment. Uric acid values obtained in mol/l are 0.31, 0.37, 0.39 and 0.32 for treatments 1, 2, 3 and 4 respectively while those of creatinine are 0.77, 0.78, 0.79 and 0.73 (mol/l) for treatments 1, 2, 3 and 4 respectively. ALP values obtained are 56.01, 53.12, 50.44 and 51.66 (iu/l) for treatments 1, 2, 3 and 4 respectively. Uric acid, creatinine and ALP were not significantly affected (P >0.05) by the dietary inclusion of ATMP. The uric acid and creatinine values slightly increased from treatments 1 to 3 after which the values declined. According to Ibrahim Albokhadaim (2012), uric acid is major product from nitrogen metabolism. The uric acid and creatinine values agree with the reports of Bolu *et al* (2009) on the serum biochemistry of broiler chickens but contrary to the reports of Saleh *et al* (2014) on biochemical indices of rabbits fed graded levels of browse forage. The ALP values (50.44 – 56.01 iu/l) were contrary to the (69.25-79.00 iu/l) reported by Bolu et al (2009) for broiler chickens.

The SGPT values obtained are 31.07, 32.11, 31.50 and 31.53 (U/l) for treatments 1, 2, 3 and 4 respectively while those of SGOT in (U/l) are 30.23, 30.91, 31.46 and 30.41 for treatments 1, 2, 3 and 4 respectively, the values were not significantly (P>0.05) influenced by the different inclusion of ATMP. The SGPT and SGOT values of 31.07 - 31.53 (U/l) and 30.23 – 31.46 (U/l) were comparable to 33.03 – 36.99 (U/l) and 30.38 – 33.58 (U/l) respectively reported by Tyas *et al* (2013) but contrary to the reports of Alagbe, J.O (2016) on the effects of feeding varying levels of tiger nut on the blood profile of grass cutters. According to Iyayi (1994) SGPT and SGOT are mostly influenced by the presence of anti-nutrients in an experimental diet, this observation suggests that the animals were able to tolerate the anti-nutrients in turmeric. Alagbe, J.O (2017) reported the presence of saponin, alkaloid, tannin, sterol, phenol and flavenoid at 1.03%, 0.66%, 1.01%, 0.02%, 0.72% and 0.38% respectively during the phytochemical analysis of turmeric powder.

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