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Effect of phytase and protease supplementations on growth performance and carcass characteristics of broiler chickens fed sub-optimal levels of crude protein

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***Target Audience**:* Poultry Farmers, Feed Millers, Animal Scientists, Feed Additives Suppliers

Abstract

Two hundred and seventy (270) day old unsexed Arbor Acre broiler chicks were assigned to nine dietary treatments comprising three replicates of 10 chicks each. During the starter phase birds were fed the control 23% crude protein (CP) without enzyme (T1), 21% CP without enzyme (T2), 21% CP plus phytase (PHY) (T3), 21% CP plus protease (PRO) (T4), 21% CP plus PHY and PRO (T5); and 19% CP without enzyme (T6), 19% CP plus PHY (T7), 19% CP plus PRO (T8), 19% CP plus PHY and PRO (T9). Phytase and protease supplementation followed the same arrangement in all the phases, but the crude protein were 23, 20 and 18% CP (in T1); 21, 18 and 16% CP (in T2, T3, T4 and T5) and 19, 16 and 14% CP (in T6, T7, T8 and T9) during starting, growing and finishing phases respectively. Feed and water were given unrestrictedly. The feed intake and weights of birds were recorded on weekly basis. Nutrient digestibility was determined at each phase and carcass evaluation was done at the end of the experiment. From 0-56 days, PHY supplementation significantly ($P < 0.05$) improved final body weight and daily weight gain in suboptimal CP diets (T3). Enzymes had no significant effect on the carcass characteristics except drumstick, which was highest ($P < 0.05$) on PRO treatments. The addition of phytase or protease alone and in combination increased the available nutrient levels and there was a significant effect of enzymes supplementation on nutrient digestibility. It could be concluded that phytase, protease and their combination improved performance, nutrient digestibility and carcass cut part of broiler chickens fed suboptimal crude protein diets.

Keywords: Feed additives; enzymes; suboptimal crude protein; carcass

Description of Problem

Poultry diet is made up of largely cereal grains, cereal by-products and oilseeds meal; and a high proportion (about 60 to 70%) of phosphorus (P) in these feed ingredients is bound as the salts of phytic acid, in the form of phytates (1). One of the most important anti-nutritional factors for non-ruminant animals is phytate, because of its abundance in many feed ingredients of plant origin. However, monogastric animals lack capacity to efficiently utilize phytate-P and this poses a challenge. This challenge is ascribed to the ability of phytate to form complexes with other dietary nutrients, such as minerals, proteins, free amino acid, and starch (2). The applications of phytase feed enzymes in pig and poultry diets generate bioavailable P and consequently reduce the P load on the environment (3). Also, in recognition of the 'extra-phosphoric effects' of phytase, some nutritionists elect to place matrix values on phytase feed enzymes for protein/amino acids and energy, in addition to calcium (Ca) and P [(4),(5) and (6)]. There is the likelihood that phytate negatively influences protein and energy utilization in poultry and these effects could be ameliorated by phytase. Nevertheless, there is still no consensus as to the extent that phytase enhances protein and energy utilisation. Several studies show that responses in amino acid digestibilities following phytase supplementation are variable and the underlying mechanisms have not been completely understood (5). Phytase supplementation has been reported to increase the apparent ileal digestibility of crude protein and amino acid by a three-way interaction between phytate, proteolytic enzymes and protein-AA in the digestive contents (8), (9) and (10). However, it was pointed out that phytase supplementation does not consistently improve the digestibility of crude protein and amino acids (11). Reducing dietary crude protein has been a strategy to decrease nitrogen excretion and ammonia production, but growth performance can be adversely affected if adequate amounts of dietary amino acids are not provided (12), (13) and (14). Phytase supplementation has been reported to improve amino acid utilization (15, 16) and nitrogen excretion may also be reduced by supplementing diets with phytase. Another very important enzyme used in monogastric nutrition is protease. Exogenous protease enzymes have been used as a feed additive in recent years to increase the digestibility of protein in the diets of monogastric animals. Addition of protease to diets with sub-optimal crude protein levels improved daily weight gain and decreased daily feed intake and feed conversion ratio (17) and (18). Also, a reduction in dietary crude protein by 24% and 28% for starter and finisher broiler chickens respectively had no deleterious effect on the performance when protease was added to the broiler diet (18). The objective of this study therefore, was to evaluate the effect of phytase and protease, and their combination on the growth performance and carcass characteristics of broiler chickens fed sub-optimal levels of dietary crude protein.

Materials and Methods

Experimental site

The experiment was carried out at the Teaching and Research Farm of Obafemi Awolowo University, Ile-Ife, Osun State.

Experimental diets

Starter's diet

Nine experimental diets were fed during the starter phase which were: the control (23% CP) without enzyme (T1), 21% CP without enzyme (T2), 21% CP plus PHY (T3), 21% CP plus PRO (T4), 21% CP plus PHY and PRO (T5); and 19% CP without enzyme (T6), 19% CP plus PHY (T7), 19% CP plus PRO (T8), 19% CP plus PHY and PRO (T9). Crude protein levels in suboptimal diets compared to the control (i.e. feeding standard for broilers) were reduced by 2% (23 in T1 vs. 21% in T2 to T5) and 4% (23 in T1 vs. 19% in T6 to T9).

Grower's diet

During the grower phase, the birds were fed 20% CP control diet without enzyme (T1), 18% CP without enzyme (T2), 18% CP plus PHY (T3), 18% CP plus PRO (T4), 18% CP plus PHY and PRO (T5); and 16% CP without enzyme (T6), 16% CP plus PHY (T7), 16% CP plus PRO (T8), 16% CP plus PHY and PRO (T9). Crude protein levels in suboptimal diets compared to the control (i.e. feeding standard for broilers) were reduced by 2% (20 in T1 vs. 18% in T2 to T5) and 4% (20 in T1vs 16% in T6 to T9).

Finisher's diet

During the finisher phase, birds were fed 18% CP control diet without enzyme (T1), 16% CP without enzyme (T2), 16% CP plus PHY (T3), 16% CP plus PRO (T4), 16% CP plus PHY and PRO (T5); and 14% CP without enzyme (T6), 14% CP plus PHY (T7), 14% CP plus PRO (T8), 14% CP plus PHY and PRO (T9). Crude protein levels in suboptimal diets compared to the control (i.e. feeding standard for broilers) were reduced by 2% (18 in T1 vs. 16% in T2 to T5) and 4% (18 vs. 14%; T6 to T9).

The phytase and protease enzymes were supplemented at 0.1 g/kg and 0.05 g/kg diet, respectively. Diets in each phase were formulated to be isocaloric as shown in Table 1. The duration for starter, grower and finisher phases were 0-21 days, 21-42 days and 42-56 days respectively. The control diets in each phase were formulated using National Research Council recommendation (19) as a guide.

Experimental Birds and Management

Two hundred and seventy (270) unsexed day-old, arbor acre broiler chicks with initial body weight of 47.60 ± 0.25 g were randomly allocated to the dietary treatments formulated in three phases, with ten birds per replicate in a completely randomized design. Birds were fed experimental diets in the three phases; starter, grower and finisher. Feed and water were supplied unrestrictedly for the period of the experiment. Routine management procedures were observed.

Data Collection

Birds were weighed on weekly basis and weekly feed offered and left over were weighed. At the end of the experiment, feed intake, weight gain and feed conversion were evaluated. In the digestibility trial, a total of 27 birds per phase and one bird per replicate were selected to determine apparent nutrient digestibility of diets at each phase using total collection method. At the starter, grower and finisher phases, birds were moved to metabolic cage on day 13, 35 and 49 respectively and acclimatized for a period of five days at each phase. The birds were put in cages with trays underneath to collect excreta for three days. At 56 days of age, one bird per replicate (bird with representative weight of each treatment) making a total of three birds per treatment was slaughtered after fasting for 12 hours for carcass evaluation.

Chemical and Statistical Analyses

Analyses of feed and excreta were performed at the Poultry Meat Research Laboratory of Department of Animal Sciences, Obafemi Awolowo University, Ile-Ife, Osun State, Nigeria. Diets and excreta were analyzed for proximate compositions according to standard procedures (20). Data generated were subjected to one-way analysis of variance (ANOVA) using the General Linear Models procedure of SAS (21). Significant differences among treatments were separated at 5% significance level by Duncan's Multiple Range Test.

Table 1: Gross composition of experimental diets fed to broiler chicken in the three phases[1]

	Starter (0 - 3 weeks)			Grower (3 - 6 weeks)			Finisher (6 - 8 weeks)		
Ingredients	23% CP **T1**	21% CP **T2**	19% CP **T6**	21% CP **T1**	19% CP **T2**	16% CP **T6**	18% CP **T1**	16% CP **T2**	14% CP **T6**
Maize	52.26	56.00	59.00	57.39	59.83	62.10	59.83	62.66	64.36
SBM[2]	37.30	30.04	24.58	27.92	23.33	17.00	24.45	19.08	12.08
Wheat Offal	4.75	7.00	8.38	8.90	8.40	10.07	8.40	9.30	11.41
HQCP[3]	1.10	2.10	3.4	2.20	4.73	6.86	4.85	6.38	9.33
Bone meal	1.10	1.40	1.4	0.80	0.80	0.96	0.75	0.77	0.80
Limestone	1.10	0.60	0.60	0.92	0.92	0.92	0.92	0.92	0.92
Fish meal	1.50	1.50	1.50	1.00	1.00	1.00	-	-	-
DL-meth.	0.19	0.19	0.19	0.10	0.12	0.14	0.07	0.09	0.12
L-lysine	0.20	0.31	0.45	0.27	0.37	0.45	0.21	0.33	0.48
Premix	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Salt	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Total	**100**	**100**	**100**	**100**	**100**	**100**	**100**	**100**	**100**
Calculated analysis (%)									
ME (kcal/kg)	2961.98	2968.69	2971.99	2966.23	2977.41	2963.32	2974.54	2979.70	2981.68
Crude protein (%)	23.01	21.04	19.27	20.01	18.43	16.44	18.07	16.43	14.28
CF (%)	3.98	3.88	3.79	3.91	3.83	3.78	3.90	3.81	3.45

[1]T1 (non-enzyme supplemented diet of 23, 21 and 18% CP at starting, growing and finishing phases respectively); T2 (non-enzyme supplemented diet of 21, 18 and 16% CP at starting, growing and finishing phases respectively); T3 (T2 plus PHY); T4 (T2 plus PRO); T5 (T2 plus PHY and PRO) and T6 (non-enzyme supplemented diet of 19, 16 and 14% CP at starting, growing and finishing phases respectively); T7 (T6 plus PHY); T8 (T6 plus PRO); T9 (T6 plus PHY and PRO). Enzymes were added to the diets at the expense of maize; making a total of nine diets at each phase.
[2]SBM – Soya bean meal
[3]HQCP – High quality cassava peel (fine mash)

Results and Discussion

The performance characteristics of broiler chicken fed diets with varying levels of crude protein are presented on Table 2. There were significant effects of phytase and protease supplementation on final body weight (FBW), daily weight gain (DWG) and daily feed intake (DF) during the starter phase. Birds fed enzyme-supplemented diets at 21% CP (T3, T4 and T5) had improved FBW and DWG compared to the control (T1). Birds fed diet with combination of phytase and protease at the starter phase (T5) had the highest ($P<0.05$) FBW and DWG compared to the control (T1), but this was similar to the phytase supplemented (T3), protease supplemented (T4) and non-enzyme supplemented 19% CP (T6). Addition of phytase or protease individually, or their combination to diet with suboptimal crude protein (21% CP) significantly improved the final body weight by 4.71%, 3.83% and 10.06% respectively when compared to the non-enzyme supplemented diet (T2). The daily feed intakes of broilers fed diets with reduced crude protein of 21 and 19% with or without enzyme supplementation were significantly higher than the control diet (T1) with 23% crude protein. The daily feed intake ranged between 45.41 - 51.75 g during the starter phase. The feed conversion ratios (FCR) at the starter phase ranged from 1.84 to 2.01, but were not significantly different among the treatments, though T5 had the best FCR and the highest ($P<0.05$) DWG. There was also a significant effect of crude protein on daily feed intake, such that the daily feed intake of the highest crude protein control diet was inferior to all the lower crude protein treatment diets.

At the grower phase (Table 2), addition of phytase, protease and their combination increased daily weight gain by 13%, 9.9% and

2.5% respectively in the 18% CP compared to non-enzyme supplemented control diet. Phytase supplementation to 18% crude protein diet (T3) increased the daily feed intake of the birds by 5.1% compared with birds fed non-enzyme supplemented diet (T2). However, the feed conversion ratios were similar across the treatments. The supplementation of phytase, protease or their combination to 16% CP diets (T7 to T9) had no significant effect on all response variables compared to the non-enzyme supplemented 16% CP diet (T6), but DWG of these treatments tend to be lower than the higher CP diets of 18 and 20%.

At finishing phase (Table 2), improved final body weight was observed among the enzyme-supplemented group (T3, T4 and T5) at 16% crude protein. Birds fed phytase-supplemented diet (T3) had the highest final body weight and daily weight gain. The body weight gain was improved by 16.79% when phytase was supplemented. Protease also improved the daily weight gain by 13.65% when compared to non-enzyme supplemented diet (T2). The overall performance of broiler chickens fed suboptimal crude protein diets with or without enzyme supplementation is also shown in Table 2. There was a significant ($P<0.05$) effect of enzyme supplementation on final body weight and daily weight gain. Birds fed diets with phytase, protease and their combination (T3, T4 and T5) had better FBW and DWG and the values ranged from 2253.64 to 2429.73g and 39.39 to 42.53g respectively, and T3 had the highest FBW and DWG. Phytase and protease supplementation improved the daily weight gain by 12.84% and 9.8% respectively when compared with the non-enzyme supplemented diet T2. There was no significant effect of enzyme supplementation on T7, T8 and T9.

Lack of significant effect of enzyme supplementation observed on T7, T8 and T9 across the three phases may be because of reduction of crude protein (23 to 19% CP, 20 to 16% CP and 18 to 14% CP at starter, grower and finisher phases respectively) in the diets which may be too low for the birds to express their full genetic potential in terms of growth and daily weight gain. The improvement observed in the performance of the birds fed phytase-supplemented diets could be due to the hydrolytic effect of phytase enzyme on the feed ingredients, thereby enhancing the release of nutrients. Positive effect of protease on performance of the birds could be attributed to its complementary effect with the limited levels of endogenous enzymes. The improvement in FBW and BWG observed in birds fed protease supplemented diets was in agreement with the earlier findings (18) where improvement in FBW and BWG when broilers were fed reduced crude protein diets supplemented with protease was reported**.** The increased feed intake observed in this study was in harmony with earlier report (22) in which protease supplementation resulted in increased BWG and feed intake. The significant effect of phytase, protease and their combination observed in this study on performance supported the report of Cowieson and Adeola (10) who stated that phytase, protease or their combination improved the performance of broiler chickens when fed diets nutritionally marginal in energy, calcium and phosphorus for a period of 28 days from day old. The combined effect of the enzymes significantly ($P<0.05$) improved broiler performance during starting phase but decreased in the later stages. This was in line with the work of Yuan *et al.* (23) who concluded that the growth of broilers was significantly improved during days 1- 21 but decreased in the later stage when non-starch polysaccharide (NSP) enzyme was combined with protease.

The carcass characteristics of broiler chickens fed enzyme supplemented diets are shown in Table 3. There was no significant (P>0.05) effect of the protease, phytase or their combination on the carcass characteristics of broiler chickens except for the drumstick which was significantly (P<0.05) higher in birds fed protease supplemented diets T4 and T8 compared to other treatments and the values ranged from 12.85 in T1 to 16.25% in T8. This finding was in line with the observation of Ajayi (18) who reported improvement in some of the carcass characteristics of broiler chickens fed enzyme-supplemented diets with reduced crude protein. The values for dressing percentage ranged from 69.08 – 77.89% with the control (T1) and enzyme-supplemented diet (T9) having the highest and lowest dressing percentages respectively. Although, there was no significant effect of enzyme supplementation on breast meat but birds fed protease supplemented diet (T4) had the highest numerical value among the birds fed diets with reduced crude protein. Also, the abdominal fat had increased numerical values as the crude protein inclusion decreased across the treatments. This observation may be due to high energy to protein ratio which led to energy in excess of what was required for growth due to relatively low protein content; hence, this energy was stored in the form of fat (24). The result of this study was in line with the findings of Malherios *et al.* (25) who concluded that broiler chickens fed with low protein diet had higher fat deposition compared to low lipid or carbohydrate diet. Also, in a comparison of low protein and normal protein diets in broiler chickens (26), it was found that low protein diets caused a significant increase in the abdominal fat content.

The nutrient digestibilities of the broiler chickens at the starter and grower phases are presented in Table 4. At the starter phase, the crude protein digestibility of the enzyme-supplemented diets at 21% and 19% CP differed significantly (P<0.05) from the non-enzyme supplemented diets, with T5 (PRO plus PHY) having the best nutrient digestibility at 21% CP. Also, there was a significant (P<0.01) difference in the ash values, and these ranged from 52.13 to 73.06 %. At the grower phase, there was a significant effect of the phytase and protease on dry matter digestibility (DM) at 18% CP, and phytase supplemented diet (T3) had the highest DM digestibility, followed by the protease supplemented diet (T4), while the non-enzyme supplemented diet (T6) had the lowest DM digestibility. At the finisher phase (Table 4), there were significant (P<0.05) differences in dry matter, crude protein and nitrogen free extract digestibilities, and the values ranged from 69.61 to 77.04%, 66.95 – 83.00%, and 88.01 – 94.91% respectively. Phytase supplemented diet (T3) had the highest crude protein digestibility, followed by protease supplemented diet (T4) at 16% CP. The improved nutrient digestibility observed in this study supported earlier findings (27) that supplementation of exogenous enzymes in young chicks' diets improved nutrient digestibility. The results also agreed with the study of Angel *et al.* (28) who found that addition of protease to low protein diet fed to birds for three weeks of age totally compensated for performance losses, resulting in enhancement of crude protein digestibility. The improvement in crude protein digestibility observed in this study could be as a result of a three-way interaction between phytate, proteolytic enzymes and protein-AA in the digestive contents, which consequently increased the apparent ileal digestibility of crude protein and amino acid (8) and (9). There was no significant (P>0.05) effect of enzyme-supplementation on crude protein digestibility at the grower phase and this agreed with the report (11) that enzyme-supplementation does not consistently improve crude protein and amino acid digestibilities.

Conclusion and Application

It was concluded that;

1. The addition of phytase, protease individually or in combination improved growth performance and nutrient digestibility of broiler chickens fed suboptimal crude protein diets.
2. There was a synergistic effect of phytase and protease supplementation on the growth performance and crude protein digestibility of broiler chickens fed suboptimal crude protein diet at starter phase.
3. Enzyme supplementation had no significant effect on carcass characteristics except the drumsticks, with birds fed protease-supplemented diets having the highest drumsticks.
4. Therefore, for optimal performance, 21% CP, 18% CP and 16% CP at starter, grower and finisher phases respectively may be adequate for broiler chickens if phytase or protease enzyme would be supplemented.

Table 2: Performance of broiler chicken fed sub-optimal levels of crude protein with or without enzyme supplementation at three phases[1]

Parameter	T1	T2	T3	T4	T5	T6	T7	T8	T9	SEM
0 – 21 days										
Initial body weight (g/bird)	47.92	47.78	47.85	47.58	48.18	47.58	47.88	47.82	45.85	0.25
Daily weight gain (g/bird)	21.68^{c}	22.47^{bc}	23.69^{ab}	23.46^{abc}	25.22^{a}	23.76^{ab}	22.99^{bc}	22.38^{bc}	22.69^{bc}	0.24
Daily feed intake (g/bird)	45.41^{b}	49.51^{a}	51.25^{a}	51.75^{a}	51.29^{a}	50.58^{a}	50.69^{a}	49.62^{a}	49.57^{a}	0.41
Feed conversion ratio	1.90	1.99	1.96	2.00	1.84	1.93	2.00	2.01	1.98	0.02
21- 42 days										
Initial body weight (g/bird)	503.29^{c}	519.61^{bc}	545.30^{ab}	540.29^{bc}	577.73^{a}	546.44^{ab}	530.63^{bc}	517.87^{bc}	522.24^{bc}	5.16
Daily weight gain (g/bird)	42.08^{abc}	40.61^{bc}	46.74^{a}	44.65^{ab}	41.64^{abc}	36.87^{c}	36.37^{c}	39.97^{bc}	39.08^{bc}	0.81
Daily feed intake (g/bird)	124.07^{a}	122.82^{ab}	129.42^{a}	122.32^{ab}	120.84^{ab}	119.12^{ab}	119.33^{ab}	112.79^{bc}	107.19^{c}	1.48
Feed conversion ratio	2.97	3.02	2.78	2.75	2.93	3.23	3.29	2.82	2.75	0.05
42 – 56 days										
Initial body weight (g/bird)	1387.06^{bcd}	1372.51^{bcd}	1526.91^{a}	1477.94^{ab}	1452.12^{abc}	1320.67^{cd}	1294.31^{d}	1357.21^{bcd}	1342.87^{bcd}	18.70
Daily weight gain (g/bird)	56.12^{abc}	53.66^{abc}	64.49^{a}	62.14^{a}	57.25^{ab}	40.77^{c}	44.85^{bc}	43.03^{bc}	46.35^{bc}	2.08
Daily feed intake (g/bird)	185.48^{abc}	186.62^{abc}	198.66^{a}	193.68^{a}	191.99^{ab}	177.59^{bc}	196.51^{a}	175.91^{c}	177.19^{bc}	2.07
Feed conversion ratio	3.47	3.48	3.11	3.20	3.43	4.41	4.51	4.09	3.84	0.14
0 – 56 days										
Initial body weight (g/bird)	47.92	47.78	47.85	47.58	48.18	47.58	47.88	47.82	45.85	0.25
Final body weight (g/bird)	2172.75^{bcd}	2123.76^{cde}	2429.73^{a}	2347.91^{ab}	2253.64^{abc}	1891.45^{f}	1922.18^{ef}	1959.67^{ef}	1991.73^{def}	40.33
Daily weight gain (g/bird)	37.94^{bcd}	37.07^{cde}	42.53^{a}	41.08^{ab}	39.39^{abc}	32.93^{f}	33.47^{ef}	34.14^{ef}	34.75^{def}	0.72
Daily feed intake (g/bird)	112.33^{abcd}	113.61^{abc}	119.92^{a}	116.07^{ab}	114.91^{abc}	110.27^{bcd}	115.32^{ab}	107.07^{cd}	105.23^{d}	1.08
Feed conversion ratio	2.86^{c}	2.95^{bc}	2.72^{c}	2.73^{c}	2.84^{c}	3.23^{ab}	3.33^{a}	3.03^{abc}	2.92^{bc}	0.05

[1]T1 (non-enzyme supplemented diet of 23, 21 and 18% CP at starting, growing and finishing phases respectively); T2 (non-enzyme supplemented diet of 21, 18 and 16% CP at starting, growing and finishing phases respectively); T3 (T2 plus PHY); T4 (T2 plus PRO); T5 (T2 plus PHY and PRO) and T6 (non-enzyme supplemented diet of 19, 16 and 14% CP at starting, growing and finishing phases respectively); T7 (T6 plus PHY); T8 (T6 plus PRO); T9 (T6 plus PHY and PRO) at the expense of maize.

^{abc} Means within the same row with different superscripts are significantly different ($p<0.05$), SEM; standard error of means

Table 3: Carcass Characteristics of broiler chickens fed enzyme-supplemented diets (%)

Parameter	TI	T2	T3	T4	T5	T6	T7	T8	T9	SE	P
Dressing %	77.89	72.55	71.37	72.07	70.57	74.94	71.35	70.45	69.08	0.7	0.26
Drumstick	12.85^{c}	14.68^{a}	15.05^{a}	16.21	15.28^{a}	14.58	15.04^{a}	16.25	15.67^{a}	0.2	0.01
Thigh	16.91	16.82	16.06	16.98	17.82	16.10	15.73	16.53	15.94	0.1	0.12
Wings	10.80	11.17	9.92	11.03	10.58	10.20	10.31	11.29	12.56	0.2	0.27
Breast	34.07	30.74	32.80	33.44	30.52	32.03	32.17	26.25	26.40	0.7	0.12
ABF	1.70	1.73	2.05	2.19	3.05	2.38	2.69	3.05	3.07	0.1	0.21

[1]T1 (non-enzyme supplemented diet of 23, 21 and 18% CP at starting, growing and finishing phases respectively); T2 (non-enzyme supplemented diet of 21, 18 and 16% CP at starting, growing and finishing phases respectively); T3 (T2 plus PHY); T4 (T2 plus PRO); T5 (T2 plus PHY and PRO) and T6 (non-enzyme supplemented diet of 19, 16 and 14% CP at starting, growing and finishing phases respectively); T7 (T6 plus PHY); T8 (T6 plus PRO); T9 (T6 plus PHY and PRO) at the expense of maize.

[abc] Means within the same row with different superscripts are significantly different ($p<0.05$)

ABF = Abdominal fat, % = percentage

Table 4: Nutrient digestibility of broiler chickens fed enzyme-supplemented diets at starter, growing and finisher phases (%)

Parameter	**T1**	T2	T3	T4	T5	T6	T7	T8	T9	SEM	P value
Starter											
Dry matter	81.00	76.63	79.24	79.19	81.34	76.43	80.38	82.37	84.85	1.16	0.84
Crude	87.34^{ab}	85.89^{bc}	86.80^{ab}	87.43^{ab}	89.75^{a}	82.72^{c}	89.25^{ab}	87.95^{ab}	87.12^{ab}	0.54	0.04
Crude fibre	25.47	13.26	21.55	14.36	14.21	13.71	19.15	19.80	18.21	1.21	0.18
Ether extract	84.56^{ab}	87.96^{a}	76.54^{b}	87.39^{a}	90.65^{a}	88.93^{a}	92.18^{a}	88.11^{a}	92.54^{a}	1.33	0.09
Ash	71.86^{a}	71.92^{a}	73.06^{a}	58.74^{c}	63.78^{b}	52.13^{d}	64.19^{b}	71.41^{a}	65.86^{b}	1.66	<.01
NFE	86.62	88.65	87.48	89.88	89.96	87.35	90.33	89.58	88.62	0.38	0.15
Grower											
Dry matter	71.88^{bc}	69.49^{cd}	76.29^{a}	72.09^{bc}	75.28^{ab}	67.44^{d}	69.32^{cd}	71.27^{bcd}	73.42^{abc}	0.61	0.01
Crude protein	81.09^{a}	76.25^{ab}	79.00^{a}	78.27^{a}	77.58^{a}	64.81^{b}	72.12^{a}	78.27^{a}	70.60^{ab}	1.14	0.12
Crude fibre	37.94	35.74	30.61	33.88	28.90	28.19	31.21	32.52	31.61	1.94	0.99
Ether	83.14	79.85	75.97	77.21	81.80	78.05	79.48	78.36	79.67	0.84	0.73
Ash	63.73^{ab}	56.49^{abc}	40.78^{bc}	45.78b^{c}	39.87^{c}	32.80^{c}	35.13^{c}	72.08^{a}	34.95^{c}	3.61	0.02
NFE	90.80^{b}	91.24^{b}	90.19^{b}	88.18^{b}	91.26^{b}	88.34^{b}	91.53^{b}	96.39^{a}	89.98^{b}	0.62	0.03
Finisher											
Dry matter	75.97^{ab}	70.75^{c}	72.82^{abc}	71.99^{bc}	77.04^{a}	73.02^{abc}	69.61^{c}	69.83^{c}	76.35^{ab}	0.66	0.01
Crude protein	77.79^{bc}	76.41^{c}	83.00^{a}	82.10^{ab}	76.34^{c}	66.95^{d}	77.17^{bc}	73.71^{c}	77.72^{bc}	1.12	0.01
Crude fibre	39.95	30.42	31.95	39.35	32.61	31.29	35.79	39.44	36.40	1.38	0.66
Ether extract	74.08^{cd}	81.04^{abc}	80.72^{abc}	77.31^{abcd}	83.69^{a}	74.94^{bcd}	82.42^{ab}	79.19^{abcd}	72.15^{d}	1.07	0.06
Ash	29.81^{bc}	45.47^{a}	39.15^{ab}	33.09^{abc}	23.01^{c}	27.41^{bc}	32.15^{abc}	33.20^{abc}	33.60^{abc}	1.83	0.14
NFE	88.01^{e}	90.03^{de}	90.07^{de}	94.91^{ab}	91.29^{bcd}	91.43^{bcd}	94.46^{a}	90.85^{cde}	93.29^{acd}	0.52	0.01

[1]T1 (non-enzyme supplemented diet of 23, 21 and 18% CP at starting, growing and finishing phases respectively); T2 (non-enzyme supplemented diet of 21, 18 and 16% CP at starting, growing and finishing phases respectively); T3 (T2 plus PHY); T4 (T2 plus PRO); T5 (T2 plus PHY and PRO) and T6 (non-enzyme supplemented diet of 19, 16 and 14% CP at starting, growing and finishing phases respectively); T7 (T6 plus PHY); T8 (T6 plus PRO); T9 (T6 plus PHY and PRO) at the expense of maize.

[abc] Means within the same row with different superscripts are significantly different ($p<0.05$)

NFE - Nitrogen free extract

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