



## **Response of Broiler Chicken to Microbial Phytase: Effects on Phytin-Phosphorus, Serum Biochemistry and Carcass Characteristics**

**O. T. Daramola<sup>1\*</sup>**

<sup>1</sup>*Department of Agricultural Technology, Federal Polytechnic, P.M.B 5351, Ado-Ekiti, Ekiti-State, Nigeria.*

### **Author's contribution**

*The sole author designed, analysed, interpreted and prepared the manuscript.*

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### **ABSTRACT**

**Aim:** The experiment was conducted to determine phytin-phosphorus in feed, pre-caeca digesta and faecal droppings of broiler chickens and to investigate serum metabolites and carcass characteristics of broiler chicken as affected by phytase supplementation.

**Study Design:** The experiment employed a completely randomized design. All data generated were subjected to analysis of variance,  $P=0.05$ .

**Place and Duration of Study:** The study was carried out at the Teaching and Research farm of the Ekiti State University, Ado-Ekiti, Nigeria between February and April, 2011.

**Methodology:** Two hundred and forty unsexed day-old Anak 2000 strain broiler chicken were used in a 56-day feeding trial. The birds were allotted to five treatments with 4 replicates per treatment of 12 birds per replicate. Diet 1 was the reference diet with no phytase supplementation but with both plant and animal protein sources. Diets 2 and 3 were duplicate diets with enzyme supplementation only in diet 3. Diets 4 and 5 were also duplicate diets with enzyme supplementation only in diet 5. Diets 2 and 3 had groundnut cake as the major plant protein ingredient while diets 4 and 5 had soybean cake as the major protein ingredient. They were fed

\*Corresponding author: E-mail: [olajumoke.daramola2016@gmail.com](mailto:olajumoke.daramola2016@gmail.com);

ad-libitum. Phytin-phosphorus in feed, pre-caeca digesta and faecal droppings of broiler chickens were determined and the effect of phytase supplementation on serum metabolites and carcass analysis of broiler chicken were also investigated.

**Result:** The levels of phytin-phosphorus in feed exceeded that detected in pre-caeca digesta of ileo-caeco-colonic junction likewise the levels of phytin-phosphorus in pre-caeca digesta exceeded that detection in faecal droppings. All diets supplemented with or without phytase had no significant influence on all the serum metabolites of broiler finisher investigated in this experiment ( $P>0.05$ ) but a slight increase in the values of urea, creatinine, cholesterol, triglyceride and high density lipoprotein in the serum of birds with phytase supplementation. There were no significant ( $P>0.05$ ) differences recorded for the liveweight, dressed weight, eviscerated weight and all carcass cuts percentage of broiler finisher.

**Conclusion:** The supplementation of broiler diets with or without phytase constituted no danger to the health status of broiler birds. In carcass analysis phytase supplementation promoted muscle accretion.

*Keywords: Phytin-phosphorus; carcass; serum; phytase; broiler chicken.*

## 1. INTRODUCTION

Recently, there has been an effort to improve the nutritive worth of feedstuffs using phytase. Large proportion of phosphorus in plant materials as phytate form (salts of phytic acid) and about 70% is not available to be used by monogastric animals [1]. The vulnerable portability of poultry to exploit phosphorus phytate in grains [2] is attributed to their decreased secretion of phytase in the intestines of birds. Large amount of phosphorus in birds droppings cause environmental pollution in areas with intensive poultry production [3]. Phytase has the ability to breakdown undigested phytic acid in grains and oilseeds releasing digestible calcium and phosphorus in monogastric animals [4].

Microbial phytase positively influence nutrients, it may increase the availability of phosphorus in corn-soybean diets by 15-45% and decreases it in the waste up to one-third without affecting productive performance [5]. One of mechanism that has been suggested by [6] is that dietary phytate, the substrate for exogenous phytase is an anti-nutrient beyond its influence on phosphorus solubility having the capacity of secretion and absorptions dynamics in the gastro intestinal tract. The mechanism by which phytate and phytase alter secretory and absorptive physiology are not fully understood but are thought to be related to the reactive nature of phytate and electrostatic aggregation of dietary protein in the gastric phase of digestion. The study aimed to determine phytin-phosphorus in feed, pre-caeca digesta and faecal droppings of broiler chickens and to investigate the effect of phytase on serum metabolites and carcass analysis of broiler chickens as affected by phytase supplementation.

## 2. MATERIALS AND METHODS

**Experimental Location** The study was carried out at poultry unit of the Teaching and Research farm, Ekiti State University, Ado-Ekiti, Nigeria.

### 2.1 Experimental Animals and Management

Two hundred and forty day old broiler chicks of Anak 2000 breed were used in the experiment. There were five experimental diets with four replicates per treatment. Twelve birds were allotted per replicate, amounting to 48 birds per treatment in a completely randomized design (CRD) experiment. The birds were offered feed ad-libitum and water on daily basis throughout the experimental period which lasted for 8 weeks.

### 2.2 Experimental Diets

Five experimental diets were formulated to be iso-nitrogenous and iso-caloric and the ingredients were predominantly plant products. Diet 1 was the reference diet with no phytase supplementation with protein sources of both plant and animal origins. Diets 2 and 3 were duplicate diets with enzyme supplementation only in diet 3. Diets 4 and 5 were also duplicate diets with enzyme supplementation only in diet 5. Diets 2 and 3 had groundnut cake as the major plant protein ingredients while diets 4 and 5 had soy bean as the major plant protein ingredients. All diets were supplemented with feed grade DL-methionine and lysine. The inclusion rate of phytase was 0.1/kg for this study.

**Table 1. Composition of experimental diets (g/100 g) for broiler chicks**

Ingredients	1	Diets				5
		2	3	4		
		Phytase inclusion				
	Reference diet	-	+	-	+	
Maize	45.00	45.00	45.00	45.00	45.00	
Groundnut cake	15.00	40.00	40.00	0.00	0.00	
Soyabean	25.00	0.00	0.00	40.00	40.00	
Wheat offal	8.70	10.70	10.70	10.70	10.70	
Fishmeal (72% CP)	2.00	0.00	0.00	0.00	0.00	
Bone meal	2.50	2.50	2.50	2.50	2.50	
Oyster shell	0.50	0.50	0.50	0.50	0.50	
NaCl	0.50	0.50	0.50	0.50	0.50	
DL-Methionine	0.15	0.15	0.15	0.15	0.15	
L-Lysine	0.15	0.15	0.15	0.15	0.15	
Premix	0.50	0.50	0.50	0.50	0.50	
Total	100.00	100.00	100.00	100.00	100.00	
<b>Calculated composition (%)</b>						
Crude protein	24.00	23.72	23.72	23.32	23.32	
Crude fibre	4.32	3.81	3.81	4.89	4.89	
Metabolisable energy (Kcal/kg)	2777.21	2719.69	2719.69	2759.69	2759.69	
<b>Analysed composition (%)</b>						
Crude protein	24.23	23.51	23.51	23.53	23.53	
Crude fibre	5.30	5.51	5.52	5.67	5.64	

*Negative sign (-) means no phytase inclusion; positive sign (+) means phytase inclusion at 250 FTU/g minimum activity Inclusion rate of phytase in diet*

**Table 2. Composition of experimental diets (g/100 g) for broiler finisher birds**

Ingredients	1	Diets				5
		2	3	4		
		Phytase inclusion				
	Reference diet	-	+	-	+	
Maize	62.00	62.00	62.00	62.00	62.00	
Groundnut cake	10.00	25.00	25.00	0.00	0.00	
Soyabean	15.00	0.00	0.00	25.00	25.00	
Wheat offal	6.70	8.70	8.70	8.70	8.70	
Fishmeal (72% CP)	2.00	0.00	0.00	0.00	0.00	
Bone meal	2.50	2.50	2.50	2.50	2.50	
Oyster shell	0.50	0.50	0.50	0.50	0.50	
NaCl	0.50	0.50	0.50	0.50	0.50	
Methionine	0.15	0.15	0.15	0.15	0.15	
Lysine	0.15	0.15	0.15	0.15	0.15	
Premix	0.50	0.50	0.50	0.50	0.50	
Total	100.00	100.00	100.00	100.00	100.00	
<b>Calculated composition</b>						
Crude protein (%)	19.88	18.93	18.93	18.68	18.68	
Crude fibre (%)	3.73	3.49	3.49	4.17	4.17	
Metabolisable energy (Kcal/kg)	2935.87	2888.35	2888.35	2913.35	2913.35	
<b>Analysed composition</b>						
Crude protein (%)	21.16	20.48	20.47	20.76	20.74	
Crude fibre (%)	5.24	5.43	5.43	5.37	5.36	

*Negative sign (-) means no phytase inclusion; positive sign (+) means phytase inclusion at 250 FTU/g minimum activity inclusion rate of phytase in diets*

### 3. DATA COLLECTION

#### 3.1 Pre-caeca Digesta Study

The experimental birds were cut opened from jugular vein. The terminal two-third of section between Meckel diverticulum and 2cm anterior to the ileo-caeco-colonic junction was severed. The contents from each bird from replicates of all diets were flushed out with distilled water into a sterile bottles. The Pre-caeca digesta was taken to laboratory for determination of phytin-phosphorus. The five formulated diets, pre-caeca digesta and droppings from experimental birds were taken to the laboratory for the determination of phytin-phosphorus. The extraction and precipitation of phytin in the experimental diets, pre-caeca digesta and droppings from experimental birds were done by Wheeler and Ferrel's method [7] while iron in the precipitate was determined by Makowers's method [8]. Phytin was determined by using a 4:6 Fe/P ratio to calculate phytin-phosphorus and multiplying the phytin-phosphorus by 3.55 as suggested by Young and Greaves [9].

#### 3.2 Blood Sample Collection

Blood samples for serum metabolites were collected in vacutainer tubes without anticoagulants and sent to laboratory. The tubes were kept in a slanting wooden rack and the blood samples were allowed to clot overnight. The serum was separated clearly by decanting after the blood samples were spun in a centrifuge at 300 rpm, 4°C and 10 minutes. The serum sample was kept in sterile vacutainer tubes and kept deep frozen prior to analysis to determine cholesterol as outlined by [10]. The triglyceride and high density lipoprotein (HDL) were measured using commercial kits while low density lipoprotein (LDL) was measured by indirect method using the [11] formula.  $LDL\text{-cholesterol (mg/dL)} = \text{Total cholesterol} - \text{Triglyceride}/5 - \text{HDL-cholesterol}$ .

Following slaughtering, the carcass were scalded at 75°C in a water bath for about 30seconds after defeathering. The dressed chickens were eviscerated and the measurements of the carcass traits were taken. The following carcass measurement were taken liveweight, dressed weight, eviscerated weight, thigh, drumstick, back, breastweight, shank, wing, head and neck. All carcass except the dressed and eviscerated weight were expressed as percentage of liveweight.

All data collected in the studies were subjected to analysis of variance using the SPSS software package Duncan's Multiple Range Test of one way ANOVA (SPSS 17.0 for windows) was used to analyse the mean differences of the same parameter. Significant differences were considered where necessary at a level of ( $p < 0.05$ ).

### 4. RESULTS AND DISCUSSION

The phytin-phosphorus in feed, pre-caeca digesta and faecal droppings of broiler chickens fed experimental diets is presented in Table 3. The phytin-phosphorus level in experimental diets ranged from  $0.58 \pm 0.01$  mg/g in diet 3 to  $0.68 \pm 0.01$  mgg<sup>-1</sup> in reference diet. The phytin-phosphorus concentration in the pre-caeca digesta of the ileo-caeco-colonic junction ranged from  $0.24 \pm 0.01$  mgg<sup>-1</sup> in diet 5 to  $0.34 \pm 0.01$  mg/g in reference diet. Phytin-phosphorus concentration in faecal droppings was highest in diet 2 followed by reference diet, diet 4, diet 5 and diet 3 at  $0.19 \pm 0.02$  mgg<sup>-1</sup>,  $0.14 \pm 0.01$  mgg<sup>-1</sup>,  $0.11 \pm 0.02$  mgg<sup>-1</sup>,  $0.08 \pm 0.02$  mgg<sup>-1</sup> and  $0.07 \pm 0.01$  mgg<sup>-1</sup>, respectively. Expectedly, the levels of phytin-phosphorus in feed exceeded that detected in the pre-caeca digesta likewise the levels of phytin-phosphorus in pre-caeca digesta exceeded that detected in faecal droppings. Noteworthy that less phytin-phosphorus was noticeable in the pre-caeca digesta and faecal droppings from experimental birds on phytase supplemented diets. This implied that breakdown of phytin-complex and absorption of nutrients had taken place in the digestive system. This could therefore explain the decline in the concentration of phytin-phosphorus in the pre-caeca digesta and faecal droppings compared with the initial concentration in the experimental diets. Absorption of the fluid portion of the digesta has been reported to take place at the lower portion of the digestive system hence concentrating the excreta voided. This findings were consistent with the reports of several workers that reported an improvement in phytate phosphorus utilization [12,13] showed that microbial phytase supplementation of broiler diets increased the availability of nutrients and decreased the amount of phosphorus in the droppings.

The data on serum metabolites are given in Table 4. All diets supplemented with or without phytase had no significant influence on all the serum metabolites of broiler finisher investigated in this experiment ( $P < 0.05$ ). However, there was

**Table 3. Phytin-phosphorus in feed, pre-caeca digesta and faecal droppings of broiler finisher birds fed experimental diets**

Parameters	1	Diets				5
		2 3 4				
		Phytase inclusion				
Reference diet	-	+	-	+		
Feed (mg/g)	0.68±0.01	0.59±0.01	0.58±0.01	0.62±0.01	0.62±0.01	
Pre-caeca digesta (mg/g)	0.34±0.01	0.32±0.01	0.25±0.01	0.30±0.01	0.24±0.01	
Faecal droppings (mg/g)	0.14±0.01	0.19±0.02	0.07±0.01	0.11±0.01	0.08±0.01	

*Negative sign (-) means no phytase inclusion; positive sign (+) means phytase inclusion at 250 FTU/g minimum activity inclusion rate of phytase in diets*

**Table 4. Serum metabolites of broiler finisher birds fed experimental diets**

Parameters	1	Diets				5
		2 3 4				
		Phytase inclusion				
Reference diet	-	+	-	+		
Urea (mmol/L)	0.95±0.07	0.95±0.0466	0.98±0.03	0.96±0.02	0.97±0.09	
Creatinine (µmol/l)	65.35±0.76	5.30±0.54	65.49±0.82	65.24±0.52	65.62±0.78	
Cholesterol(µmol/l)	2.90±0.34	2.87±0.15	2.91±0.52	2.90±0.56	2.94±0.42	
Triglyceride	0.26±0.07	0.26±0.17	0.28±0.05	0.27±0.74	0.29±0.06	
HDL	1.96±0.53	1.98±0.49	2.08±0.53	2.06±0.74	2.10±0.47	
LDL	0.42±0.02	0.43±0.03	0.45±0.04	0.42±0.14	0.46±0.05	

*HDL: High density lipoprotein; LDL : Low density lipoprotein. Means with different superscript on the same row differ significantly (p<0.05). Negative sign (-) means no phytase inclusion; positive sign (+) means phytase inclusion at 250 FTU/g minimum activity inclusion rate of phytase in diets*

**Table 5. Carcass characteristics of broiler finisher birds fed experimental diets**

Parameters	1	Diets				5
		2 3 4				
		Phytase inclusion				
Reference diet	-	+	-	+		
Liveweight (g)	1769.50±62.42	1752.00±64.50	1755.00±42.03	1759.50±62.91	1765.00±62.42	
DW(%)	91.05±0.17	88.57±0.07	89.46±0.03	88.47±0.07	89.49±0.03	
EW (%)	80.23±0.06	79.34±0.26	80.00±0.02	77.97±0.34	79.95±0.26	
Thigh (%)	8.73±0.03	7.68±0.01	8.57±0.05	7.66±0.03	8.56±0.06	
Drumstick (%)	9.48±0.02	9.03±0.02	9.41±0.03	9.02±0.01	9.47±0.03	
Back (%)	17.14±0.05	16.83±0.02	17.14±0.04	16.60±0.17	17.12±0.02	
Breastweight (%)	17.65±0.07	16.32±0.02	17.51±0.03	16.37±0.04	17.62±0.01	
Shank (%)	2.57±0.02	2.41±0.01	2.52±0.01	2.39±0.01	2.53±0.02	
Wing (%)	4.11±0.08	3.94±0.06	4.10±0.07	3.68±0.34	4.10±0.07	
Head (%)	3.71±0.02	3.41±0.01	3.62±0.02	3.43±0.03	3.64±0.01	
Neck (%)	4.39±0.01	4.15±0.01	4.38±0.02	4.14±0.03	4.39±0.02	

*DW: Dressed weight; EW: Eviscerated weight. Means with different superscript on the same row differ significantly (p<0.05). Negative sign (-) means no phytase inclusion; positive sign (+) means phytase inclusion at 250 FTU/g minimum activity inclusion rate of phytase in diets*

a slight increase in the values of urea, creatinine, cholesterol, triglyceride and high density lipoprotein in the serum of birds with phytase supplementation. The results also corroborated the earlier work of [14] and [15] they reported that phytase supplementation had no

adverse effect on serum metabolites of broiler birds.

The result on carcass characteristics of broiler birds fed diets with or without phytase are given in Table 5. The liveweight, dressed weight and

eviscerated weight of birds were not significantly ( $p>0.05$ ) influenced by the experimental diets. This implies that supplementing the diets of broiler birds with or without phytase had no effect on liveweight, dressed weight and eviscerated weight of the broiler birds. All percentage of carcass cuts (thigh, drumstick, back, breast weight, shank, wing, head and neck for all the treatments were not significantly ( $p<0.05$ ) influenced by the experimental diets. The result agreed with the report of [16] that phytase supplementation had no effect on carcass cuts percentage. However, trend of increase in weight of carcass cuts seems higher in diets 3 and 5 (phytase supplemented diets) compare to diets 2 and 3 (diets without phytase supplementation) suggesting that the phytase supplementation promoted better muscle accretion in the birds.

## 5. CONCLUSION

It can be concluded that supplementation of broiler diets with or without phytase constituted no danger to the health status of broiler birds. In carcass analysis, inclusion of phytase in the diets promoted significant muscle accretion.

## CONSENT

It is not applicable.

## ETHICAL APPROVAL

Animal Ethic committee approval has been taken to carry out this study.

## COMPETING INTERESTS

Author has declared that no competing interests exist.

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