

Effect of Dietary Supplementation of *Piper guineense* Seed (African Black Pepper) and *Curcuma longa* (Turmeric) Powder on Performance, Hematological and Biochemical Indices of Finisher Broiler Chickens

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Accepted November 22, 2023

In a quest to achieve optimum output in poultry products through the use of organic livestock production with natural spices in our locality as phytogetic feed additives, this study was conducted to determine the effect of *Piper guineense* (PG) seed and *Curcuma longa* (CL) powder on performance, hematological and biochemical indices of finisher broiler chickens. A total of 270 one-day-old unsexed broiler chicks (Cobb 500) were raised on litter-floored pens and had ad libitum access to water and feed for 56 days. The birds were randomly allocated to nine dietary treatments with 10 birds per treatment replicated 3 times laid out in a 3 by 3 factorial in a completely randomized design (CRD) having three dietary inclusions of *Piper guineense*, *Curcuma longa* and a combination of (*Piper guineense* + *Curcuma longa*) at inclusion levels of 0, 2 and 4 %. Experimental diets were formulated such that they contained 0, 2 and 4 % *Piper guineense*, 0, 2 and 4 %, turmeric and 0, 2 and 4 % *Piper guineense* + *Curcuma longa* on a 1:1 basis, respectively. The result revealed that birds fed Turmeric and their combination (PG + CL) obtained the highest significant ($P<0.05$) final weight, weight gain and feed intake. Birds fed 4% dietary additive obtained increased final weight, weight gain, feed intake and the best Feed Conversion Ratio (FCR). All hematological parameters were not significant ($P>0.05$) except for Pack cell volume (PCV), Hb and RBC. The PCV value of birds on *Curcuma longa* and their combinations (PG + CL) was significantly ($P<0.05$) higher. Dietary additives at 4 % significantly ($P<0.05$) increased the PCV, Hb and Red Blood Cell (RBC) of the birds compared to others. The serum result of dietary additives at 4 % showed significant ($P<0.05$) higher TP, Albumin, Globulin and glucose. Significantly ($P<0.05$) higher cholesterol, triglyceride, LDL and VLDL values were obtained in birds fed 0% diet. Meanwhile, the HDL of birds at 4 % and 2 % were higher. The creatinine, ALT, ALP, AST and uric acid were not affected. In conclusion, the inclusion of different dietary additives either singly or their combination levels resulted to better growth performances and did not hamper with the health.

Keywords: Prebiotics Performance, Hematological and serum biochemical indices

INTRODUCTION

The emergence of drug-resistant side effects of antimicrobials and the harmful residual toxicity effects of drugs observed recently in both animals and man regarding the use of antibiotics in many countries have encouraged the poultry industry to venture into organic livestock production. This has facilitated the use of natural feed additives which are known as phyto-genic feed additives that can enhance productivity in livestock birds. Moreover, herbs, spices and plant extracts have been considered as alternative ingredients for the health and nutrition of poultry birds (Manan et al., 2012). They have been reported as good appetite and digestion stimulants, enhancing physiological function, promoting good health and improving body performance (Frankic et al., 2009). Puvaca et al. (2013) observed that phyto- additives in poultry nutrition have attracted more attention for their potential role as alternatives to antibiotic growth promoters. Windisch et al. (2008) revealed that herbs and spices enhanced the resistance of animals exposed to varying stress conditions and also increased the absorption of essential nutrients, thereby promoting the growth of the animal. These phyto-genic substances have been reported to exhibit antioxidant, anti-proliferate, anti-carcinogenic, anti-inflammatory, immunomodulatory, antidiarrheic, hypolipidemic, detoxifying, digestion- stimulating and flavoring properties (Grashorn, 2010). Several authors (Stanacev et al. 2011; Dhama et al., 2014, 2015) reported the enhancement of these phyto-genic substances in growth performance, carcass meat safety and quality in the animal. It also serves as immune enhancement and health protection in livestock. Among these phyto-genic feed additives are *Piper guineense* and *Curcuma longa*.

Piper guineense is a flowering plant that belongs to the family Piperaceae. It bears small drupe fruits of about five millimeters in diameter and red when fully matured. The seeds are otherwise called peppercorn when dried and are used as spice and seasoning (Dutta and Dutta, 1997) as well as preservatives. The plant has been reported to originate from India and has been rated the most important for its fruit. *Piper guineense* has been observed for its broad economic values which include culinary preservative, cosmetic and medicinal. Many Authors attested that it enhances good health and it's nutritionally safe for humans (Scott et al., 2004). The genus Piper is made up of almost 1,050 species of tropical shrubs and small trees, most of which are important as spices, flavoring agents and medicine (Mabberley, 2008).

Members with economic importance include; *P. nigrum* (black pepper) (Satyal and Setzer, 2012), *P. methysticum* (nacroctic pepper) and *P. longum* (long pepper) (Zaveri et al., 2010). The essential oils of piper species have been analyzed and reported for their biological properties (Moura Do et al., 2012). Cabuk et al. (2003) reported that the essential oil from *P. guineense* leaf and seed promotes the digestive process in birds and farm animals. According to (Platel and Srinivasan, 2000), piperin and curcumin found in *P. guineense* can also influence the activity of pancreatic digestive enzymes, such as amylase, protease and lipases, thereby enhancing the digestive system. Furthermore, piperine and curcumin promote digestion and reduce the time feed passes through the digestive tract. The use of herbs and spices as probiotics with growth-promoting properties have been reported to increase feed stability and enhance the gastrointestinal ecosystem through interference with the growth of pathogenic organisms. In addition, Windisch et al. (2008) reported that animal fed diets supplemented with herbs and species are usually more resistant when exposed to different stress conditions and also with higher absorption of essential nutrients, thereby enhancing the growth performance of the animal.

Turmeric powder (*Curcuma longa*) belongs to the family of ginger (Zingiberaceae) and is known to possess phenolic compounds (curcuminoids) that act as antioxidant and anti-inflammatory substances. Curcuminoids such as curcumin, desmethoxycurcumin and bisdemethoxycurcumin, are yellow turmeric pigments that have antioxidative, anticarcinogenic and anti-inflammatory properties (Nishiyama et al., 2005). Curcumin is the most bioactive ingredient in turmeric. It represents 3- 5% of the curcuminoids in turmeric rhizomes and is a strong phenolic antioxidant (Stankovic 2004; Jaggi 2012). Turmeric benefits in poultry nutrition include improved growth performance of broiler chickens and endogenous digestive enzyme secretion and activating immune responses and antimicrobial and antioxidant activities (Dorman and Deans, 2000; Burt, 2004; Khan et al., 2012).

With reference to the above, an effort has been made to search for available organic phyto-genic substances in our locality that can enhance growth performance without residual effects on human health however, information on the combination effects of *Piper guineense* and *Curcuma longa* as growth promoters and at higher inclusion levels are

scars. This research therefore, evaluated the effect of *Piper guineense* seed, *Curcuma longa* (turmeric) powder and their combination at higher levels on the performance, hematological, and biochemical indices of finisher broiler chickens.

MATERIALS AND METHODS

Experimental site

The experiment was conducted at the Poultry Unit, Livestock Teaching and Research Farm, Federal University Dutse, Jigawa State. Dutse is located at longitude 9.34°E and Latitude 11.76°N and has an elevation of 431.36 meters above sea level. There is usually a hot diurnal temperature and comparatively cooler at night during the last 2-3 months of the dry season which is followed by a wet season between the months of June and September, Weather and Climate (2020).

Processing of test ingredient

Dried buds of *Piper guineense* and *Curcuma longa* (as dried roots) were purchased commercially. The *Piper guineense* buds and *Curcuma longa* were both cleaned and air-dried for 24 hours prior to milling. Thereafter, they were milled individually in a kitchen blender to fine particle sizes and stored in air-tight bags until incorporation into the formulated diets at appropriate levels of inclusion.

Experimental Design and bird's management

Two hundred and seventy (270) one-day-old commercial mixed sex broiler chicks (Cobb 500) were randomly allocated to nine dietary treatments laid out in a 3 by 3 factorial arrangement of nine (9) treatments having three dietary inclusion of *Piper guineense* seed, *Curcuma longa* and combination of *Piper guineense* seed + *Curcuma longa* at inclusion levels of (0, 2 and 4 %) replicated three times with ten chicks per replicate. The experimental diets were formulated such that the diets contained 0, 2, 4 % *Piper guineense* seed, 0, 2, 4 % *Curcuma longa* and 0, 2, 4% *Piper guineense* seed + *Curcuma longa* at 1:1. Feed and clean water were supplied *ad libitum*. The birds were reared intensively on a deep litter (dried wood shavings) housing system. Normal vaccination programs and medication schedules were strictly adhered to.

Data collection and parameters measured

Growth performance

Individual BW and feed consumption in each pen were recorded every 7 days (28, 42 and 56) to calculate daily gain, feed intake and feed conversion ratio. Feed intake was determined daily by measuring the feed leftovers; body weight gain and feed conversion ratio were also computed between 28-56d of the study.

Total feed intake (g) = Total feed supplied (g)-Total feed left over (g)

Average feed intake (g/bird) = total feed intake/number of birds

Total weight gain=final weight-initial weight

Feed conversion ratio=Total feed intake (g)/ Total weight gain (g), % Mortality= Number of dead birds/Total number of stocked birds X100

Blood parameter

At 56 days of the study, blood samples were collected from 8 randomly selected birds per treatment (2 per replicate) to determine the blood profile of the birds. Blood collection was done through brachial vein puncture using needles and syringes (Frandsen et al., 1986). Each blood sample was emptied into 2 sets of well-labeled sample bottles; the sample containing anti-coagulant was used for the analysis of hematological profiles while the plain bottle without anti-coagulant was used to analyze the serum bio-chemical profiles of the birds. Hematological parameters were determined using a hematology analyzer (HA) model 6000. Analysis of the biochemical indices was conducted using the clinical chemistry semi-auto-analyzer and a commercial biochemical assay kit. Serum enzymes: alanine transaminase (ALT) and aspartate serum transaminase (AST) were analyzed using commercial kits (Qualigens India. Pvt. Ltd., Catalogue number 72201-04).

Statistical analysis

Data obtained from this study were subjected to analysis of variance using SAS (2010) to separate the main effects (*Piper guineense* inclusion, *Curcuma longa* inclusion) and interaction effect (*Piper guineense* inclusion × *Curcuma longa* inclusion). Significant means were separated using Duncan's Multiple Range Test as contained in the above statistical package.

RESULTS

Table 1 shows the basal composition of experimental diet (g/Kg). The main and interactive effects of different dietary additive levels of *Curcuma longa* (CL), *Pipper guineense* (PG) and their combination (CL +PG) on the growth performance of finisher broiler chickens are presented in **Table 2**. Birds fed *Curcuma longa* and their combination (PG + CL) obtained the highest significant ($P<0.05$) final weight, weight gain and feed intake compared to birds on PG. Birds fed turmeric and their combination had significantly ($P<0.05$) increased feed intake than those fed the PG diet. The feed conversion ratio (FCR) of the birds on different dietary additives were not significantly ($P>0.05$) different across the treatment. Although, birds on PG+ CL and PG singly recorded numerically better FCR compared to birds on control diet. Survivability was similar across the treatments.

The inclusion of the different dietary additives regardless of inclusion levels significantly ($P<0.05$) increased the final live weight, weight gain, and feed intake compared to birds on the control diet which recorded the lowest of the parameters. Birds fed different dietary additives at 4 % inclusion level obtained the best and increased final weight, weight gain, feed intake and FCR. Closely followed were birds on 2% inclusion of different additives while birds on control diet recorded the lowest values.

The interactive result in **Table 3** showed that there existed a significant ($P<0.05$) interaction effect of different dietary additives on final weight, weight gain, feed intake and FCR of the birds. Birds fed 4% different dietary additive inclusion regardless of either single or their combinations had significantly ($P<0.05$) higher values of final weight, weight gain and feed intake while the lowest values of these parameters were obtained in birds fed control diets. Closely followed were birds on 2% different dietary additive inclusion level. Generally, there was a slightly significant ($P<0.05$) interactive increase as the different dietary additive inclusion levels increased across the treatment group. Survivability was similar across the treatments.

The main and interactive effects of different dietary additives of Turmeric, *Pipper guineense* (PG), and their combination (CL +PG) on the hematological of finisher broiler chickens are presented in **Tables 4** and **5**.

Results revealed that all parameters measured were not significant ($P>0.05$) except for Pack cell

volume (PCV), Hb and RBC. The PCV value of birds on turmeric and their combinations (PG + CL) were significantly ($P<0.05$) higher compared to birds on PG singly diet. Similar trend was observed for Hb and RBC of birds fed turmeric and their combinations (PG + CL) having significantly ($P<0.05$) higher Hb and RBC while birds fed PG diets recorded the lowest values for Hb and RBC. Other parameters were not significantly ($P>0.05$) influenced by the inclusion of dietary additives. The inclusion of different dietary additives at 4% significantly ($P<0.05$) increased PCV, Hb and Red Blood Cell (RBC) of the birds compared to birds fed 2% level while birds on 0% (control diet) recorded the least of the parameters. Other parameters were not altered by the treatment imposed. MCHC value increased with the increased inclusion of different additives. Birds on 4% different dietary additive recorded the highest MCHC compared to birds on 2% while the lowest was obtained in birds fed control diet (0%).

The interaction effect of the different dietary additive inclusion levels is presented in **Table 5**. The result revealed that there existed a significant ($P<0.05$) interaction influence on PCV, Hb and RBC of the birds. Birds on 4% different dietary additives (PG, CL and their combination) recorded a significant ($P<0.05$) increased PCV. Birds on 4% level of *Curcuma longa* (CL) and their combination (PG+ CL) had significantly ($P<0.05$) higher RBC and Hb compared to other groups while the least value was obtained for the control diet. Birds on 4% CL and their combination (PG+ CL) obtained the highest Hb, closely followed were birds on 4% PG, 2% CL, 2% PG and their combinations (2% PG+ CL) while the least was recorded for birds on the control diet. Similar trend was observed for RBC where 2% PG, 2% CL and their combination (2% CL + PG) were significantly ($P<0.05$) lower compared to birds on 4% CL and their combination (4% CL + PG) while the least value was noticed in birds fed control diets. The Mean corpuscular hemoglobin concentration (MCHC) interaction revealed a significant ($P<0.05$) increase as the inclusion of the different additives increased. Birds fed 4% CL +PG recorded the highest MCHC compared to birds on 4% *Curcuma longa* which are statistically similar. Closely followed were birds on 4% PG, 2% (PG + CL), 2% CL and 2% PG while control recorded the lowest MCHC.

The main and interactive effects of different dietary additive levels of *Curcuma longa* (CL), *Pipper guineense* (PG), and their combination (CL +PG) on serum biochemical indices of finisher broiler chickens

Table 1. Gross composition (g/kg) of the experimental diets (5-8weeks)

Feed additives	CL (%)			PG (%)			CL+ PG (%)		
MSP inclusion (g/kg)	0	2	4	0	2	4	0	2	4
Ingredients									
Maize	59	59	59	59	59	59	59	59	59
SBM	17.8	17.8	17.8	17.8	17.8	17.8	17.8	17.8	17.8
GNC	8	8	8	8	8	8	8	8	8
Fish Meal	3	3	3	3	3	3	3	3	3
Wheat offal	8	8	8	8	8	8	8	8	8
Bone meal	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Oyster shell	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Lysine	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Methionine	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35
Vitamins/Minerals Premix	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Common salt	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
CL	+	+	++	-	-	-	-	-	-
PG	-	-	-	+	+	++	-	-	-
CL +PG	-	-	-	-	-	-	+	+	++
Total	100	100	100	100	100	100	100	100	100
Calculated Analyses									
ME (MJ/kg)	11.15	11.15	11.15	11.15	11.15	11.15	11.15	11.15	11.15
Crude Protein (%)	20.86	20.86	20.86	20.86	20.86	20.86	20.86	20.86	20.86
Ether Extract (%)	3.95	3.95	3.95	3.95	3.95	3.95	3.95	3.95	3.95
Crude Fiber (%)	3.18	3.18	3.18	3.18	3.18	3.18	3.18	3.18	3.18
Ether extract	3.69	3.69	3.69	3.69	3.69	3.69	3.69	3.69	3.69

Vitamin/ Minerals premix supplied per kg diet vita, 8,000iu; vit D3,1440iu; VitE,21.6mg; VitK3,2.7mg; VitB1,1.8mg; VitB2,3.6mg; VitB6,2.7mg; Niacin,21.6mg; VitB12,0.018mg; FolicAcid,0.54mg; Panthothenic acid,9.0mg; Biotin,0.036mg; Choline chloride,270mg; Zinc,27mg; Mn,108mg; Fe,18mg; I2,0.72mg; Se,0.072mg; Cu,1.44mg; Co,0.14 Turmeric, Pipper guineense (PG) and their combination (Turmeric + PG); Inclusion levels: 0, 2% = +, 4%= ++

are presented in **Tables 6** and **7**. The result revealed that there was no significant ($P>0.05$) influence of the different dietary treatments on all serum biochemical indices of the birds. Numerically, birds on *Curcuma longa* and their combination (PG + CL) recorded higher values of all the serum biochemical indices measured. All measured parameters were within the normal range for healthy birds.

The inclusion of different dietary additives at 4% revealed significant ($P<0.05$) higher TP, Albumin, Globulin and glucose compared to birds on 2% level of inclusion while birds on 0% recorded the least of

these parameters. Significantly ($P<0.05$) higher cholesterol and triglyceride, LDL and VLDL values were obtained in birds fed 0% as compared with birds on diets 2% and 4% which were statistically similar. Although 4% inclusion numerically had an increased value of these parameters. However, the HDL of birds fed 4% inclusion of different dietary additives were significantly ($P<0.05$) increased when compared to birds on 2% different dietary additives while the least was observed in birds fed control diet. The creatinine, ALT, ALP, AST and uric acid were not affected ($P>0.05$) by the treatment imposed. The

Table 2. Main effects of *Curcuma longa* (CL), *Pipper guineense* (PG) and levels of inclusion on the performance of finisher broiler chickens (5-8weeks)

Parameters	Level of inclusion of additives (%)							
	CL	PG	CL + PG	SEM	0	2	4	SEM
Initial Weight (g/bird)	1100.44	1100.44	1100.44	0	1100.44	1100.44	1100.44	0
Final weight (g/bird)	2280.52 ^a	2272.44 ^b	2286.66 ^a	21.93	2163.40	2266.40	2288.89	10.46
Weight gain (g/bird)	1180.08 ^a	1172.18 ^b	1186.22 ^a	15.82	1062.96	1165.96	1188.45	11.22
Feed intake (g/bird)	2454.84 ^a	2388.44 ^b	2421.22 ^a	22.40	2370.43	2444.42	2452.42	34.32
FCR	2.08	2.04	2.04	0.06	2370.43	2.09	2.06	0.10
Survivability (%)	100.00	100.00	100.00	0.10	98.00	100.00	100.00	0.72

^{abc} Means on the same row having different superscripts are significantly different ($P < 0.05$); FCR, feed conversion ratio.

values were similar among the treatment groups. The interactive effect revealed that all serum biochemical indices measured were not significantly ($P > 0.05$) influenced except for total protein, HDL and LDL of the birds. Birds fed 4% of CL, PG and their combination (PG+ CL) recorded a significant ($P < 0.05$) increased HDL compared to birds fed 2% Turmeric, PG and their combination (PG+ CL) while the lowest was obtained for birds on control diet. The LDL of birds on control diet was the highest compared to other treatment groups. Birds fed 4% inclusion of PG+ CL recorded the lowest LDL. Closely followed were birds on 2% PG + CL, 4% CL and 4% PG which were comparably lower to birds on 2% *Curcuma longa* diet. Both 4% and 2% inclusion of different dietary additives resulted in high significant TP. The higher the level of inclusion the better increased TP noticed irrespective of the different feed additives.

DISCUSSION

The significant increase noticed in final weight and weight gain of birds fed Turmeric and their combination (PG+CL) corroborates the work of Effiong and Ochagu (2019) who reported positive increase in final weight and total weight gain compared to control group when fed *P. guineense* to broiler chickens. Similarly, the work agreed with Anyanwu et al. (2021) who reported significant final weight in finisher broiler fed cooked *Curcuma longa*

at 2% level. The superiority noticed in the 4% inclusion of different dietary additives regardless of the type in all growth parameters showed that all the natural feed additives either singly or combined were well utilized and could enhance the digestibility of the birds for efficient and maximum physiological growth. In addition, significant slight increase occurred as the different dietary additive inclusion increased among the treatment groups. This portrays that higher dosage doesn't alter the physiological state of the animal negatively. The increased growth performance recorded for birds on CL + PG compared to PG singly regardless of the level is a reflection that there existed a great synergy between PG + CL for efficient nutrient utilization. Similarly, the inclusion of Turmeric, PG singly regardless of the level and 2% (PG+ CL) were as good as 4% (PG+ CL). This also shows that these natural phytochemical substances have traits of growth promoting agents. Cabuk et al. (2002) reported that PG seed contains essential constituent Dillapiole (44.8%) and myristicin (9.8%) which promotes digestive process in birds and other farm animals. Consequently, the piperin and curcumin present in PG and CL could have influenced the activities of pancreatic digestive enzymes such as amylase, protease and lipases, thereby aids the digestive system of the birds (Platel and Srinivasan, 2000). Moreover, the utilization of herbs and spices as phytobiotic with growth promoting properties have been proven to increase feed stability and enhance the gastrointestinal ecosystem through interference with the growth of

Table 3. Interactive effects of *Curcuma longa* (CL) , *Pipper guineense* (PG), their combination and levels of inclusion on the performance of finisher broiler chickens (5-8weeks).

	CL			PG				CL+ PG			
Parameters	0	2	4	0	2	4		0	2	4	SEM
Initial wt (g/bird)	1100.44	1100.44	1100.44	1100.44	1100.44	1100.44		1100.44	1100.44	1100.44	0
Final wt (g/bird)	2168.66 ^c	2280.66 ^b	2286.24 ^a	2168.89 ^c	2262.88 ^b	2275.50 ^b		2170.52 ^c	2275.50 ^b	2278.50 ^a	31.29
Wt gain (g/bird)	1068.22 ^c	1184.22 ^b	1185.80 ^a	1068.45 ^c	1162.44 ^b	1175.06 ^b		1070.08 ^c	1175.06 ^b	1178.06 ^a	14.47
Feed intake	2360.82 ^c	2430.42 ^b	2438.20 ^a	2370.33 ^c	2446.20 ^b	2444.84 ^b		2342.43 ^c	2444.84 ^b	2452.64 ^a	33.29
FCR	2.21 ^a	2.05 ^c	2.06 ^c	2.22 ^a	2.10 ^b	2.07 ^c		2.19 ^a	2.07 ^c	2.08 ^c	0.04
Survivability (%)	98.00	100.00	100.00	97.00	100.00	100.00		98.00	100.00	100.00	0.14

^{abc} Means on the same row having different superscripts are significantly different ($P < 0.05$); FCR, feed conversion ratio.

pathogenic organisms (Effiong and Ochagu, 2019).

The better feed intake observed in birds fed turmeric and their combination with reduction in birds fed PG singly affirmed the report of Effiong and Ochagu (2019), who noted reduction in feed intake of starter chicks fed 0.6% PG diet. The reduction in feed intake of birds fed PG could be attributed to PG's pungent alkaloid nature. Piperine is a pungent alkaloid with a modulating effect on other phytochemicals (Srinivasan, 2007). The improved feed intake noted in birds fed turmeric and their combination could be attributed to the curcumin present in both which brought about great synergy to express their abilities to digest-stimulate flavoring properties which improved the efficiency of

nutrient absorption that translated into increased final weight.

Report has shown that phytogenic feed additives enhance performance, feed conversion ratio, carcass meat safety and quality in animals (Stanacev et al., 2011; Dhama et al., 2014, 2015). The increased feed intake with higher weight gain at 4% of broiler chickens fed combination PG+ CL could be attributed to improvement in palatability and the quick digestive effect of these natural products. *Curcuma longa* has been found to increase the secretion of gastrointestinal enzymes, endogenous digestive enzyme secretion and activate immune response and antimicrobial and antioxidant activities (Dormans and Deans, 2000; Burt, 2004; Khan et al., 2012) while PG

contains capsaicinoids which could have increased the activities of digestive enzyme in gastric mucosa possibly enhance the synthesis of bile acids in the liver and their excretion in bile which beneficially enhance lipid digestion and absorption. The result here contradicts the report of Anyanwu et al. (2021) who reported a decrease in feed intake of finisher broiler chicken fed additives compared with the control diet. The best FCR noticed in birds fed different dietary additives regardless of inclusion level could be attributed to the growth promoting properties present. This implied that the nutrient requirements of the birds were adequately met. Several authors have reported improved FCR and body weight gain following phytogenic blends (Ciftci et al., 2005; Jamroz et al., 2005; Brenes and Roura,

Table 4. Main effects of *Curcuma longa* (CL), *Pipper guineense* (PG), their combination and levels of inclusion on hematological parameters of finisher broiler chickens.

Parameters	Level of inclusion of additives (%)			SEM				SEM
	CL	PG	CL + PG		0	2	4	
PCV (%)	39.87 ^a	38.13 ^b	40.57 ^a	1.56	36.55 ^c	38.72 ^b	39.56 ^a	2.82
Hb (g/dl)	9.88 ^b	9.92 ^b	10.62 ^a	0.62	11.66 ^b	11.96 ^b	12.74 ^a	1.06
RBC (x10 ¹² /l)	2.86 ^b	2.82 ^b	2.93 ^a	0.21	2.44 ^b	2.66 ^b	3.22 ^a	0.04
WBC (10 ⁹ /l)	5.22	5.18	4.93	0.17	5.32	5.16	5.31	0.82
LYM (%)	74.22	73.64	73.77	2.60	71.72	74.86	73.46	1.88
MCV (fl)	142.07	146.65	146.03	4.76	150.11	146.80	148.20	4.22
MCH (pg)	40.66	40.64	41.10	2.06	40.26	40.42	40.46	2.26
MCHC (g/dl)	28.81	27.28	29.96	2.16	25.20 ^b	27.42 ^{ab}	29.88 ^a	1.62

^{abc} Means on the same row having different superscripts are significantly different ($P < 0.05$); PCV, packed cell volume; Hb, hemoglobin; RBC, red blood cells; WBC, white blood cell; LYM, lymphocytes; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin; MCHC, mean corpuscular hemoglobin concentration.

2010). Similarly, this work affirmed the report of Giannenas et al. (2003) who noted a significant improvement in body weight and FCR of birds fed diet supplemented with oregano essential oil compared to the control diet. This corroborates the report of Mondal et al. (2015) and Samarasinghe et al., (2003) who reported improvement in feed efficiency of broiler chickens fed a Turmeric diet. Meanwhile, the report is at variance with Anyanwu et al. (2021) who reported no significance in FCR of birds fed different additives. The hematological parameters measured revealed that the PCV, Hb and RBC were higher for birds fed different dietary additives compared to control diets. It is an indication that both CL and PG singly and their combination were adequate for sufficient iron in the blood in maintaining the oxygen-carrying capacity of the birds to support life. The significant increase of these measured parameters noticed in 4% PG and their combination showed that at higher dosages health of the birds can still be improved and maintained without negative effects. Okpuzor et al. (2009) reported an increase in the RBC, Hb and PCV which is suggestive of polycythemia and positive erythropoiesis. The work agreed with the observation of Adegoke et al. (2018)

who reported a significant increase of PCV, Hb and RBC in finisher birds fed additives (*Curcuma longa* and Cayenne pepper) compared to birds on control diet. The white blood cell values were within the normal range for healthy birds. The report is at variance with Al-sultan (2003) who reported *Curcuma longa* (CL) inclusion in broiler diet to have increased the WBC of the birds. The MCHC of birds on 4% CL + PG were increased compared to control diet. This indicates sufficient iron in the blood. The values were within the reference range (26-35g/dl) across the group as reported by Bounous and Stedman (2000).

The significant increase observed in TP, Albumin and Globulin in birds fed different dietary additives especially at higher levels could be attributed to the curcuminoid present which has been absorbed and utilized of the essential vitamins and protein for stem cell formation of serum protein and albumin. The increased TP was suggestive of better enhancement of the phytogetic substance in protein synthesis in the liver. The work agreed with Uhegbu et al. (2015) who reported increased TP in birds fed additives. Similarly, Kumari et al. (2007) recorded significantly higher TP and globulin with low Albumin in broiler birds fed diet supplemented with 1kg turmeric. In

Table 5. Interactive effects of *Curcuma longa* (CL), *Pipper guineense* (PG), their combination and levels of inclusion on hematological parameters of finisher broiler chickens.

Parameters	CL			PG			CL + PG			SEM
	0	2	4	0	2	4	0	2	4	
PCV	36.70 ^c	37.60 ^b	38.64 ^a	36.62 ^c	37.86 ^b	38.82 ^a	36.82 ^c	37.64 ^b	38.92 ^a	2.13
Hb	9.62 ^c	10.38 ^b	11.12 ^a	9.60 ^c	10.82 ^b	10.42 ^b	9.80 ^c	10.65 ^b	11.08 ^a	1.22
RBC	2.54 ^c	2.72 ^b	2.98 ^a	2.48 ^c	2.65 ^b	12.68 ^b	12.62 ^b	2.70 ^b	2.88 ^a	0.11
WBC	5.12	5.34	5.26	5.01	25.30	5.38	5.18	5.26	5.44	0.23
LYM	63.88	71.24	64.64	64.01	64.46	4.22	64.44	64.64	63.01	1.23
MCV	146.50	148.20	146.80	144.26	148.32	147.88	146.65	143.07	144.03	11.23
MCH	40.44	40.46	40.54	41.02	40.88	40.64	40.64	40.66	40.22	3.72
MCHC	28.38 ^d	29.80 ^b	31.46 ^{ab}	27.08 ^d	29.96 ^b	30.03 ^b	27.28 ^d	29.07 ^b	32.61 ^a	3.12

^{abc} Means on the same row having different superscripts are significantly different ($P < 0.05$); PCV, packed cell volume; Hb, hemoglobin RBC, red blood cells; WBC, white blood cell; LYM, lymphocytes; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin MCHC, mean corpuscular hemoglobin concentration.

Table 6. Main effects of *Curcuma longa* (CL), *Pipper guineense* (PG), their combination and levels of inclusion on biochemical indices of finisher broiler chickens.

Parameters	Level of inclusion of Additives (%)							SEM
	CL	PG	CL+PG	SEM	0	2	4	
Total protein (g/dl)	6.86	6.74	6.96	0.36	5.12 ^b	5.84 ^{ab}	6.96 ^a	0.86
Albumin (g/l)	2.93	2.86	3.60	0.23	2.56 ^b	2.58 ^b	3.04 ^a	0.46
Globulin (g/L)	3.93	3.88	3.36	0.70	2.56 ^c	3.26 ^b	3.92 ^a	0.36
Glucose (mg/dl)	184.78	182.33	186.44	4.70	170.11 ^b	176.23 ^b	187.22 ^a	3.62
Alanine aminotransferase (u/L)	41.50	40.62	41.01	1.53	41.24	41.34	40.88	2.06
Alkaline phosphatase (u/L)	31.38	31.05	31.48	1.06	33.00	33.22	34.68	1.06
Aspartate aminotransferase (u/L)	97.84	96.94	98.02	5.84	99.60	90.44	91.30	3.12
Uric acid (mmol/l)	4.42	4.43	4.30	0.19	3.42	3.29	3.34	0.68
Creatinine (mmol/l)	20.51	20.25	20.64	1.13	19.20	20.42	20.44	1.16
Cholesterol (mmol/l)	104.44	106.24	106.88	5.49	108.86 ^a	92.60 ^b	94.22 ^b	8.25
Triglyceride (mmol/l)	99.88	99.26	96.86	4.83	120.43 ^a	94.13 ^b	92.24 ^b	2.84
High-density lipoprotein (mmol/L)	62.85	66.74	66.18	2.86	63.46 ^c	68.02 ^b	72.35 ^a	5.60
Low-density lipoprotein (mmol/L)	20.25	22.75	21.40	0.96	19.23 ^a	14.72 ^b	14.66 ^b	0.38
Very low-density lipoprotein (mmol/L)	19.74	19.86	19.88	0.86	21.72 ^a	16.57 ^b	17.86 ^b	2.06

^{abc} Means on the same row having different superscripts are significantly different ($P < 0.05$)

addition, Al-Noori et al. (2011) observed a significant increase in TP of broiler birds fed 0.5% and 1% turmeric. Meanwhile, Emadi et al. (2007) reported no

significant effect of turmeric at 0.25%, 0.5% and 0.75% on TP and Albumin of 21-day-old broiler chickens. This also attested to Qasem et al. (2016)

Table 7. Interactive effects of *Curcuma longa* (CL), *Pipper guineense* (PG), their combination and levels of inclusion on biochemical indices of finisher broiler chickens.

	CL				PG			CL+PG		
Parameters	0	2	4	0	2	4	0	2	4	SEM
Total protein (g/dl)	5.72 ^b	6.64 ^a	6.76 ^a	5.70 ^b	6.66 ^a	6.82 ^a	5.82 ^b	6.42 ^a	6.64 ^a	0.54
Albumin (g/l)	2.50	2.96	2.94	2.45	2.93	2.99	2.76	2.86	2.94	0.24
Globulin (g/L)	3.22	3.68	3.82	3.25	3.73	3.83	3.06	3.56	3.70	0.19
Glucose (mg/dl)	172.44	184.88	186.72	174.11	186.40	188.62	170.46	185.65	187.23	3.85
ALT (U/L)	40.88	41.46	42.24	40.50	41.88	41.86	40.75	41.68	41.86	2.87
ALP (U/L)	30.88	32.82	32.47	31.20	31.66	31.84	31.30	32.60	33.86	2.16
AST (U/L)	90.12	92.22	92.40	90.60	92.84	93.48	90.44	93.40	92.98	3.08
Uric acid (mg/dl)	4.06	4.36	4.29	4.42	4.43	4.30	4.42	4.32	4.23	0.78
Creatinine (mg/dl)	20.32	20.61	20.46	20.25	20.51	20.64	20.15	20.64	20.35	1.26
Cholesterol (mmol/l)	105.32	98.88	101.64	106.24	92.64	102.84	106.24	98.84	96.66	6.56
Triglyceride (mmol/l)	121.02	96.86	98.64	120.43	94.13	95.42	120.86	92.96	89.16	5.88
HDL (mmol/L)	63.87 ^c	67.96 ^b	70.28 ^a	63.46 ^c	67.86 ^b	72.65 ^a	62.89 ^c	68.24 ^b	75.40 ^a	5.60
LDL (mmol/L)	22.46 ^a	18.12 ^c	17.52 ^{cd}	22.75 ^a	19.25 ^b	17.40 ^{cd}	20.88 ^{ab}	17.88 ^{cd}	16.42 ^d	6.42
VLDL (mmol/L)	21.72	18.57	17.86	20.40	17.76	17.80	22.43	19.88	19.86	3.13

^{abc} Means on the same row having different superscripts are significantly different ($P < 0.05$); ALT, alanine aminotransferase; ALP, alkaline phosphatase; AST, aspartate aminotransferase; HDL, high density lipoprotein; LDL, low density lipoprotein; VLDL, very low density lipoprotein.

who reported higher serum total protein in birds fed additives compared with control diet. The author also reported the non-significance of creatinine in turmeric diet, this is in line with the present study.

The slight increase in the glucose of birds fed different dietary additives regardless of inclusion level compared to the control diet could be attributed more to *Curcuma longa* which may have increased the activity of glucose transporters and they are family of transmembrane proteins that help to carry glucose across the plasmalemma (Kumari et al., 2007). On contrary to the report of Qasem et al. (2016) and Gowda et al. (2009) who noted a decline in the glucose of broiler birds fed 444mg/kg of turmeric diet. Similarly, Arshami et al. (2013) reported no significant effect of turmeric at 5, 15 and 25g/kg fed laying hens on TP, Albumin and glucose. Meanwhile, this research recorded significant influence of additives on glucose.

The ALT, ALP and AST reports showed no significant effect of the different dietary additives. Consequently, Kumari et al. (2007) and Mehala and Moorthy (2008) noted that the activity of these liver enzymes such as ALT, AST, ALP AND LDH were not

affected by the inclusion of *Curcuma longa* additives. Likewise, Qasem et al. (2016) and Gowda et al. (2009) reported no significant effect of the dietary inclusion of turmeric supplement compared with birds on the control diet on the liver enzymes (AST, ALT and GGT) these reports agreed with the present research. Meanwhile, Emadi and Kermanshahi (2007); Hosseini-Vashan et al. (2012) opinioned decreased ALT, ALP and AST for broiler chickens fed 0.25%, 0.5% and 0.75%; (0.4% and 0.8%) CL supplement respectively.

The inclusion of additives significantly lowers cholesterol, triglyceride, LDL and VLDL of the birds compared to birds on the control diet. Meanwhile, the inclusion of different dietary additives most especially the combination at 4% increased the HDL of the birds. Triglycerides are synthesized in the liver from fatty acids protein and glucose when they are above The body's current need and are stored in adipose tissue (Esubonteng, 2011). The significant reduction observed with increasing dietary additives may be attributed to the regulation of lipid metabolism in a favorable sense. The higher HDL of birds fed 4% dietary additives regardless of the type as compared

to other groups could be attributed to enhanced hypocholesterolemic mechanism and the hypolipidemic action of phytogetic feed additives. The reduction of LDL and VLDL noticed in 2% and 4% of different additives either singly or their combination could be attributed to the process of antioxidant and anti-peroxide lowering action on LDL or the decrease in hepatic production of very low-density lipoprotein (VLDL) which serve as the precursor of LDL in the blood circulation system (Kim et al., 2009). This work agreed with earlier works of Al-Kassie et al. (2011) and Uhegbu et al. (2015) who reported reduced cholesterol in broiler birds fed black pepper at 0.5% to 1% and 25-75mg/kg to albino rats.

CONCLUSION

The inclusion of different dietary additives such as *Piper guineense* (PG) and *Curcuma longa* (CL) either singly or their combination could be used as phytogetic substances in a broiler's diet for improved growth performance and healthy living. In addition, higher inclusion levels of up to 4% pose no threat to growth performance and blood parameters.

Conflict of interest statement

There is absolutely no conflict of interest with any individual or organization regarding the materials discussed in the manuscript.

Source of fund: Self sponsor

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