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Performance and haematoserological influence of black and alligator peppers and their composite mix in broiler chickens

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ABSTRACT

Background: The experiment was carried out to assess the influence of varying inclusions of black pepper, alligator pepper and their composite mix on the performance, haematology and serum biochemical components of broiler chickens.
Methods: Two hundred and ten (210) day – old mixed sex Cobb 500 broiler chicks were allocated to diets containing 0.25 and 0.50 g/kg each of black pepper, alligator pepper and their composite mix tagged treatments: B and C, D and E, F and G representing 0.25 and 0.50 g/kg black pepper, alligator pepper and the composite mix respectively. Treatment A was the control/basal diet. Each treatment was further divided 3 times with 10 birds per representative group. The birds received the experimental diets and water ad – *libitum* for 6 weeks (42 days). The initial and final weight gain as well as the feed intake were captured and recorded to determine the feed efficiency. At 42nd day, 5 birds per experimental sub-group were selected randomly and blood collected from the jugular veins for determination of haematological and serum biochemical indices.
Results: The total weight gain, feed intake and feed conversion ratio of the broilers fed black and alligator peppers and their composite were significantly ($P < 0.05$) better than the bird on the control diet. However, the phyto-additive did not have a significant ($P > 0.05$) influence on the haematological parameters of the birds when compared with the control. There was a significant ($P < 0.05$) reduction in the serum cholesterol and blood alkaline phosphatase concentrations while the serum total protein was significantly ($P < 0.05$) increased.
Conclusion: Overall, the phytogens individually improve the performance and haematoserological components of the birds but the composite mix had better effect on the performance and serological indices of the broilers.

INTRODUCTION

The availability of various medicinal plants consumed as spices in sub-Saharan African countries is abundant. Among these plants are black pepper (*Piper guineense)* and alligator pepper (*Aframomum melegueta*). Black and alligator peppers are reported to be rich in both medicinal and nutritive values (Nosiri et al., 2013, Uhegbu et al., 2015a). The suitability of these herbs as medicinal plants is not farfetched from their rich phytochemical components such as alkaloids, glycosides, tannins, flavonoids, sterols, triterpenes, and oils which are responsible for their therapeutic and antimicrobial properties (Okigbo and Igwe, 2007, Doherty *et al*., 2010). Previous studies have highlighted the potentials of the seeds of these spices or their extracts in alternative medicine. They possess potent antioxidant (Onoja et al., 2014), antibacterial (Ilic et al., 2010), antidiarrheal (Umukoro and Ashorobi, 2003), anti-inflammatory (Umukoro and Ashorobi, 2005), antihypercholesterolemic and blood modulator effects (Adefegha et al., 2017).

Considering the recent concerns on the extensive use of antibiotics in broiler production (Adu and Olarotimi, 2020), dietary inclusion of black and alligator peppers could provide a potent alternative in the management of diseases as well as enhancing the performance of broiler chickens. Since the blood profiles of an animal forms the basis for determining its health status and this is directly linked with performance, the effects of the diets on the haematogical and biochemical indices, therefore, plays a pivotal role in the overall well being and the productive performance of the animal. For instance, the dietary inclusion of alligator pepper meal was discovered to significantly promote weight gain and recommended for sustainable fish production (Kwankwa et al., 2020). Effiong and Ochagu (2019) also reported a significant increase in daily weight gain and feed conversion ratio of broiler chickens fed diets containing 0.4 % black pepper seed meal. In another study, Al-Kassie et al. (2011) included 0.50 to 1.00% of black pepper in broilers' diets and recorded an improved body weight gain, feed intake and conversion ratio. They also observed a reduced cholesterol, Hb, RBC and H/L ratio concentrations.

On the other hand, black pepper extracts administered to albino rats at 25 - 75 mg/kg BW was reported to significantly increase the serum proteins and packed cell volume concentration while serum cholesterol was significantly reduced (Uhegbu et al., 2015b). The effects of black pepper on serum enzymes were also recorded by Uhegbu et al. (2015a). They observed significant reductions in alanine transaminase, aspartate transaminase and alkaline phosphatase in female albino rats given aqueous seed extract of black pepper at 5 ml/BW. The red blood cells, white blood cells and hemoglobin concentration of the rats were significantly increased. Nwozo et al. (2013), in another their report, also observed a significant decrease in the elevated levels of serum alanine transaminase and aspartate transaminase activities of Wistar rats administered 200 to 400 mg/kg BW of alligator pepper. The hypoglycemic effect of alligator pepper was also documented in previous studies (Akhani et al., 2004; Al-Amin et al., 2006; Ilic et al., 2010). The safety of black and alligator peppers was already assured as they did not have adverse effects on the hematology and blood coagulation, blood pressure, or heart rate of the animals treated (Weidner and Sigwart, 2000). The aim of the present study is to examine the individualistic and synergistic effects of these herbal additives on the performance, haematology and serum biochemistry of broiler chickens.

MATERIALS AND METHODS

Both black and alligator pepper seeds were sourced and identified at the University's herbarium. The seeds were separated carefully from the twig and all other extraneous materials. The seeds were air-dried under a shade till a moisture content of 10 -11 % was achieved. Thereafter, the dried seeds were pulverized into powder to make black pepper seed powder (BPSP) and alligator pepper seed powder (APSP) using an electric blender. A total of two hundred and ten (210) day-old mixed-sexed Cobb 500 broiler chicks were purchased from a reputable hatchery and used for the experiment at the Poultry Unit of the Teaching and Research Farm, The Federal University of Technology, Akure. On arrival, they were weighed and randomly assigned to seven (7) treatments comprising treatments A (control), B (basal + {0.25 g BPSP/kg}), C (basal + {0.50 g BPSP}/kg), D (basal + {0.25 g APSP}/kg), E (basal + {0.50 g APSP}/kg), F (basal + {0.125 g BPSP + 0.125 g APSP}/kg) and G (basal + {0.25 g BPSP + 0.25 g APSP}/kg). Each treatment was repeated 3 times with 10 birds per replication in a completely randomized design. Basal broiler starter and finisher diets (Table 1) were formulated and fed to the birds at 0–3 weeks and 4-6 weeks, respectively. The respective total weight gain and feed intake of the birds were calculated as summations of the weekly weight gains and feed intake throughout the experimental period. The feed conversion ratio (FCR) was determined as:

$$FCR = \frac{\text{Total Feed Intake (g)}}{\text{Total Weight Gain (g)}}$$

On the last day of the experiment (42nd day), five (5) birds per replicate were randomly selected and fasted overnight. Blood samples were collected through jugular veins puncture and blood was collected into bottles with EDTA for determination of haematological indices. Another set of blood samples were collected into dry clean plain centrifuged glass tubes for determination of serum biochemical parameters. Serum samples were harvested after plain blood samples were centrifuged for

10 minutes at 3000 rpm. The serum samples were used for the determination of serum enzymes [alanine transaminase (ALT), aspartate transaminase (AST) and alkaline phosphatase (ALP)], cholesterol and proteins [total protein (TP), globulin (GLB) and albumin (ALB)]. The haematological indices determined were packed cell volume (PCV), haemoglobin concentration (Hb), red blood cell counts (RBC), Mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) and white blood cell counts (WBC). The WBC differential counts (granulocytes, lymphocytes and monocytes) were equally determined.

Table 1: Ingredients Composition of the Experimental Diets

Ingredients	Starter	Finisher
Maize	51.00	50.00
Soybean meal	15.00	13.00
Groundnut cake	15.00	13.00
Fish Meal (72%CP)	5.00	0.00
Rice Bran	0.00	13.30
Corn bran	10.60	6.00
Bone Meal	1.50	2.50
Limestone	1.00	1.00
Salt	0.35	0.35
Lysine	0.10	0.30
Methionine	0.20	0.30
Broiler Premix	0.25	0.25
Total	100.00	100.00
Calculated Nutrients		
ME (Kcal/Kg)	**2985.82**	**2964.16**
Crude Protein (%)	**22.60**	**18.56**
Calcium (%)	**1.28**	**1.24**
Phosphorus (%)	**0.52**	**0.56**
Lysine (%)	**1.15**	**1.08**
Methionine (%)	**0.56**	**0.57**
Crude Fibre (%)	**3.68**	**4.85**

Statistical Analyses

All data collected were subjected to One-Way analysis of variance (ANOVA) using SAS (2008) version 9.2. Duncan Multiple Range Test of the same software was used for means comparison where significant differences existed.

RESULTS AND DISCUSSION

The results of the performance of the birds fed different experimental diets are in Table 2. There was an increase in the total weight gain (TWG) among the birds fed diet containing 0.25 g/kg BPSP (Diet B) though not significantly ($P > 0.05$) different from the weights of the birds on the control diet (Diet A). However, doubling the inclusion rate of BPSP to 0.50 g/kg diet (Diet C) significantly ($P < 0.05$) increased the TWG of the birds when compared with the birds on the control diet. Furthermore, the inclusions of 0.25 and 0.50 g/kg APSP significantly ($P < 0.05$) increased the TWG among the birds on diets D and E when compared with the birds on the control diet. The composite mix of BPSP and APSP at 0.25 and 0.50 g/kg equally significantly ($P < 0.05$) increased the TWG of the birds when compared with the broilers on the control diet. The TWG recorded among the birds fed the composite mix was far better than the weights by the birds on all other diets with birds fed 0.50 g/kg BPSP-APSP mix recording the higher significant ($P < 0.05$) means when compared with birds on either of the phyto-additives. The total feed intake (TFI) of the birds on the two phyto-additives were significantly ($P < 0.05$) higher than that of the birds on the control diet. However, the varying inclusions of the composite mix in the diet further enhanced the TFI of the birds on diets F and G with the broilers on diet G recording the higher significant ($P < 0.05$) TFI when compared with birds on other diets. For the feed conversion ratio (FCR), the phyto-additives positively influenced the FCR. The inclusion of 0.25 g/kg BPSP did not significantly ($P > 0.05$) enhance the FCR unlike the inclusion of 0.50 g/kg where a significant ($P < 0.05$) reduction in FCR was recorded. Both the inclusions of 0.25 and 0.50 g/kg APSP in the diet significantly ($P < 0.05$) improved the FCR of the birds when compared with the control. However, broilers on diets F and G (0.25 and 0.50 g/kg composite mix) presented the best ($P < 0.05$) FCR.

The improved TWG, FI and FCR recorded among the birds fed the varying inclusions of the phytogens could be as a result of the antioxidant properties of black and alligator peppers. Antioxidants have been credited with having the potentials to positively enhance the performance of poultry birds (Biswas et al 2011). The improved TWG, FI and FCR among the birds fed diets containing varying inclusions of the phytogens and their composite indicated that broilers were able to covert feed to muscles when fed diets supplemented with phytogenic additives. Weight gain, feed intake and feed conversion ratio are usually the most reliable yardsticks considered in the measurement of the productivity of diets fed to broiler chickens. The enhanced performance recorded in all treatments was indicative that broilers would perform well on diets containing black and alligator peppers at the inclusion rates of 0.25 to 0.50 g/kg diet. It also indicated that the best performance could be obtained by feeding the composite mix of the two phytogens as they conferred synergistic effects on the birds. The excellent performance recorded by the individual phytogen and their composite could be due to their rich bioactive components such as tannin, alkaloid, phytate and saponins. These phytochemicals were previously explained to increase the growth performance of animals (Enemor *et* al., 2014). The finding in the present study justified the claims of previous studies which documented improved performances in different classes of livestocks fed black and alligator peppers (Al-

Kassie et al., 2011, Effiong and Ochagu, 2019, Kwankwa et al., 2020).

The results of the haematological indices of the broilers fed different levels of the two phytogenic compounds and their composite are in Table 3. From this study, the haemogram, white blood cell counts and the differentials were not significantly ($P > 0.05$) influenced by the inclusions of the phytogens and their composite. The serum biochemical indices' results of the broilers are equally displayed in Table 4. It was observed that the serum cholesterol concentration of the birds on the control diet was significantly ($P < 0.05$) higher than the cholesterol concentrations recorded by the birds on all other diets except diet B where a non-significant ($P < 0.05$) reduction in cholesterol concentration was recorded. For the serum enzymes, the inclusions of varying amounts of the phytogens significantly ($P < 0.05$) lowered serum concentration of ALP except among the birds fed diet containing 0.25 g/kg BPSP where there was no statistical ($P > 0.05$) difference between their ALP concentration and that of the birds on the control diet. The serum total protein concentrations of the birds fed varying inclusions of the phytogenic compounds were significantly ($P < 0.05$) enhanced when compared with the birds on the control diet. However, the globulin and albumin concentrations of the birds were not significantly ($P > 0.05$) different from what were recorded by the control birds.

The non-significant influence recorded in the haematological indices among birds fed the varying inclusions of the phytogens and their composite indicated that the health status of the birds was not compromised. The various haematological indices studied were within their standard haemtological reference values range. For instance the haematological values recorded in this study were within the reference values of RBC ($2.0 – 4.0 \times 10^6 mm^3$), Hb (7.0 – 13.0 g/dl), PCV (25 – 45%), MCV (90 – 140 fl), MCH (33 – 47 pg) and MCHC (26 – 35 g/dl) (Harrison and Lightfoot, 2005). Furthermore, the consistence in white blood cells (WBC) differential counts recorded across all the treatment diets in this study showed that the dietary inclusions of the phytogens and their composite had no significant effect on lymphocyte, monocytes, heterophil, eosinophil, and basophil when compared with birds on the control diet. This indicated that the bird's immunity was not compromised but rather enhanced. The present study gave credence to the fact that black and alligator peppers and their composite were safe as phyto-additives in broiler nutrition as previously stated that they did not have adverse effects on the hematology of farm animals (Weidner and Sigwart, 2000).

The increased transaminase in the bloodstream is always linked with hepatocellular damage. Hence, the insignificant differences recorded in ALT and AST in this study pointed to the fact that the phytogens and their composite at the inclusion rates used were not inimical to the livers of the birds. However, the significant reduction in the serum ALP observed indicated that these phytogens and their composite could be useful in lowering increased blood concentration of the enzyme which pointed to the therapeutic effect of these phytogens. This study confirmed the findings of Nwozo et al. (2013) and Uhegbu et al. (2015a) who documented a significant reduction in the elevated levels of serum transaminase activities of animals administered alligator pepper and black pepper respectively. The hypoglycemic effect of these phytogens and their composites were clearly illustrated in this study as the birds on the control diet had the highest serum cholesterol. This agreed with previous reports on the hypoglycemic potentials of both black and alligator peppers (Uhegbu et al., 2015b, Ilic et al., 2010). The non-significant difference observed in the GLB and ALB in this study indicated that the inclusion of the phytogens and their composite did not interfere with protein metabolism in the liver of the birds. However, the increased total protein was suggestive that inclusion of these phytogens enhances protein synthesis in the liver of the animals which agreed with the report of Uhegbu et al. (2015b).

Table 2: Performance of Broilers fed Diets Containing BPSP, APSP and their Composite

Parameters	A	B	C	D	E	F	G	SEM	P-Values
IBW	33.00	33.12	33.08	31.99	34.20	33.09	33.14	1.00	0.19
TWG	2600^{c}	2800^{bc}	2900^{b}	2958^{b}	2890^{b}	3270^{ab}	3490^{a}	110	0.03
TFI	3500^{c}	3600^{b}	3600^{b}	360^{ab}	3611^{b}	3635^{ab}	3650^{a}	140	0.01
FCR	1.35^{a}	1.29^{ab}	1.24^{b}	1.22^{b}	1.25^{b}	1.11^{c}	1.05^{c}	0.06	0.01

Means in a row without a common superscript letter differ ($P<0.05$) significantly. IBW = Initial Body Weights, TWG = Total Weight Gain, TFI = Total Feed Intake, FCR = Feed Conversion Ratio. Diets: A (Control/Basal), B (Basal + 0.25 g BPSP/kg diet), C (Basal + 0.50 g BPSP/kg diet), D (Basal + 0.25 g APSP/kg diet), E (Basal + 0.50 g APSP/kg diet), F (Basal + 0.25 g BPSP-APSP Mix/kg diet), G (Basal + 0.50 g BPSP-APSP Mix/kg diet).

Table 3: Haematology of Broilers fed Diets Containing BPSP, APSP and their Composite

Parameters	A	B	C	D	E	F	G	SEM	P-Value
PCV (%)	40.00	38.00	35.00	39.00	37.00	34.00	37.00	2.80	1.21
RBC ($x10^6$ mm^3)	4.00	3.51	3.92	4.50	3.60	4.11	5.02	0.83	0.56
MCHC (g/dl)	33.00	33.00	32.00	33.00	32.00	32.00	32.00	0.62	1.18
MCV (fl)	100.00	110.00	95.00	89.00	100.00	87.00	80.00	17.00	0.92
MCH (pg)	33.00	37.00	32.00	30.00	35.00	29.00	27.00	5.64	1.51
Hb (g/dl)	13.00	13.00	12.00	13.00	12.00	11.00	12.00	0.94	0.35
WBC ($x10^9$/l)	2.91	2.90	3.11	2.52	3.90	2.33	2.11	0.91	0.26
GRA ($x10^9$/l)	0.95	1.04	1.21	0.85	1.32	0.90	0.89	0.32	1.45
LYM ($x10^9$/l)	1.90	1.90	1.88	1.64	2.51	1.44	1.23	0.66	1.69
MON ($x10^9$/l)	0.04	0.02	0.04	0.03	0.05	0.02	0.02	0.02	1.11

Values are means and SEM (Standard Error of Means). Means in a row without a common superscript letter differ significantly (P<0.05). PCV = Packed Cell Volume, RBC = Red Blood Cells, MCHC = Mean Corpuscular Haemaglobin Concentration, MCV = Mean Corpuscular Volume, MCH = Mean Corpuscular Haemoglobin, Hb = Haemoglobin, WBC = White Blood Counts, GRA = Granulocytes, LYM = Lymphocytes, MON = Monocytes. Diets: A (Control/Basal), B (Basal + 0.25 g BPSP/kg diet), C (Basal + 0.50 g BPSP/kg diet), D (Basal + 0.25 g APSP/kg diet), E (Basal + 0.50 g APSP/kg diet), F (Basal + 0.25 g BPSP-APSP Mix/kg diet), G (Basal + 0.50 g BPSP-APSP Mix/kg diet).

Table 4: Serum Biochemical Components of Broilers fed Diets Containing BPSP, APSP and their Composite

Parameters	A	B	C	D	E	F	G	SEM	P-Values
Chol	6.84^a	5.40^{ab}	3.55^c	4.18^b	3.52^c	3.16^c	3.05^c	0.72	0.01
ALP	2200^a	2200^a	2100^b	1700^c	1200^d	1600^c	1300^d	620	0.01
ALT	32	32	31	33	32	31	33	1.8	0.95
AST	120	110	120	120	120	110	120	12	1.23
TP	33^b	36^a	36^a	36^a	37^a	36^a	37^a	4.5	0.03
ALB	12	12	10	8.7	10	12	12	3.4	0.43
GLB	22	24	26	28	26	24	25	4.7	0.11

Values are means and SEM (Standard Error of Means). Means in a row without a common superscript letter differ significantly (P<0.05). Chol. (Cholesterol), Alkaline phosphatase (ALP), Alanine transaminase (ALT), Aspertate transaminase (AST), TP (Total Protein), ALB (Albumin), GLB (Globulin). Diets: A (Control/Basal), B (Basal + 0.25 g BPSP/kg diet), C (Basal + 0.50 g BPSP/kg diet), D (Basal + 0.25 g APSP/kg diet), E (Basal + 0.50 g APSP/kg diet), F (Basal + 0.25 g BPSP-APSP Mix/kg diet), G (Basal + 0.50 g BPSP-APSP Mix/kg diet).

CONCLUSIONS

This study concluded that the varying inclusions of black pepper, alligator pepper and their composite had significant enhancement potentials on the performance of broiler chickens without any adverse effect on the blood profiles of the animals indicating healthy performance. The phyto-additives also had the potential of reducing the cholesterol level of the birds thereby providing lean meat which is healthy to the consumers. The composite mix of these phytogens are therefore recommended at the 0.25 to 0.50 g/kg inclusion rate as the synergistic effect of the composited proved to be of better performance than the individualist effect of each phytogen.

Competing interests

The author declares no competing interest.

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