The Effects of Microbial Phytase on Serum Calcium and Phosphorus Levels and Alkaline Phosphatase Activities in Broilers Fed Diets Containing Different Levels of Phosphorus

Tanay Bilal¹, Selcuk Atis² & Onur Keser¹

ABSTRACT

Background: Addition of the phytase enzyme to the diet of monogastric animals is an effective and practical method for both utilization of phytate-P and P disposal. Besides hydrolytic effects on phytate, phytase may also improve the availability of minerals. Alkaline phosphatase (ALP) plays an important role in bone mineralization. Since dietary P levels decrease, serum ALP level has been shown to increase. It was reported that pigs and broilers fed phytase had lower serum ALP level. The present study aimed to investigate the effect of microbial phytase on serum calcium (Ca), inorganic phosphorus (Pi) levels and ALP activities in broilers fed diets containing different P levels.

Materials, Methods & Results: A total of 144 one-day-old male broiler chicks (Cobb 500 stain) were used in this study. The birds were housed in metabolic cages (33 cm × 33 cm × 40 cm) under environmentally controlled conditions for 42 days. The fluorescent lights were on for 24 h each day. Diets and water were provided ad libitum. Starter diet was given in the first three weeks of the study, and grower diet was given from the beginning of the fourth to the end of the sixth week. The study was designed as a 3 × 2 factorial arrangement with three levels, low, medium and high, of nonphytate phosphorus (nPP) (0.18, 0.30 and 0.45 %), and two levels of phytase (0 and 600 U/kg diet). Blood samples were collected from 5 broilers in each group on days 21 and 42 and analyzed for serum Ca and Pi levels and ALP activities. Serum Ca concentrations were affected only by dietary P levels between 0-21 days (P < 0.05), but there were no differences between groups for serum Ca concentrations for 22-42 days. Phytase supplementation had no effect on serum Ca concentrations. During the experiment, serum P concentrations increased linearly depending on dietary P levels. Phytase supplemented groups fed low (0.18%) and medium (0.30%) P diets had higher serum P concentrations than those of unsupplemented groups (P < 0.05). However, serum P concentrations were not affected by phytase supplementation in group fed high (0.45%) P diet. In unsupplemented groups, dietary P levels and ALP activity were inversely proportional and serum ALP activities were significantly decreased by the increase in the dietary P levels during the experiment (P < 0.05). When it was compared to unsupplemented groups, ALP activities were significantly lower in phytase supplemented groups (P < 0.05). In phytase supplemented groups, there were no difference for ALP activities between groups fed low and medium P diet, but group fed high P diet had significantly lower ALP activities than other supplemented groups (P < 0.05).

Discussion: Serum P concentrations were affected by dietary P levels and increased linearly depending on dietary P levels. Although phytase supplementation significantly increased serum P concentrations during the study, serum Ca levels were affected only in first three weeks of the experiment. ALP is a metalloenzyme that plays an important role in bone mineralization. As expected, dietary P levels and serum ALP activity were inversely proportional in this study. The decrease in serum ALP activity in groups supplemented phytase was a response to increasing P levels in diets in supplemented phytase groups and this result can be accepted as an indicative of the increase in phytate-P utilization in broilers fed low-P diet. Similar results have already been observed in prior studies with chickens and turkeys.

Keywords: microbial phytase, calcium, phosphorus, alkaline phosphatase, broiler
INTRODUCTION

In raw materials of plant origin commonly used in poultry diets such as soybean and corn, 60-80% of the P is in the form of phytate phosphorus, an organically bound form of phosphorus [16]. When it was compared to other sources of phosphorus, the digestion and utilization of phytate phosphorus is very low, therefore, the addition of inorganic phosphorus to diets for monogastric animals increases the cost of the diet [9]. Phytase is an enzyme which hydrolyzes phytate to inositol and inorganic phosphate. Addition of the phytase enzyme to the diet of monogastric animals is an effective and practical method for both utilization of phytate-P and P disposal [21]. Studies with broiler chickens fed corn-soybean diets indicate phytate-P utilization of between 10 and 53% [2,6]. It was reported that addition of phytase increased dietary P availability to 65% and reduced P excretion by 50% in broilers [19]. Similarly, phytase supplemented diets reduced the need to P and decreased P excretion [3,8].

Alkaline phosphatase (ALP) plays an important role in bone mineralization. Since dietary P levels decrease, serum ALP level has been shown to increase [10]. It was reported that pigs and broilers fed phytase had lower serum ALP level [8,11].

The aim of this study was to investigate the effect of microbial phytase on some serum indices (Ca, Pi and ALP activities) in broilers fed diets containing different P levels.

MATERIALS AND METHODS

Animals and Diets

A total of 144 one-day-old male broiler chicks (Cobb 500 stain) were used in this study. The birds were housed in metabolic cages (33 cm × 33 cm × 40 cm) under environmentally controlled conditions for 42 days. The fluorescent lights were on for 24 h each day. Diets and water were provided ad libitum. A basal diet containing maize and soybean meal was prepared in accordance with the requirements of broilers [14]. Starter diet was given in the first three weeks of the study, and grower diet was given from the beginning of the fourth to the end of the sixth week. Composition and calculated nutrients in diets were presented in Table 1. The study was designed as a 3 × 2 factorial arrangement with three levels, low, medium and high, of nonphytate phosphorus (nPP) (0.18, 0.30 and 0.45 %), and two levels (0 and 600 U/kg diet) of microbial phytase (Natuphos® 600 from Aspergillus niger).

Collection of Samples and Analysis

Blood samples were collected from 5 broilers in each group on day 21 and 42. Samples were stored at -20 °C for further analysis. Serum Ca and Pi levels and ALP activities were analysed by using commercial kits in Technicon RA-1000 Autoanlyser.

Statistical Analysis

All data were compared by using analysis of variance (ANOVA, Duncan’s multiple-range test) between groups. Results are presented as mean ± standard error. All statistical analysis was performed by using software package program. A significant level of P < 0.05 was employed in the analysis of data from groups.

RESULTS

The effects of microbial phytase on serum Ca and Pi levels and ALP activities in broilers fed diets containing different phosphorus levels were presented in Table 2. Serum Ca concentrations were affected only by dietary P levels between 0-21 days (P < 0.05), but there were no differences between groups for serum Ca concentrations for 22-42 days. Phytase supplementation had no effect on serum Ca concentrations. During the experiment, serum P concentrations increased linearly depending on dietary P levels. Phytase supplemented groups fed low (0.18%) and medium (0.30%) P diets had higher serum P concentrations than those of unsupplemented groups (P < 0.05). However, serum P concentrations were not affected by phytase supplementation in group fed high (0.45%) P diet.

In unsupplemented groups, dietary P levels and ALP activity were inversely proportional and serum ALP activities were significantly decreased by the increase in the dietary P levels during the experiment (P < 0.05). When it was compared to unsupplemented groups, ALP activities were significantly lower in phytase supplemented groups (P < 0.05). In phytase supplemented groups, there were no difference for ALP activities between groups fed low and medium P diet, but group fed high P diet had significantly lower ALP activities than other supplemented groups (P < 0.05).
Table 1. Diet ingredients and nutrient compositions of the starter (d 0-21) and grower (d 21-42) diets containing low, medium and high level of nonphytate phosphorus (nPP).

<table>
<thead>
<tr>
<th>Ingredients (%)</th>
<th>Starter (d 0-21)</th>
<th>Grower (d 22-42)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Low nPP</td>
<td>Midium nPP</td>
</tr>
<tr>
<td>Maize (%)</td>
<td>50.95</td>
<td>49.08</td>
</tr>
<tr>
<td>Soybean meal (%)</td>
<td>35.00</td>
<td>35.00</td>
</tr>
<tr>
<td>Maize gluten (%)</td>
<td>4.00</td>
<td>2.50</td>
</tr>
<tr>
<td>Fullfat soybean (%)</td>
<td>2.42</td>
<td>5.36</td>
</tr>
<tr>
<td>Sunflower oil (%)</td>
<td>3.43</td>
<td>3.62</td>
</tr>
<tr>
<td>Limestone (%)</td>
<td>2.10</td>
<td>1.64</td>
</tr>
<tr>
<td>Dicalcium phosphate (%)</td>
<td>0.42</td>
<td>1.12</td>
</tr>
<tr>
<td>Salt (%)</td>
<td>0.34</td>
<td>0.34</td>
</tr>
<tr>
<td>Lysine (%)</td>
<td>0.36</td>
<td>0.34</td>
</tr>
<tr>
<td>DL-methionine (%)</td>
<td>0.18</td>
<td>0.20</td>
</tr>
<tr>
<td>Vitamin C (%)</td>
<td>0.10</td>
<td>0.10</td>
</tr>
<tr>
<td>Premix* (%)</td>
<td>0.50</td>
<td>0.50</td>
</tr>
<tr>
<td>Baciferm (%)</td>
<td>0.10</td>
<td>0.10</td>
</tr>
<tr>
<td>Anticoccidial (%)</td>
<td>0.10</td>
<td>0.10</td>
</tr>
</tbody>
</table>

Disruption

Phytase supplementation had no effect on serum Ca concentrations. Similarly, it was reported that phytase supplementation had no significant effect on plasma Ca at the low or recommended dietary Ca levels [17]. Also, in a study on weanling horses, plasma Ca concentrations were not affected by any of the diet-phytase combination [12].

Serum P concentrations were affected by dietary P levels and increased linearly depending on dietary P levels. When it was compared to unsupplemented groups, phytase supplementation significantly increased serum P concentrations during the experiment. Similarly, it was reported that phytase supplementation to low-P diets increased P concentrations in plasma [4]. Improved plasma P content was also
observed when phytase was added to the diets [15,18]. However, phytase supplementation to low-P diets increased plasma P level by 8% in broilers fed different levels of phosphorus [20], and these findings were agree with the results of the present study.

Serum ALP activities were significantly decreased by the increase in the dietary P levels. The phytase supplementation decreased serum ALP activity and this decrease was greater in lowest P diet ($P < 0.05$). ALP is a metalloenzyme that play an important role during mineralization [13]. The decrease observed in this study in serum ALP activity was a response to increasing P levels in diets in supplemented phytase groups. Similar results have already been observed in chickens [7] and turkeys [1]. The decreased serum ALP activities in phytase supplemented groups in the present study were also agreed with the results found in broilers [5,21]. Similar results have been also reported in a study carried out to investigate the effect of dietary phytase and high available phosphorus corn on broiler chicken performance [8]. However it was observed that serum ALP levels decreased in chicks that received high levels of phosphorus or phytase, most likely the result of a downregulation of ALP activity, suggesting that the synthesis of this protein is dependent on phosphorus levels [8].

CONCLUSIONS

In conclusion, phytase supplementation improved phosphorus utilization and increased serum P concentration in broilers fed low-P diets. However, phytase can play a role in modifying serum ALP activity by decreasing it in broilers fed low-P diet.

MANUFACTURERS
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Ethical approval. This study was approved by Istanbul University Veterinary Faculty Ethical Committee (Protocol No:2009/128)

Declaration of interest. The authors declare no conflict of interests. The authors alone are responsible for the content and writing of the paper.

REFERENCES


