Improving Phytate Phosphorus Utilization by Adding Phytase to Low Protein, Phosphorus and Calcium Corn-Soybean Broiler Diets

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Received: *20/12/2007*

Abstract: Two hundred and eighty eight one-day old unsexed Arbor Acres broiler chicks were used to evaluate the effect of microbial phytase enzyme supplementation to broiler corn-soybean meal diets low in crude protein (CP), inorganic P (IP) and Ca on the utilization of phytate P. Eight treatments with three replicates (12 birds/rep.) per treatment were used. Positive control and three basal diets were formulated. The positive control diet contained 22/20% CP, 0.45/0.39% IP and 1.00/0.90% Ca (starter/grower). Basal 1, 2 and 3, low CP, contained 20/18%; basal 2 and 3, low IP, contained 0.30/0.25%; basal 3, low Ca, contained 0.67/0.60%; (starter/grower). Microbial phytase was supplemented with 0 and 1000 FTU/kg diet for the control and the three basal diets. Birds growth performance, bone mineralization, carcass characteristics, blood parameters and economical efficiency were evaluated. Results of the overall period (1-6 wks) showed that phytase supplementation to control diet improved growth performance. Reducing CP (20/18%) only or with low IP (0.30/0.25%) decreased body weight gain (BWG) and daily feed intake (DFI) and worsened ($P \leq 0.05$) feed conversion ratio (FCR). However, the addition of phytase (1000 FTU/kg) to low CP only or with low IP diets increased BWG and improved ($P \leq 0.05$) FCR and values were comparable to those of the control. The reduction of Ca (0.67/0.60%) in bird low CP-IP diet decreased BWG and worsened ($P \leq 0.05$) FCR compared to that of the control. Supplementation of phytase to low CP-IP-Ca diet restored ($P \leq 0.05$) BWG and FCR and values were even better than those of the control. Tibia ash increased ($P \leq 0.05$) when phytase was added to all diets, with no significant differences between control and the three basal supplemented diets. Plasma Ca decreased ($P \leq 0.05$) when phytase was added to low CP-IP-Ca diet in comparison with all treatments. Phytase numerically elevated plasma P with significant differences only when added to low CP-IP diet. Phytase increased ($P \leq 0.05$) plasma protein when added to low CP diets. Except the abdominal fat, experimental treatments had no significant effect on carcass characteristics. While abdominal fat was increased ($P \leq 0.05$) with control + phytase diet, it was decreased ($P \leq 0.05$) with low CP-IP + Phytase diet. Supplementation of phytase to low CP, low CP-IP and Low CP-IP-Ca diets increased relative economical efficiency (REE) over those with no supplementation and over the control diet with phytase. In conclusion, phytase supplementation to low CP-IP-Ca broiler diet significantly improved BWG, FCR and tibia ash (%) and resulted in the highest REE.

Keywords: Broilers, protein, phosphorus, calcium, phytase, performance, bone ash.

INTRODUCTION

Protein, calcium and phosphorus are essential nutrients in the production of animal food. Protein and phosphorus are of special interest due to their excessive presence in manure causing environmental pollution problems. The majority of phosphorus in plant diets is contained in chemical structure, called phytic acid or its salt, which is known as phytate (Pallauf and Rimbach, 1997). Phytate P is relatively unavailable to poultry. Corn and soybean meal are commonly used in all practical diets for poultry. The National Research Council (1994) reported that corn has a total 0.28 % of phosphorous, whereas available phosphorus is only 0.08 %. In addition to reducing the phosphorus availability to poultry, phytates are associated with a number of anti-nutritional effects, largely because they can chelate divalent cations such as Ca, Mg, Fe, Zn, Cu and Mn and can also reduce protein availability (Ravindran *et al.*, 1995 and Bedford and Schulze, 1998). Phytic acid also reduces the activity of pepsin, trypsin and α- amylase (Sebastian *et al.*, 1998). Nutritionists have utilized inorganic P supplements (e. g., mono and dicalcium phosphates) over the years to meet the crucial need for supplemental P. Thus, an improvement in the utilization of phytate P would reduce the need to add inorganic P in feed and to a more important extent it decreases P and N excretion in the manure, therefore reduces the pollution problems.

Poultry are lacking in intestinal phytase (myo-inositol-hexaphosphate phosphohydrolase), the enzyme that hydrolyzes phytate to inositol and inorganic phosphorus (Sebastian *et al.*, 1998 and Yan *et al.*, 2001). Phytase was shown to improve the bioavailability of dietary P, Ca, Zn, Mn and Cu (Simons *et al.*, 1990 and Zanini and Sazzad, 1999). Dilger *et al.* (2004) concluded that microbial phytase improved performance, bone mineralization and P utilization in broiler chicks. In addition, Biehl and Baker (1997) reported that phytase have a small but significant positive effect on improving utilization of the essential amino acids in chicks fed corn-soybean meal basal diets. Improved utilization of dietary phytate P caused by phytase supplementation not only reduces inorganic P supplementation and reduces P excretion but also could enable producers to feed low protein diet, and reduce N excretion (El-Nagmy *et al.*, 2004). Rutherfurd *et al.* (2004) reported that microbial phytase improved phytate P utilization and total digestibility as well as true ileal amino acid digestibility for broiler chicks fed low P corn-soybean diet.

The influence of Ca on the efficacy of phytase was investigated on broiler by Sebastian *et al.* (1996); Driver *et al.* (2005); Bozkurt *et al.* (2006); Yan *et al.* (2006).

They observed that a wider Ca/P ratio depressed the weight response of broilers and turkeys fed suboptimum P and increased the amount of phytase needed. Broilers fed diets low in Ca and supplemented with phytase resulted in superior growth and mineral utilization compared with those fed on a diet with high Ca content. The addition of phytase improved body weight and tibia ash. At 0.5 and 0.7% Ca, dietary Ca was a limiting factor in maximizing tibia ash regardless of P level or phytase supplementation (Yan *et al.*, 2006).

The objective of this study was to evaluate the effect of microbial phytase enzyme supplementation to broiler corn-soybean meal diets low in crude protein, inorganic P and Ca on the utilization of phytate P. Growth performance, bone mineralization, carcass and blood parameters were also evaluated.

MATERIALS AND METHODS

The present study was carried out at the Poultry Experimental Farm, Faculty of Agriculture, Suez Canal University, Ismailia, Egypt.

Birds and management:

Two hundred and eighty eight one-day old unsexed Arbor Acres broiler chicks were obtained from a commercial hatchery (Ismailia Misr Poultry Company, Ismailia, Egypt). Chicks were weighed individually, wing-banded and randomly distributed into 24 groups of 12 chicks per pen. Chicks were caged in brooder batteries with wire mesh floors. Eight treatments with three replicates per treatment were used. Feed was supplied *ad-libitum* and birds had free access to water. The artificial light was provided 24 hours daily through the experimental period, which lasted for 6 weeks. All birds were kept under the same management conditions.

Diets and treatments:

Chicks were fed two types of diets, the starter diet from 1-3 wks of age and the grower diet from 4-6 wks of age. Experimental diets were corn-soybean meal formulated by using least-cost linear program of diet formulation. Except protein and energy, NRC (1994) was used as nutrient guideline for broiler chicken requirements. The composition and calculated analysis of the positive control and three different diets for starter and grower periods are shown in Table (1).

Eight treatments were designed to investigate the utilization of phytate P in corn-soybean meal diets as influenced by microbial phytase, crude protein (CP), inorganic phosphorous (IP) and calcium (Ca) levels. Growth performance, bone mineralization, carcass and blood parameters were evaluated. The positive control diet contained 22/20% CP, 0.45/0.39% IP and 1.00/0.90% Ca (starter/grower). Compared to the control diet, reductions in CP, IP and Ca were made in basal diets for starter and grower periods. Basal 1, 2 and 3 (low CP) contained 20/18%; basal 2 and 3 (low IP) contained 0.30/0.25%; basal 3 (low Ca) contained 0.67/0.60% (starter/grower).

Microbial phytase was supplemented with 0 and 1000 units per kg diet (FTU/kg) for the control and the three basal diets. All diets were isocaloric (3000 kcal ME/kg diet) and fed in mash form. One unit FTU is defined as the quantity of enzyme that liberates 1 µmol of IP/min from 0.0015 sodium phytate at pH 5.5 and 37°C.

Growth performance:

Birds were observed daily for mortality. Birds individual body weight (BW) and pen feed consumption were recorded weekly. Body weight gain (BWG), average daily feed intake (DFI) and feed conversion ratio (FCR) were calculated. At the end of the study, the economic efficiency was calculated from the input-output analysis based upon the difference in both growth rate and feeding cost.

Blood, bone and carcass measurements:

At the end of the experimental period (6 weeks of age), 2 birds were chosen near to the average body weight from each replicate. Selected birds were individually weighed and slaughtered to a complete bleeding. Blood samples were collected from each bird in heparinized tube at slaughtering. Plasma was separated by centrifugation at 3000 rpm for 15 minutes and stored at -20°C for subsequent analysis. Protein, Ca, and P were determined in plasma by colorimetric method using commercial kits, following the procedure described by the manufacture.

The center toes of the left and right feet were removed at the fourth joint from the claw. All the toes within each replicate of each treatment were pooled. The pooled samples were dried to a constant weight at 100°C and ashed in a muffle furnace at 550°C for 24 hours. Toe ash was expressed as a percentage of dry weight (Bliss, 1945). The left tibia from each bird was separated. All adhering tissues were removed, bones were dried, ground, fat extracted, then dried and weighed. The dry fat-free bones were ashed as described before. Tibia ash was expressed as a percentage of the fat-free bone weight.

Slaughtered birds were used to evaluate carcass characteristics. Eviscerated carcass, liver, heart, spleen, empty gizzard and abdominal fat were weighed individually and recorded to the nearest gram. Eviscerated carcass (dressing), giblets, total edible portions (carcass weight plus giblets weight), spleen, and abdominal fat were expressed as a percentage of the live body weight.

Statistical analysis:

The experimental design was completely randomized. Data were statistically analyzed using General Linear Models (GLM) procedures of SAS (SAS institute, 1998). Means comparisons between treatments were performed using Duncan's multiple-range test (Duncan, 1955).

RESULTS AND DISCUSSION

Growth performance:

Starting period:

Effects of different treatments on body weight (BW), body weight gain (BWG), daily feed intake (DFI) and feed conversion ratio (FCR) during the starting period (1-3 weeks of age) are shown in Table (2).

There were no significant differences in average one day old BW of chicks with an overall mean of 50.9 g (data not shown). In comparison with the control diet (T1), lowering CP (20%; T3) significantly reduced birds BW and BWG. Reducing IP (0.30%) with low CP (T5) gave the lowest ($P \leq 0.05$) BW, BWG and DFI, compared with those of the control diet (T1) and the low CP diet (T3). The poorest FCR resulted from feeding chicks diet No. 3 and 5 but data showed no significant differences in FCR as a result of feeding diets 1, 2, 3, 4, 5, 7 & 8. In addition, feeding diet of low Ca (0.67%) and low CP-IP (T7) had no significant effect on growth performance in comparison with that of the control diet. Supplementation of phytase (1000 FTU/kg diet) to control (T2), low CP (T4) and low CP-IP-Ca (T8) diets improved growth performance of broiler chicks over that for chicks fed the same diets without phytase supplementation with no significant differences between treatments supplemented with phytase (T2, 4, & 8) and the others without phytase (T1, 3 & 7). However, supplementation of phytase to low CP-IP diet (T6) significantly improved ($P \leq 0.05$) BW, BWG, DFI and FCR compared with no phytase supplementation (T5). Also, the improvements in BW, BWG and FCR resulted from phytase supplementation to low CP-IP (T6) diet were significantly better than those of the low CP diet without phytase. The best growth performance resulted from feeding birds control + phytase (T2) followed by low CP-IP-Ca + phytase (T8), control (T1) and low CP-IP + phytase (T6) diets.

Growing period:

Effects of different treatments on BW, BWG, DFI and FCR during the growing period (4-6 weeks of age) are shown in Table (3).

In comparison with the control diet (T1), lowering CP level (18%; T3) reduced ($P \leq 0.05$) BW, BWG and worsened FCR. Reducing IP (0.25%) with low CP (T5) significantly reduced BW and worsened FCR. The reduction in BWG and DFI was not significant in comparison with that of the control. There were no significant difference in growth performance between birds fed low CP (T3) and low CP-IP (T5) diets. In addition, lowering Ca level (0.60%) with low CP-IP (T7) resulted in poor ($P \leq 0.05$) FCR, besides, BW, BWG, and DFI decreased with no significant differences compared to those of the control diet (T1). Birds fed low CP-IP-Ca (T7) had better ($P \leq 0.05$) BW (compared with T3 & 5) and FCR (compared with T3). Diets supplemented with phytase (1000 FTU/Kg diet; T2, 4, 6, & 8) improved broiler chicks' growth performance. There were significant differences in BW between diets supplemented with phytase (T2), Low CP-IP (T6) and low CP-IP-Ca (T8) in comparison with those fed diets with no phytase supplementation (T1, 5 & 7). BWG improved significantly when phytase was supplemented to low CP-IP-Ca diet (T8) compared with no phytase supplementation (T7). FCR improved ($P \leq 0.05$) with phytase supplementation to low CP (T4) and low CP-IP-Ca (T8) diets, in comparison with those of T3 and T7 diets (no phytase supplementation). The highest BW and BWG resulted from birds fed control + phytase diet (T2), followed by low CP-IP-Ca + phytase (T8) and control (T1) diets. However, the best FCR resulted from birds fed low CP-IP-Ca + phytase (T8) diet in comparison with those fed T3, T4, T5 & T7 diets. Birds fed control diet + phytase (T2) had the highest DFI.

Overall period:

Effects of different treatments on BW, BWG, DFI and FCR during the overall period (1-6 weeks of age) are shown in Table (4).

In comparison with the control diet (T1), lowering CP (20/18%; T3), IP (0.30/0.25%) with low CP (T5) and Ca (0.67/0.60%) with low CP-IP (T7) in birds diet reduced overall BWG with significant differences observed only between T1 & T5. Phytase supplementation (1000 FTU/kg diet; T2, 4, 6 & 8) improved overall BWG. In addition, supplementation of phytase to low CP-IP-Ca (T8) significantly improved overall BWG compared with no phytase supplemented diet (T7), low CP-IP with/out phytase (T6 & 5) and low CP with/out phytase (T4 & 3) diets. The highest overall BWG resulted from birds fed control diet + phytase (T2) followed by low CP-IP-Ca + phytase (T8) and control (T1) diets. Birds fed control diet + phytase (T2) had the highest ($P \leq 0.05$) DFI compared with those fed low CP with/out phytase (T4 & 3) and low CP-IP with/out phytase (T6 & 5) diets. The poorest ($P \leq 0.05$) FCR resulted from birds fed low CP (T3) followed by low CP-IP (T5) then low CP-IP-Ca (T7) diets, compared with those of the rest of the treatments. Phytase supplementation to low CP (T4), low CP-IP (T6) and low CP-IP-Ca (T8) diets significantly improved FCR. In addition, supplementation of phytase to control diet (T2) resulted in the highest DFI while FCR was comparable to that of the control diet (T1). The best FCR resulted from feeding birds low CP-IP-Ca + phytase (T8) diet followed by control diet with/out phytase (T2 & 1) and low CP-IP + phytase (T6) diet with no significant differences between these treatments. The overall mortality rate was 2.43% which was in normal average. The highest mortality (8.33%) resulted from feeding birds low CP diet (T3) followed by control diet + phytase (5.55). There were no significant differences in birds' mortality (%) as a result of treatments 1, 2, 4, 5, 6, 7 & 8.

Data reported herein show that phytase supplementation to control diet (T2) numerically improved growth performance over that of the control (T1) with significant differences in BW during the growing period (4-6 wks) only. Hydrolysis and subsequent utilization of phytate-associated nutrients including protein, lipids, carbohydrates, and minerals may be involved in these growth improvements (Ravindran *et al.*, 1995). In agreement with that, Ahmed *et al.* (2000) and Yan *et al.* (2000) noted no significant effect on FCR due to phytase supplementation to broiler diets. On the other hand, Simons *et al.* (1990) and Yan *et al.* (2001) reported that phytase supplementation improved dietary phytate P bioavailability. Also, El-Ghamry *et al.* (2005) stated that phytase addition to broiler commercial corn-soybean diets improved BWG.

Birds BW, BWG and DFI decreased whereas FCR and mortality were worsened upon the reduction of CP

(20/18%) in birds' diet in comparison with those of the control with/out phytase. However, the addition of phytase (1000 FTU/kg) to low CP diets improved birds BW, BWG and decreased DFI. FCR and livability improved significantly as a result of phytase addition to the low CP diet and the values were close to those of the control diet. In harmony to the data stated herein, Biehl and Baker (1997), Sebastian *et al.* (1997) and Kornegay *et al.* (1998) showed that the addition of 300-400 FTU phytase/kg low CP diets restored the reduced performance and improved ileal AA digestibility. Also, livability, growth and FCR were improved by the addition of phytase to low CP diets (Waldroup *et al.*, 2000). Also and in agreement to present data, Attia *et al.* (2001) noticed that reducing CP decreased broiler growth performance, whereas birds' growth and FCR were totally recovered by phytase addition at 700 FTU/kg diet. The improvement in growth rate upon the addition of phytase to low CP diet was mainly due to improving nutrients liberation (Ravindran *et al.*, 2000) and utilization of phytate P. Besides and in agreement to the data found here, Abd-Elsamee (2002) demonstrated that phytase supplementation to diet containing medium level of CP improved ($P<0.05$) the average values of broiler performance in comparison to that of the control group. Some other investigators reported that adding phytase to broiler chick diets had positive impact on protein availabilities of broiler diets and improved performance (Simons *et al.*, 1990, Sebastian *et al.*, 1997, and Attia *et al.*, 2001). The improvement in bird performance and FCR upon the addition of phytase to low CP diet could be explained that phytase prevents the formation of protein-phytate complexes by the prior hydrolysis of phytate and thus birds get the full benefit of the CP and the other nutrients from phytate hydrolysis (Selle *et al.*, 2006).

The data stated herein show that bird BW, BWG, DFI and FCR were worsened and mortality was in normal average upon feeding broilers low CP-IP diet (T5), whereas chicks growth performance and livability were improved upon feeding birds low CP-IP diet + phytase (T6) and the data were comparable to those of the control. In harmony with that, Bozkurt *et al.* (2006) mentioned that FCR of broilers fed low CP-IP diet + phytase was significantly better than that of broilers fed low CP-IP diet. However, they reported that neither phytase supplementation nor diet nutrient density and dietary P level had a significant effect on broiler mortality. On the contrary, they concluded that phytase (500 FTU/kg diet) could not compensate for reduced CP even if this was slightly lower than the level of standard control diet. The level of enzyme, and IP used in the present study were different from those of Bozkurt *et al.* (2006). Ravindran *et al.* (2000) stated that phytase is more efficacious in diets containing low level of inorganic P, presumably high P level inhibits phytase activity and reduces the extent of phytate hydrolysis.

The reduction of Ca (0.67/0.60) in birds low CP-IP diet did not cause significant negative effect on birds' performance, DFI, or mortality rate in comparison with that of the control. However, FCR was worsened during the growing period only and as a result of that the overall (1-6 wks) FCR was significantly poorer than that of the control. Phytase supplementation to low CP-IP-Ca diet restored birds FCR and all parameters measured were comparable ($P \leq 0.05$) or even numerically better than that of the control. In harmony with that, Mohamed *et al.* (2005) found that the addition of phytase to corn-soybean meal broiler diets low in IP and Ca significantly increased BWG and FI of 5 weeks old birds. In addition, El-Sherbiny *et al.* (2005) stated that decreasing dietary Ca level increased BW, slightly decreased FI and significantly improved FCR. Addition of phytase increased ($P \leq 0.05$) weight gain, FI and improved FCR, which agree with the present data. They also stated that birds fed 1.00/0.90% Ca (starter/grower) diet without phytase addition gained ($P \leq 0.05$) less weight compared to those fed diets containing 0.65/0.55% or 0.85/0.75% Ca with phytase addition. Birds fed diet containing 0.85/0.75% Ca + phytase recorded the highest BWG in comparison with birds fed lower (0.65/0.55%) or higher (1.00/0.90%) dietary Ca without phytase. This could be explained that increasing Ca ion concentrations reduces the extent of protein-phytate complex formation in soybeans as Ca competes with protein for the binding sites of phytate (Prattley *et al.*, 1982). In addition, Wise (1983) noted that Ca has negative effect on phytase efficacy because the formation of insoluble Ca-phytate complexes renders the substrate less susceptible to phytase activity.

Bone ash and plasma blood measurements:

Effects of different treatments on bone ash (tibia & toe) and plasma blood (Ca, P & protein) parameters at the end of the experimental period are presented in Table (5).

Tibia and toe ash (%):

Lowering CP level (20/18%; T3), IP (0.30/0.25%) with low CP (T5) and Ca (0.67/0.60%) with low CP-IP (T7) in the diet significantly decreased tibia bone ash compared with that of the rest of the treatments. Phytase supplementation (1000 FTU/kg diet) to low CP (T4), low CP-IP (T6) and low CP-IP-Ca (T8) diets significantly increased tibia bone ash (%) to values comparable to those of the control (T1). Phytase supplementation to control diet (T2) significantly increased tibia bone ash to values superior to those of the control (T1). There were no significant differences in toe ash as a result of the different diets. The highest numerical improvements in toe ash were observed in control diet + phytase (T2) followed by control (T1) and the low CP-IP-Ca + phytase (T8) diets.

In harmony to present findings, Sebastian *et al.* (1996) and Bozkurt *et al.* (2006) demonstrated that the percentages of tibia ash increased significantly by the addition of phytase to low CP-IP diet. Sohail and Roland (1999) and Attia *et al.* (2003) indicated that supplementing phytase in grower diets containing reduced levels of IP and Ca significantly improved broiler bone strength. Also and similarly to the data stated herein and previous reports, Driver *et al.* (2005) noted that broiler bone quality responses to phytase were greatest at low IP levels and high Ca levels and these responses decreased when Ca level was reduced and IP increased. On the contrary, El-Sherbiny *et al.* (2005) stated that decreasing dietary IP-Ca increased tibia and toe ash (%) significantly. However, and in agreement to the data stated herein, they reported that phytase supplementation to low IP-Ca diet increased tibia ash ($P \leq 0.05$). Besides, Mohamed *et al.* (2005) and Yan *et al.* (2006) noticed that phytase addition to low IP-Ca diets increased tibia ash significantly. Phytase addition to diets with 0.30 or 0.35% IP increased tibia ash at 0.7 and 0.9% Ca, whereas no effect was seen with 0.5% Ca, which indicates that phytase had minimum effects in releasing Ca (Yan *et al.*, 2006). In the present study it was found that phytase had superior effect on tibia ash when added to diets with 20/18% CP + 0.30/0.25% IP + 0.67/0.60% Ca (starter/grower). Yan *et al.* (2006) stated that Ca to total P ratio in the diet ranging from 1.5 – 2.16 did not seem to negatively affect the magnitude of responses. The ratio used herein between Ca and IP was 2.0. Excess of Ca over that ratio could affect phytase efficiency by binding phytate or depressing phytase activity. The improvement in tibia ash (%) with phytase addition to the diets could be considered as a good indication of increased bone mineralization (Sebastian *et al.*, 1996). This increment in tibia ash as phytase added to the control, low CP, low CP-IP, or low CP-IP-Ca diets could be attributed to the release of inorganic P and perhaps other nutrients from the phytate molecule, by phytase enzyme, and subsequently increase phosphorus and other nutrients availability and its utilization by bone (Watson *et al.*, 2006).

Plasma blood measurements:

While plasma Ca of birds fed low CP-IP-Ca + phytase diet (T8) was lower ($P \leq 0.05$) than that of the rest of the treatments, no significant differences were noticed in bird plasma Ca as affected by the rest of the different diets (T1to T7). Birds fed low CP-IP (T5) diet had the lowest ($P \leq 0.05$) plasma P compared with that of the rest of the treatments. Phytase supplementation (1000 FTU/kg diet) to low CP-IP (T6) diet increased ($P \leq 0.05$) plasma P to a level comparable with the low CP (T3) diet. This increment was lower ($P \leq 0.05$) than the values resulted from feeding birds control with/out phytase (T2 & 1), low CP with phytase (T4) and low CP-IP-Ca with/out phytase (T8 & 7) diets. There were no significant differences in plasma P as a result of feeding birds the rest of the diets (T1, 2, 3, 4, 7 & 8). Numerical reduction in bird plasma protein resulted when birds were fed low CP (T3), low CP-IP (T5), and low CP-IP-Ca with/out phytase (T8 & 7) diets compared to the control diet with/out phytase (T2 & 1) and low CP-IP with phytase (T6) diet. Supplementation of phytase to low CP (T4) diet resulted in the highest ($P \leq 0.05$) plasma protein compared to low CP (T3) and low CP-IP-Ca with/out phytase (T8 & 7) diets. In agreement to the data stated herein, Roberson and Edwards (1994) reported that phytase addition had no significant effect on plasma Ca. In contrary to present data, Attia *et al.* (2001) stated that plasma Ca increased significantly when phytase (700 FTU/kg) was supplemented to intermediate protein diet. No significant differences in plasma Ca were noticed in this study between the control, low CP or low CP + phytase diets.

Carcass characteristics:

Effects of different treatments on carcass characteristics as a percentage of live body weight (dressing, giblets, total edible parts, abdominal fat and spleen) at the end of the experimental period are presented in Table (6).

No significant differences were noticed in each of the dressing, giblets, edible parts or spleen percentages of birds supplemented with different treatments which is in agreement with Kornegay *et al.* (1998), Abd-Elsamee (2002) and El-Medany and El-Afifi (2002) who stated that low CP diet with/out phytase had no significant effect on carcass characteristics. Also, El-Sherbiny *et al.* (2005) found that dressing (%), liver, heart and gizzard (% of BW) were not affected by phytase addition to low IP-Ca diets which is in harmony with present data. However, Abd El-Hakim and Abd-Elsamee (2004) and El-Husseiny *et al.* (2006) noticed that chicks fed low IP diet + phytase recorded the highest dressing (%) compared to those fed the control diet.

On the other hand, some treatments affected ($P \leq 0.05$) bird abdominal fat (%). In comparison with the control diet, lowering CP level (20/18%; T3), IP (0.30/0.25%) with low CP (T5) and Ca (0.67/0.60%) with low CP-IP (T7) resulted in increasing ($P \leq 0.05$) abdominal fat (%). This was explained by Summers *et al.* (1992) who stated that by reducing dietary crude protein, the energy used for degradation of protein was spared and stored as fat. Although supplementation of phytase (1000 FTU/kg diet) to control diet (T2) increased ($P \leq 0.05$) abdominal fat%, it decreased ($P < 0.05$) abdominal fat (%) of birds when added to their low CP-IP (T6) diet. However, supplementation of phytase to low CP (T4) and low CP-IP-Ca (T8) diets had no significant effect on bird abdominal fat. This agrees with the data reported by El-Sherbiny *et al.* (2005). They stated that the addition of phytase significantly decreased abdominal fat (%). On the other hand, Mohamed *et al.* (2005) noted that abdominal fat (%) was not affected significantly by phytase addition to low IP-Ca diets.

Relative economical efficiency (REE):

Effects of different treatments on REE at the end of the experimental period are presented in Table (7).

Although birds fed control diet supplemented with phytase (1000 FTU/kg diet) had the highest BW, those birds revealed the lowest REE. This was due to the high feed cost while bird FCR was comparable with those fed on the control diet. Supplementation of phytase to low CP (T4), low CP-IP (T6) and Low CP-IP-Ca (T8) diets increased REE over those with no supplementation and over the control with phytase (T2) diets. The highest REE was recorded with birds fed low CP-IP-Ca + phytase (T8) followed by those fed low CP-IP + phytase (T6), low CP-IP-Ca (T7), and low CP + phytase (4) diets. These improvements were due to the combined effect of low feed cost with increased BW and improved FCR.

Table (1): Composition and calculated analysis of the experimental diets fed during starter and grower periods.

Ingredients%	Starter diets				Grower diets			
	Contol	Low CP	Low CP - IP	Low CP-IP-Ca	Control	Low CP	Low CP- IP	Low CP-IP-Ca
Yellow corn	55.76	61.75	61.95	63.04	62.52	69.02	69.14	70.07
Soybean meal (44% CP)	34.01	30.00	30.90	31.70	30.00	25.21	25.85	26.15
Corn gluten (60% CP)	3.70	2.50	1.80	1.10	2.43	1.73	1.23	1.00
Sunflower oil	2.66	1.90	1.90	1.60	1.66	0.70	0.70	0.50
Dicalcium phosphate	1.72	1.80	0.90	0.90	1.40	1.30	0.68	0.68
Limestone	1.42	1.40	1.90	1.00	1.34	1.43	1.78	0.98
Salt	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35
Vit. & Min. Mixture*	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
DL-methionine	0.13	0.05	0.05	0.06	0.05	0.01	0.02	0.02
Total	**100**	**100**	**100**	**100**	**100**	**100**	**100**	**100**
Calculated analysis:**								
ME (kcal/kg diet)	3000.00	2996.09	2996.82	2999.40	3000.00	3000.00	3000.00	3011.78
Crude protein	22.00	19.97	19.97	20.00	20.00	18.00	18.00	18.07
Calcium	1.00	1.00	1.01	0.67	0.90	0.90	0.91	0.60
Inorganic phosphorus	0.45	0.46	0.30	0.30	0.39	0.38	0.25	0.25
Lysine	1.10	0.99	1.01	1.03	1.00	0.88	0.89	0.90
Methionine	0.49	0.38	0.38	0.39	0.39	0.32	0.32	0.32
Methionine + cyctine	0.91	0.78	0.78	0.78	0.78	0.69	0.69	0.69

* Each kg of Vit. & Min. Mixture contains: Vit A 4800000 IU, Vit D_3 800000 IU, Vit E 4g, Vit K_3 400mg, Vit B_1 400mg, Vit B_2 2g, Vit B_6 600mg, Vit B_{12} 4mg, Pantothenic acid 4g, Niacin 12g, Biotin 20mg, Folic acid 400mg, Choline chloride 240g, Iron 12g, Manganese 24g, Zinc 20g, Copper 2g, Cobalt 40mg, Iodine 120mg and Selenium 40mg. $CaCo_3$ was used as carrier.
** Calculation are based on tables of NRC (1994).

Table (2): Effects of dietary crude protein (CP), inorganic phosphorus (IP), calcium (Ca) and phytase levels on growth performance of broiler chicks (mean ± SE) during the starting period (1-3 wks).

T #	Diets	Phytase (FTU/kg)	Body weight (g)	Body weight gain (g)	Daily feed intake (g)	Feed conversion (g feed/g gain)
1	**Control**	**0**	613.1 ± 09.1^{ab}	561.9 ± 08.9^{ab}	43.78 ± 0.63^{ab}	1.64 ± 0.01^{ab}
2	**As T1 +**	**1000**	635.1 ± 09.0^{a}	584.2 ± 08.8^{a}	45.43 ± 1.07^{a}	1.63 ± 0.03^{ab}
3	**Low CP (20%)**	**0**	580.1 ± 11.0^{c}	530.4 ± 11.0^{c}	42.64 ± 0.67^{ab}	1.69 ± 0.02^{a}
4	**As T3 +**	**1000**	593.1 ± 06.5^{bc}	542.0 ± 06.4^{bc}	42.14 ± 0.28^{b}	1.63 ± 0.02^{ab}
5	**Low CP-IP (0.30%)**	**0**	532.7 ± 11.9^{d}	481.4 ± 11.6^{d}	38.66 ± 0.17^{c}	1.69 ± 0.01^{a}
6	**As T5 +**	**1000**	611.4 ± 06.6^{ab}	559.8 ± 06.4^{ab}	42.86 ± 0.32^{ab}	1.61 ± 0.01^{b}
7	**Low CP-IP-Ca (0.67%)**	**0**	610.6 ± 10.5^{ab}	559.4 ± 10.2^{ab}	44.32 ± 0.65^{ab}	1.66 ± 0.02^{ab}
8	**As T7 +**	**1000**	618.8 ± 10.4^{ab}	568.3 ± 10.1^{ab}	44.69 ± 2.14^{ab}	1.64 ± 0.02^{ab}
	Overall mean ± SE		599.3 ± 3.8	548.4 ± 03.7	43.07 ± 0.50	1.65 ± 0.01

a-d Numbers with different superscript in each column are significantly different at $P \leq 0.05$.

Table (3): Effects of dietary crude protein (CP), inorganic phosphorus (IP), calcium (Ca) and phytase levels on growth performance of broiler chicks (mean ± SE) during the growing period (4-6 wks).

T #	Diets	Phytase (FTU/kg)	Body weight (g)	Body weight gain (g)	Daily feed intake (g)	Feed conversion (g feed/g gain)
1	**Control**	**0**	1624.4 ± 25.1^{bc}	1011.3 ± 21.5^{ab}	106.91 ± 2.17^{abc}	2.22 ± 0.01^{cd}
2	**As T1 +**	**1000**	1728.6 ± 23.6^{a}	1092.3 ± 19.4^{a}	114.59 ± 2.20^{a}	2.21 ± 0.05^{cd}
3	**Low CP (18%)**	**0**	1445.6 ± 24.7^{e}	865.5 ± 22.8^{c}	102.75 ± 2.48^{bc}	2.49 ± 0.01^{a}
4	**As T3 +**	**1000**	1509.1 ± 28.6^{de}	916.7 ± 27.8^{bc}	99.02 ± 4.13^{c}	2.29 ± 0.08^{bc}
5	**Low CP-IP (0.25%)**	**0**	1447.7 ± 31.8^{e}	915.5 ± 25.6^{bc}	104.19 ± 3.27^{bc}	2.30 ± 0.02^{ab}
6	**As T5 +**	**1000**	1558.3 ± 12.6^{cd}	946.8 ± 13.4^{bc}	102.29 ± 1.05^{bc}	2.27 ± 0.05^{bcd}
7	**Low CP-IP-Ca (0.60%)**	**0**	1554.7 ± 35.4^{cd}	944.1 ± 29.7^{bc}	105.92 ± 1.64^{bc}	2.36 ± 0.03^{b}
8	**As T7 +**	**1000**	1678.9 ± 30.3^{ab}	1060.1 ± 23.6^{a}	108.57 ± 3.75^{ab}	2.14 ± 0.01^{d}
	Overall mean ± SE		1568.4 ± 11.1	969.0 ± 09.3	105.53 ± 1.20	2.30 ± 0.03

a-e Numbers with different superscript in each column are significantly different at $P \leq 0.05$.

Table (4): Effects of dietary crude protein (CP), inorganic phosphorus (IP), calcium (Ca) and phytase levels on growth performance of broiler chicks (mean ± SE) during the overall period (1-6 wks).

T #	Diets	Phytase (FTU/kg)	Body weight gain (g)	Daily feed intake (g)	Feed conversion (g feed/g gain)	Mortality rate%
1	Control	0	1573.3 ± 25.0^{ab}	75.35 ± 1.29^{abc}	2.01 ± 0.01^{cd}	0^{b}
2	As T1 +	1000	1677.7 ± 23.4^{a}	80.01 ± 0.64^{a}	2.01± 0.02^{cd}	5.55^{ab}
3	Low CP (20/18%)	0	1395.9 ± 24.9^{bc}	72.70 ± 0.38^{bc}	2.19± 0.01^{a}	8.33^{a}
4	As T3 +	1000	1458.0 ± 28.4^{bc}	70.58 ± 2.01^{c}	2.04± 0.03^{c}	0^{b}
5	Low CP-IP (0.30/0.25%)	0	1396.2 ± 31.6^{c}	71.43 ± 1.72^{c}	2.14± 0.01^{ab}	2.78^{b}
6	As T5 +	1000	1506.6 ± 12.4^{bc}	72.58 ± 0.66^{bc}	2.03± 0.02^{cd}	0^{b}
7	Low CP-IP-Ca (0.67/0.60%)	0	1503.5 ± 35.1^{bc}	75.12 ± 0.52^{abc}	2.10± 0.01^{b}	0^{b}
8	As T7 +	1000	1628.5 ± 29.9^{a}	76.63 ± 2.90^{ab}	1.97± 0.01^{d}	2.78^{b}
	Overall mean ± SE		1517.4 ± 11.1	74.30 ± 0.75	2.06 ± 0.02	2.43 ± 0.79

$^{a-c}$ Numbers with different superscript in each column are significantly different at $P \leq 0.05$.

Table (5): Effects of dietary crude protein (CP), inorganic phosphorus (IP), calcium (Ca) and phytase levels on bone ash and plasma blood parameters of broiler chicks at the end of the experiment (mean ± SE).

T #	Diets	Phytase (FTU/kg)	Bone ash%		Plasma blood measurements		
			Tibia	Toe	Ca (mg/dl)	P (mg/dl)	Protein (g/dl)
1	Control	0	51.2 ± 0.4^{b}	12.1 ± 0.5	11.0 ± 0.8^{a}	3.6 ± 0.3^{a}	5.5 ± 0.8^{ab}
2	As T1 +	1000	54.0 ± 0.6^{a}	12.3 ± 0.2	12.8 ± 0.8^{a}	4.1 ± 0.1^{a}	5.3 ± 0.6^{ab}
3	Low CP (20/18%)	0	48.2 ± 0.9^{c}	11.4 ± 0.3	12.4 ± 0.5^{a}	3.5 ± 0.5^{ab}	4.3 ± 0.7^{b}
4	As T3 +	1000	51.1 ± 0.6^{b}	11.9 ± 0.4	12.7 ± 0.9^{a}	4.3 ± 0.4^{a}	6.2 ± 0.3^{a}
5	Low CP-IP (0.30/0.25%)	0	46.6 ± 0.8^{c}	11.1 ± 0.2	11.3 ± 0.5^{a}	1.4 ± 0.1^{c}	4.5 ± 0.7^{ab}
6	As T5 +	1000	51.1 ± 0.4^{b}	11.5 ± 0.8	11.4 ± 0.4^{a}	2.6 ± 0.2^{b}	6.0 ± 0.5^{ab}
7	Low CP-IP-Ca (0.67/0.60%)	0	46.2 ± 1.5^{c}	11.3 ± 1.3	11.7 ± 0.2^{a}	3.8 ± 0.4^{a}	4.1 ± 0.4^{b}
8	As T7 +	1000	52.3 ±0.4^{ab}	12.1 ± 0.3	8.5 ± 0.5^{b}	4.4 ± 0.3^{a}	4.3 ± 0.6^{b}
	Overall mean ± SE		50.1 ± 0.6	11.7 ± 0.2	11.5 ± 0.3	3.5 ±0.2	5.0 ± 0.2

$^{a-c}$ Numbers with different superscript in each column are significantly different at $P \leq 0.05$.

Table (6): Effects of dietary crude protein (CP), inorganic phosphorus (IP), calcium (Ca) and phytase levels on carcass characteristics of broiler chicks at the end of the experiment (mean ± SE).

T #	Diets	Phytase (FTU/kg)	Traits %				
			Dressing	Giblets	Total edible parts	Abdominal fat	Spleen
1	Control	0	68.83 ± 0.60	5.14 ± 0.33	73.97 ± 0.26	1.17 ± 0.27^{c}	0.25 ± 0.02
2	As T1 +	1000	68.88 ± 0.91	5.31 ± 0.16	74.19 ± 0.75	2.23 ± 0.28^{ab}	0.25 ± 0.05
3	Low CP (20/18%)	0	67.30 ± 1.06	4.78 ± 0.19	72.08 ± 1.25	2.34 ± 0.08^{a}	0.20 ± 0.01
4	As T3 +	1000	71.25 ± 1.75	4.97 ± 0.30	76.21 ± 1.44	2.35 ±0.47^{a}	0.27 ± 0.08
5	Low CP-IP (0.30/0.25%)	0	65.28 ± 0.32	4.87 ± 0.30	70.15 ± 0.61	2.47 ± 0.06^{a}	0.24 ± 0.01
6	As T5 +	1000	68.89 ± 1.26	5.02 ± 0.17	73.91 ± 1.10	1.48 ± 0.28^{c}	0.20 ± 0.01
7	Low CP-IP-Ca (0.67/0.60%)	0	69.76 ± 0.82	4.67 ± 0.49	74.42 ± 1.30	2.07 ± 0.09^{ab}	0.19 ± 0.02
8	As T7 +	1000	69.79 ± 1.10	4.91 ± 0.21	74.69 ± 0.89	1.93 ± 0.02^{abc}	0.16 ± 0.01
	Overall mean ± SE		68.75 ± 0.64	4.96 ± 0.09	73.70 ± 0.63	2.01 ± 0.13	0.22 ± 0.01

$^{a-c}$ Numbers with different superscript in each column are significantly different at $P \leq 0.05$.

In agreement to the data stated herein, Attia *et al.* (2001) concluded that a satisfactory EE could be achieved by decreasing CP level up to 10% (-2% CP) when phytase was supplemented at 700 FTU/kg diet. In addition, Abd-Elsamee (2002) noted that the best values of EE were observed by feeding broiler diets containing medium level of CP + phytase. This improvement in EE could be attributed to the improvement in both growth rate and FCR with added phytase. Phytase reduces feed cost because of the improvements in FCR and the faster growth rate of broilers (Kies *et al.*, 2001).

According to the above findings, it can be concluded that for improving broiler performance, tibia ash, and REE it is recommended to decrease dietary CP (20/18%), IP (0.30/0.25%) and Ca (0.67/0.60%) levels during the starter and grower periods with addition of phytase (1000 FTU/kg diet).

Table (7): Effects of dietary crude protein (CP), inorganic phosphorus (IP), calcium (Ca) and phytase levels on economical efficiency of broiler chicks at the end of the experiment (mean ± SE).

T #	Diets	Phytase (FTU/kg)	Body weight (kg)	Feed			Revenue (PT)		Economic Efficiency	Relative economic efficiency
				Intake (kg)	Price/kg[1] (PT)	Cost (PT)	Total[2]	Net		
1	Control	0	1.624	3.165	73.68	233.20	730.80	497.60	2.134	100.00
2	As T1 +	1000	1.729	3.362	74.98	252.08	778.08	525.97	2.086	97.78
3	Low CP (20/18%)	0	1.446	3.051	68.45	208.83	650.70	441.87	2.116	99.16
4	As T3 +	1000	1.509	3.075	69.75	214.47	679.05	464.58	2.166	101.52
5	Low CP-IP (0.30/0.25%)	0	1.448	3.042	68.36	207.95	651.60	443.65	2.133	99.98
6	As T5 +	1000	1.558	3.015	69.66	210.03	701.10	491.08	2.338	109.58
7	Low CP-IP-Ca (0.67/0.60%)	0	1.555	3.146	68.22	214.60	699.75	485.15	2.261	105.94
8	As T7 +	1000	1.679	3.219	69.50	223.72	755.55	531.83	2.377	111.41

[1]Price per kg starter and grower diet + phytase.
[2] Price /kg of live body weight according to local market at the end of experimental time = 450 PT.

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تحسين الإستفادة من فيتات الفوسفور بإضافة إنزيم الفيتيز الميكروبى إلى علائق كتاكيت اللحم المنخفضة فى البروتين والفوسفور والكالسيوم والمكونة من الذرة وفول الصويا

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قسم الإنتاج الحيواني- كلية الزراعة- جامعة قناة السويس- ٤١٥٢٢ – الإسماعيلية - مصر

تم إستخدام ٢٨٨ كتوت لحم غير مجنس عمر يوم من سلالة أربورايكرز وذلك لتقييم تأثير إضافة إنزيم الفيتيز الميكروبي إلى علائق الذرة وكسب فول الصويا المنخفضة في البروتين الخام والفوسفور غير العضوي والكالسيوم على الإستفادة من فيتات الفوسفور. تم تصميم ثماني معاملات أحتوت كل معاملة على ثلاثة مكررات (١٢ طائر/ مكرر). تكونت العلائق من عليقة مقارنة موجبة وثلاث علائق قاعدية. إحتوت عليقة المقارنة الموجبة على ٢٠/٢٢% بروتين خام و ٠,٣٩/٠,٤٥% فوسفور غير عضوي و ٠,٩٠/١,٠٠% كالسيوم وذلك لكلا من علائق البادئ / والنامي..إحتوت العلائق القاعدية ١ و ٢ و ٣ على ١٨/٢٠% بروتين منخفض وأحتوت العليقتين القاعديتين ٢ و ٣ على ٠,٢٥/٠,٣٠% فوسفور غير عضوي منخفض وإحتوت العليقة القاعدية ٣ على ٠,٦٠/٠,٦٧% كالسيوم منخفض وذلك لكلا من علائق البادئ / النامي. إستخدم إنزيم الفيتيز الميكروبي في العليقة المقارنة والثلاثة علائق القاعدية بمستويين صفر و ١٠٠٠ وحدة/كجم من العليقة. تم تقييم أداء نمو الطيور وقياسات تكلس العظم وخواص الذبيحة والدم والكفاءة الاقتصادية.

أظهرت النتائج العامة لفترة ١-٦ أسابيع أن إضافة إنزيم الفيتيز لعليقة الطيور المقارنة حسنت من أداء النمو. إنخفضت الزيادة فى وزن الجسم المكتسب والمأكول اليومى من الغذاء وكذلك ساءت نسبة الكفاءة التحويلية للغذاء معنوياً عندما إنخفض البروتين الخام (٢٠/١٨%) بمفرده أو مع إنخفاض الفوسفور غير العضوي (٠,٣٠/٠,٢٥%) فى العلائق. أدت إضافة إنزيم الفيتيز (١٠٠٠ وحدة/ كجم من العليقة) للعلائق المنخفضة فى البروتين الخام فقط او المنخفضة فى البروتين الخام مع الفوسفور غير العضوي إلى تحسن فى الزيادة فى وزن الجسم المكتسب وكذلك تحسنت نسبة الكفاءة التحويلية للغذاء معنويا وكانت القيم المتحصل عليها متقاربة مع القيم المتحصل عليها لطيور المقارنة. أدى خفض الكالسيوم (٠,٦٧/٠,٦٠%) في عليقة الطيور المنخفضة في كلا من البروتين الخام والفوسفور غير العضوي إلى إنخفاض الزيادة في وزن الجسم المكتسب وتدهور في نسبة الكفاءة التحويلية للغذاء معنوياً وذلك بالمقارنة بالطيور المقارنة. أدت إضافة إنزيم الفيتيز إلى العليقة المنخفضة في البروتين الخام والفوسفور غير العضوي والكالسيوم إلى إسترجاع الزيادة فى وزن الجسم المكتسب وتحسن في نسبة الكفاءة التحويلية للغذاء معنويا وكانت القيم المتحصل عليها أفضل من قيم الطيور المقارنة.

أدت إضافة إنزيم الفيتيز إلى كل العلائق إلى زيادة معنوية فى رماد عظمة الساق مع عدم وجود إختلافات معنوية بين العليقة المقارنة والثلاث علائق القاعدية المضاف إليها إنزيم الفيتيز. أدت إضافة إنزيم الفيتيز إلى العليقة المنخفضة فى البروتين الخام والفوسفور غير العضوى والكالسيوم إلى إنخفاض معنوى فى كالسيوم بلازما الدم وذلك بالمقارنة مع باقي المعاملات. أدت إضافة إنزيم الفيتيز إلى العلائق إلى زيادة رقمية فى فوسفور بلازما الدم مع وجود إختلافات معنوية فقط عند إضافة الإنزيم إلى العليقة المنخفضة في البروتين الخام والفوسفور غير العضوي. أدت إضافة إنزيم الفيتيز إلى العليقة المنخفضة فى البروتين الخام إلى زيادة معنوية فى بروتين بلازما الدم. فيما عدا دهن البطن لم يكن للمعاملات تأثيرمعنوى على صفات الذبيحة. فبينما زاد دهن البطن معنوياً نتيجة لإضافة إنزيم الفيتيز إلى العليقة المقارنة إنخفض دهن البطن معنويا بإضافة إنزيم الفيتيز إلى العليقة المنخفضة في البروتين الخام والفوسفور غير العضوي. أدت إضافة إنزيم الفيتيز إلى العلائق المنخفضة في البروتين الخام ، المنخفضة فى البروتين الخام والفوسفور غير العضوي ، المنخفضة في البروتين الخام والفوسفور غير العضوي والكالسيوم إلى زيادة الكفاءة الاقتصادية النسبية وتفوقت هذة العلائق على العلائق التي لم يضاف لها إنزيم الفيتيز وكذلك على عليقة المقارنة المضاف إليها الإنزيم.

ويستخلص من ذلك أن إضافة إنزيم الفيتيز إلى علائق كتاكيت اللحم المنخفضة في كل من البروتين الخام والفسفور غير العضوي والكالسيوم حسنت معنوياً من الزيادة في وزن الجسم المكتسب ونسبة الكفاءة التحويلية للغذاء ونسبة رماد عظمة الساق وكذلك أعطت أعلى كفاءة اقتصادية نسبية.