INTRODUCTION

Although the energy content of wheat is lower than that for corn, the contents of many other nutrients (e.g., protein, Ca, P, and some amino acids, such as Lys, Met, Arg, Trp, and Thr) are greater in wheat (NRC, 1994). However, wheat has antinutritional factors, such as soluble nonstarch polysaccharides (NSP). Because of inadequate amounts of, or the absence of, enzyme secretion to hydrolyze these components in the gastrointestinal tract (GIT), they negatively affect the nutrient utilization and performance of monogastric animals and birds. Efforts to ameliorate the negative effects of the antinutritional components of wheat by dietary supplementation with microbial enzymes have been successful. Many studies have shown the effects of NSP-degrading enzymes on hydrolyzing NSP, reducing viscosity, and improving nutrient utilization and bird performance (Choct and Annison, 1992; Ravindran et al., 1999; Wang et al., 2005; Boguhn and Rodehutscord, 2010).

Approximately 60 to 80% of the P in plant feed ingredients is in the form of phytate and is therefore unavailable to poultry (Ravindran et al., 1995). It has thus been suggested that xylanase supplementation may improve P utilization in wheat-based diets (Kim et al., 2005). Xylanase may facilitate enzymatic phy-

Effects of xylanase and citric acid on the performance, nutrient retention, and characteristics of gastrointestinal tract of broilers fed low-phosphorus wheat-based diets

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ABSTRACT An experiment was conducted to study the effects of xylanase and citric acid on the performance, nutrient retention, jejunal viscosity, and size and pH of the gastrointestinal tract of broilers fed a low-P wheat-based diet. The experiment was conducted as a 2 × 3 factorial arrangement with 2 levels of xylanase (0 and 200 mg/kg) and 3 levels of citric acid (0, 20, and 40 g/kg). Each of the 6 dietary treatments was fed to 4 replicate pens (17 birds/pen) from 0 to 24 d of age. Chromium oxide (3 g/kg) was added to the diets as an indigestible marker to determine the apparent nutrient retention. No interaction effect was observed between xylanase and citric acid in any measured response. Xylanase did not affect feed intake but significantly increased BW gain by 3.6% (P < 0.05) from 1 to 24 d of age and improved G:F by 3.9% (P < 0.01). The inclusion of 40 g/kg of citric acid decreased (P < 0.01) BW gain and feed intake by 8.6 and 12.5%, respectively. Xylanase significantly decreased the viscosity of digesta and improved the retention of DM, CP, and energy, but did not have a significant effect on the retention of fat and P. Inclusion of 20 and 40 g/kg of citric acid in the diets increased P retention by 15.8 and 16.3% (P < 0.01), respectively. Citric acid significantly decreased the pH of crop contents (P < 0.05). In conclusion, citric acid, at the 40 g/kg inclusion level, reduced feed intake and BW gain but improved G:F and P retention. Xylanase decreased digesta viscosity, increased nutrient retention, and consequently improved the performance of broilers fed the low-P wheat-based diet. Thus, adding 20 g/kg of citric acid, especially in the starter period, and 200 mg/kg of xylanase to low-P wheat-based diets can be helpful.

Key words: broiler performance, citric acid, retention, wheat, xylanase

INTRODUCTION

Although the energy content of wheat is lower than that for corn, the contents of many other nutrients (e.g., protein, Ca, P, and some amino acids, such as Lys, Met, Arg, Trp, and Thr) are greater in wheat (NRC, 1994). However, wheat has antinutritional factors, such as soluble nonstarch polysaccharides (NSP). Because of inadequate amounts of, or the absence of, enzyme secretion to hydrolyze these components in the gastrointestinal tract (GIT), they negatively affect the nutrient utilization and performance of monogastric animals and birds. Efforts to ameliorate the negative effects of the antinutritional components of wheat by dietary supplementation with microbial enzymes have been successful. Many studies have shown the effects of NSP-degrading enzymes on hydrolyzing NSP, reducing viscosity, and improving nutrient utilization and bird performance (Choct and Annison, 1992; Ravindran et al., 1999; Wang et al., 2005; Boguhn and Rodehutscord, 2010).

The inclusion of 20 and 40 g/kg of citric acid improved G:F by 2.3 and 4.5% (P < 0.05), respectively. Xylanase significantly decreased the viscosity of digesta and improved the retention of DM, CP, and energy, but did not have a significant effect on the retention of fat and P. Inclusion of 20 and 40 g/kg of citric acid in the diets increased P retention by 15.8 and 16.3% (P < 0.01), respectively. Citric acid significantly decreased the pH of crop contents (P < 0.05). In conclusion, citric acid, at the 40 g/kg inclusion level, reduced feed intake and BW gain but improved G:F and P retention. Xylanase decreased digesta viscosity, increased nutrient retention, and consequently improved the performance of broilers fed the low-P wheat-based diet. Thus, adding 20 g/kg of citric acid, especially in the starter period, and 200 mg/kg of xylanase to low-P wheat-based diets can be helpful.

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tase activity by increasing cell wall permeability in the aleurone layer of wheat (Selle et al., 2009) or by liberating the phytate bound to P, CP, and starch (Kim et al., 2005), or both.

It has also been reported that citric acid can increase in vitro phytate dephosphorylation (Zyla et al., 1995). Boling-Frankenbach et al. (2001) evaluated the effects of adding 40 and 60 g/kg of citric acid on phytate P utilization in broilers fed P-deficient (0.20% available P) corn and soybean meal diets and reported that citric acid increased P utilization and decreased the available P requirement by approximately 0.1% of the diet. Moreover, some authors have reported positive effects of xylanase and citric acid on nutrient retention (Ao et al., 2009; Boguhn and Rodehutscord, 2010), positive effects of xylanase on viscosity and GIT size (Engberg et al., 2004; Wang et al., 2005), and a decreasing effect of citric acid on digesta pH (Ao et al., 2009). Much information is available on the effect of citric acid on the performance and P utilization of broilers fed corn-based diets (Boling et al., 2000; Biggs and Parsons, 2008; Woyengo et al., 2010). However, much less information is available on the effect of citric acid in broilers fed wheat-based diets, especially with low-P diets that include xylanase enzyme.

Thus, the objective of this study was to evaluate the effects of xylanase enzyme and citric acid, individually and in combination, on the performance, nutrient retention, and characteristics of the GIT in low-P broiler diets based on wheat.

MATERIALS AND METHODS

Birds and Diets

A total of 408 male, 1-d-old Ross 308 broilers were obtained from a commercial hatchery and randomly distributed into 24 floor pens in an environmentally controlled house. Each of the 6 treatments was replicated 4 times, with 17 birds per pen (2.5 × 1.3 m). The experimental period was divided into 2 phases: a starter phase (0 to 10 d) and a grower phase (11 to 24 d). The ingredient composition and nutrient content of the basal diets for both experimental phases are presented in Table 1. Basal diets were formulated to meet or exceed all nutrient recommendations published in the Ross rearing guideline (Aviagen, 2007), except for available P, which was 0.33 and 0.27% for the starter and grower diets, respectively (0.15% less than recommendations). The experiment was conducted as a 2 × 2 factorial arrangement with 2 levels (0 and 200 mg/kg) of xylanase enzyme (Novozymes A/S, Bagavaerd, Denmark), consisting of 1,000 fungal xylanase units/g of enzyme activity, and 3 levels (0, 20, and 40 g/kg) of citric acid (99% purity; Kimia Gharb Gostar, Kermanshah, Iran). The levels of xylanase and citric acid were chosen according to those used by Choct et al. (2004) and Boling-Frankenbach et al. (2001), respectively. To keep the diets isocaloric, citric acid was added to the basal diets in place of corn starch and sand flour. All experimental diets were free of antibiotics and were provided in mash form. Experimental diets were offered to the birds from d 1 of age. Birds had free access to feed and water. Lighting was continuous, and the temperature was maintained at 33°C from d 0 to 3 and was then gradually decreased to 24°C by the end of the third week.

Growth Trial Procedures

The experimental period lasted 24 d. On d 10 and 24, birds were weighed by pen, and feed consumption was recorded. Feed efficiency and ADG were calculated for each phase. The birds were inspected daily, and the BW of dead birds and date of death were recorded. When calculating feed efficiency, the BW of the dead birds was taken into consideration.

Metabolism Trial

For the determination of total tract nutrient retention (TTNR), 2 birds per replicate with BW similar to the average of the corresponding pen were selected and transferred to battery cages (2 birds in each) with a wire mesh bottom and excreta collection trays (60 × 30 × 30 cm, length × width × height) at 19 d of age. Each cage was equipped with feed and water cups placed outside the cage. Birds in each cage were offered the corresponding diet. The experimental diets were the same as in the growth experiment, except that Cr2O3 was added at 3 g/kg to all diets as an indigestible marker. The metabolism trial was composed of a 3-d preliminary adaptation period, followed by 2 d of excreta collection. Contamination (e.g., feathers and down) was carefully removed and the excreta collected per cage during the 2-d collection period was pooled as 1 replicate. Excreta were dried at 60°C for 48 h, finely ground, and stored in airtight plastic containers until analysis.

Sampling Procedures for Viscosity, pH, and the Size of Different Organs

At 24 d of age, 2 birds/replicate were randomly selected and euthanized using sodium thiopental, the digestive organs were excised, and jejunal digesta samples were taken for viscosity measurement. One gram of the crop contents and 1 g of digesta from the gizzard, duodenum, jejunum, ileum, and left cecum of each bird were immediately collected and placed into clean Falcon tubes. The samples were mixed with deionized water (1:10 wt/vol), and the pH of the solution was measured with a digital pH meter (Model 827, Metrohm, Herisau, Switzerland) at room temperature.

The jejunal digesta was kept on ice before centrifugation. The digesta samples of the 2 birds/pen were pooled and mixed to achieve a homogenous mixture,
which was then centrifuged at 2,150 × g at 4°C for 15 min. The supernatant (0.5 mL) from each tube was analyzed for viscosity. The viscosity of the supernatant was measured with a digital viscometer (Model DV-III, Brookfield Engineering Laboratories Inc., Middleboro, MA). The results of the viscosity measurement are stated in centipoises (cPs).

The weights of the pancreas and liver were recorded, and the lengths of the duodenum (from the ventriculus to the pancreo-biliary duct), jejunum (from the pancreo-biliary duct to Meckel's diverticulum), and ileum (from Meckel's diverticulum to the ileocecal junction) were measured with a precision of 1 mm, at 24 d of age. The weights of the pancreas and liver, and the lengths of the duodenum, jejunum, and ileum were expressed as a proportion of 100 g of BW.

### Chemical Analysis

Feeds and excreta samples were analyzed for content of DM (method 942.05; AOAC, 2000), fat [method 920.32 (AOAC, 2000) by a 1043 Soxtec HT system, Foss Tecator AB, Höganas, Sweden] and P [method 965.17 (AOAC, 1990) by a Model 2100 UV spectrophotometer, Shimadzu, Kyoto, Japan]. The total N was measured by the Kjeldahl method (Kjeltc 1030 Autoanalyzer, Foss Tecator AB), and CP was calculated as N × 6.25. Gross energy of diet and excreta samples was determined using an adiabatic bomb calorimeter (IKA-Kalorimeter, Model C400, IKA, Staufen, Germany), standardized with benzoic acid. Chromium oxide content in the experimental diets and excreta was measured according to Saha and Gilbreath (1991).

### Calculations and Statistical Analysis

In the metabolic trial, the following equation was used to calculate TTNR (Scott et al., 1976):

\[
TTNR (%) = 100 - \left( \frac{\text{diet Cr}_2\text{O}_3/\text{excreta Cr}_2\text{O}_3}{\text{nutrient in excreta/nutrient in diet}} \right) \times 100.
\]

The experiment was carried out as a completely randomized design with treatments arranged factorially. Data were analyzed as a 3 × 2 factorial arrangement (3 levels of citric acid and 2 levels of xylanase) using PROC GLM of SAS (SAS Institute, 2001). Performance data were analyzed considering the pen of birds as an experimental unit. Linear and quadratic effects were also analyzed for citric acid levels.

### RESULTS

#### Growth Performance

At 10 d of age, dietary supplementation with xylanase did not affect growth performance, but inclusion of 20 g/kg of citric acid increased (quadratic, \( P < 0.01 \))
ADFI by 5.0% and ADG by 3.7% compared with birds fed the control diet (Table 2). The inclusion of 40 g/kg of citric acid lowered feed intake, but the highest feed efficiency was observed in broilers offered 40 g/kg of citric acid (5.6% more than the control; \( P < 0.05 \)). From 11 to 24 d of age, xylanase supplementation significantly increased ADG (\( P < 0.01 \)) and G:F (\( P < 0.05 \)) by 4.5 and 4.2%, respectively. Citric acid had significant effects on ADG and ADFI; citric acid linearly decreased ADG and ADFI, with the lowest ADG and ADFI being obtained with 40 g/kg of citric acid. At 24 d of age, xylanase supplementation did not affect feed intake; however, it significantly increased ADG by 3.6% (\( P < 0.05 \)) and improved G:F by 3.9% (\( P < 0.01 \)) when compared with the nonsupplemented group. Inclusion of citric acid significantly affected broiler performance; the inclusion of 40 g/kg of citric acid decreased (\( P < 0.01 \)) ADG and ADFI by 8.6 and 12.5%, respectively, compared with the control diet. Citric acid linearly decreased ADFI and ADG but increased G:F. There was no interaction effect between xylanase and citric acid in any measured performance responses.

**Nutrient Retention at 24 d**

Xylanase supplementation significantly affected the retention of DM, energy, and CP (Table 3). In the xylanase-supplemented group, the retention of DM, energy, and CP was significantly increased by 11, 10, and 15% (\( P < 0.01 \)), respectively, compared with the control group. Xylanase did not affect fat and P retention. Citric acid did not have a significant effect on the retention of DM, energy, and CP, except for P. Inclusion of 20 and 40 g/kg of citric acid increased P retention by 15.8 and 16.3% (\( P < 0.01 \)) compared with the control group. No interaction effect between xylanase and citric acid was observed in any of measured retention responses.

**GIT pH and Digesta Viscosity at 24 d**

Xylanase did not affect pH values in any parts of the GIT of broilers (Table 4). Citric acid significantly affected the pH of crop contents. Inclusion of 20 and 40 g/kg of citric acid linearly decreased the pH value of crop contents by 0.15 and 0.31 units (\( P < 0.01 \)), respectively, compared with values for the control group. Inclusion of citric acid did not affect the pH values of the gizzard, duodenum, jejunum, and ileum, or the viscosity of jejunal digesta. Xylanase supplementation significantly affected the viscosity of jejunal digesta. Xylanase decreased the viscosity of jejunal digesta by 0.73 units (\( P < 0.01 \)) compared with values in the nonsupplemented group. No interaction effect between xylanase and citric acid was observed in GIT pH and digesta viscosity.

**Digestive Tract Measurements at 24 d**

The effects of xylanase and citric acid on the relative lengths (cm/100 g of BW) of different parts of the small intestine and relative weights (g/100 g of BW) of the liver and pancreas are presented in Table 5. Xylanase and citric acid did not have a significant effect on the relative lengths of different parts of the small intestine and relative weights of the liver and pancreas. In addition, no interaction effect was observed between xylanase and citric acid.

**DISCUSSION**

The results of this study showed that xylanase supplementation improved the performance of broilers at 24 d of age. Arabinoxylans, the major component of NSP in wheat-based diets, increase the digesta viscosity of

### Table 2. Effects of xylanase and citric acid supplementation on the performance of broilers

<table>
<thead>
<tr>
<th>Treatment</th>
<th>1 to 10 d</th>
<th>11 to 24 d</th>
<th>1 to 24 d</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>ADG (g)</td>
<td>ADFI (g)</td>
<td>G:F (g/kg)</td>
</tr>
<tr>
<td>Citric acid (g/kg)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>23.64</td>
<td>28.06</td>
<td>845</td>
</tr>
<tr>
<td>20</td>
<td>24.51</td>
<td>29.47</td>
<td>833</td>
</tr>
<tr>
<td>40</td>
<td>23.63</td>
<td>26.50</td>
<td>892</td>
</tr>
<tr>
<td>SEM</td>
<td>0.26</td>
<td>0.45</td>
<td>16</td>
</tr>
<tr>
<td>Xylanase (mg/kg)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>23.88</td>
<td>28.31</td>
<td>846</td>
</tr>
<tr>
<td>200</td>
<td>23.98</td>
<td>27.71</td>
<td>868</td>
</tr>
<tr>
<td>SEM</td>
<td>0.21</td>
<td>0.37</td>
<td>13</td>
</tr>
</tbody>
</table>

\[ P-value \]

| Xylanase | 0.751     | 0.272     | 0.253     | 0.001    | 0.891      | 0.019     | 0.022    | 0.879      | 0.008     |
| Citric acid | 0.039     | 0.001     | 0.043     | 0.027    | 0.001      | 0.186     | 0.001    | 0.001      | 0.031     |
| Linear    | 0.990     | 0.026     | 0.052     | 0.001    | 0.001      | 0.084     | 0.001    | 0.001      | 0.009     |
| Quadratic | 0.011     | 0.001     | 0.090     | 0.118    | 0.281      | 0.550     | 0.047    | 0.038      | 0.984     |

\[ ^1 \text{Means represent 4 pens of 17 chicks each per treatment. There was no interaction effect.} \]
### Table 3. Effects of xylanase and citric acid supplementation on the apparent retention (%) of DM, CP, fat, energy, and P of broilers at 24 d of age

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Retention</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>DM (g/kg)</td>
<td>CP</td>
<td>Fat (g/kg)</td>
<td>Energy</td>
</tr>
<tr>
<td>Citric acid (g/kg)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>64.67</td>
<td>56.07</td>
<td>79.70</td>
<td>70.93</td>
</tr>
<tr>
<td>20</td>
<td>65.23</td>
<td>56.00</td>
<td>81.11</td>
<td>70.55</td>
</tr>
<tr>
<td>40</td>
<td>67.58</td>
<td>57.74</td>
<td>82.65</td>
<td>71.26</td>
</tr>
<tr>
<td>SEM</td>
<td>0.98</td>
<td>1.48</td>
<td>1.44</td>
<td>0.82</td>
</tr>
<tr>
<td>Xylanase (mg/kg)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>62.37</td>
<td>52.55</td>
<td>80.23</td>
<td>67.65</td>
</tr>
<tr>
<td>200</td>
<td>69.27</td>
<td>60.65</td>
<td>82.07</td>
<td>74.17</td>
</tr>
<tr>
<td>SEM</td>
<td>0.80</td>
<td>1.21</td>
<td>1.17</td>
<td>0.67</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>P-value</th>
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<th></th>
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</thead>
<tbody>
<tr>
<td>Xylanase</td>
<td>0.001</td>
<td>0.001</td>
<td>0.283</td>
<td>0.001</td>
</tr>
<tr>
<td>Citric acid</td>
<td>0.112</td>
<td>0.651</td>
<td>0.367</td>
<td>0.828</td>
</tr>
<tr>
<td>Linear</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td>Quadratic</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
</tr>
</tbody>
</table>

1Means represent 8 birds per treatment. There was no interaction effect.

### Table 4. Effects of xylanase and citric acid supplementation on pH of different parts of the gastrointestinal tract and viscosity of jejunal digesta at 24 d of age

<table>
<thead>
<tr>
<th>Treatment</th>
<th>pH</th>
<th>Viscosity (cPs)</th>
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<tbody>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Citric acid (g/kg)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>4.88</td>
<td>2.90</td>
</tr>
<tr>
<td>20</td>
<td>4.73</td>
<td>2.92</td>
</tr>
<tr>
<td>40</td>
<td>4.57</td>
<td>3.02</td>
</tr>
<tr>
<td>SEM</td>
<td>0.07</td>
<td>0.10</td>
</tr>
<tr>
<td>Xylanase (mg/kg)</td>
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<td></td>
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<tr>
<td>0</td>
<td>4.76</td>
<td>3.04</td>
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<td>200</td>
<td>4.69</td>
<td>2.85</td>
</tr>
<tr>
<td>SEM</td>
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<td>0.08</td>
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<table>
<thead>
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<th>P-value</th>
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<tbody>
<tr>
<td>Xylanase</td>
<td>0.400</td>
<td>0.128</td>
<td>0.095</td>
<td>0.642</td>
</tr>
<tr>
<td>Citric acid</td>
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<td>0.691</td>
<td>0.331</td>
<td>0.323</td>
</tr>
<tr>
<td>Linear</td>
<td>0.008</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td>Quadratic</td>
<td>0.967</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
</tr>
</tbody>
</table>

1Means represent 4 birds per treatment; for viscosity, means represent 8 birds per treatment. There was no interaction effect.

### Table 5. Effects of xylanase and citric acid supplementation on the relative lengths of the duodenum, jejunum, and ileum (cm/100 g of BW) and relative weights of the liver and pancreas (g/100 g of BW) at 24 d of age

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Relative length</th>
<th>Relative weight</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Duodenum</td>
<td>Jejunum</td>
</tr>
<tr>
<td>Citric acid (g/kg)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>2.92</td>
<td>7.58</td>
</tr>
<tr>
<td>20</td>
<td>2.89</td>
<td>7.19</td>
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<td>2.96</td>
<td>7.94</td>
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<tr>
<td>Xylanase (mg/kg)</td>
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</tr>
<tr>
<td>0</td>
<td>2.93</td>
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<tr>
<td>200</td>
<td>2.92</td>
<td>7.65</td>
</tr>
<tr>
<td>SEM</td>
<td>0.08</td>
<td>0.21</td>
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</table>

<table>
<thead>
<tr>
<th>P-value</th>
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<th></th>
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</thead>
<tbody>
<tr>
<td>Xylanase</td>
<td>0.949</td>
<td>0.602</td>
<td>0.721</td>
<td>0.948</td>
</tr>
<tr>
<td>Citric acid</td>
<td>0.866</td>
<td>0.158</td>
<td>0.455</td>
<td>0.274</td>
</tr>
</tbody>
</table>

1Means represent 4 birds per treatment. There was no interaction effect.
and energy. These results are in agreement with those of Woyengo et al. (2010). Other authors, for example, Brenes et al. (2003) and Rafacz-Livingston et al. (2005), have reported positive effects of citric acid on the retention of P or tibia ash in broilers fed corn- and soybean-based diets. Improved retention of P might be due to the chelating effects of citric acid on Ca, resulting in increasing phytate solubility and susceptibility to hydrolysis, subsequently increasing P availability (Centeno et al., 2007). Xylanase supplementation in our study improved retention of DM, energy, and CP. Positive effects of xylanase on nutrient retention have been reported by other researchers (Ravindran et al., 1999; Boguhn and Rodehutscord, 2010). The NSP in the cell walls of cereals encapsulate starch and protein; however, they also increase the viscosity of digesta. The improved CP retention with added xylanase in wheat-based diets may be partly due to lowering the endogenous amino acid losses, resulting from the elimination of adverse effects of wheat pentosans (Angkanaporn et al., 1994). Xylanase did not affect fat retention in our study, which is in agreement with the report by Juanpere et al. (2005). This may be partly due to the age of the birds and the type of fat (soybean oil) used in this study. In young chickens, fat digestion increases with age and reaches optimal capacity after 2 wk of age (Nitsan et al., 1991). On the other hand, it has been reported that xylanase supplementation improves fat retention only in diets that include animal fat, and that it has no effect on wheat- and rye-based diets that include soybean oil (Langhout et al., 1997). In the current study, fat retention was measured at 24 d of age, and soybean oil was used as the fat source in the experimental diets. Xylanase supplementation did not affect P retention, which is in agreement with the reports by Kim et al. (2008) and Selle et al. (2009).

One of the positive effects of organic acids on P availability in poultry production has been attributed to their effects on decreasing the GIT pH (Boling-Frankenbach et al., 2001). Therefore, the pH values of different parts of the GIT were measured. The pH values of the different segments of the GIT were in the range reported by Herpol (1966), except that crop pH values in the current study were lower. As expected, citric acid decreased the pH of crop contents, in agreement with the report by Ao et al. (2009). However, citric acid did not affect the pH of the gizzard, duodenum, jejunum, ileum, and cecum, in agreement with results reported by Brown and Southern (1985) and Smulikowska et al. (2010). It has been reported that little dietary organic acid reaches the lower parts of the digestive tract (Hume et al., 1993), so this may partly be the reason for the absence of an effect on intestinal pH. Xylanase did not affect digesta pH in any part of the GIT, in agreement with the results reported by Jozeffiak et al. (2007) and Rebolé et al. (2010). In contrast with our results, Gao et al. (2008) reported that xylanase supplementation increased the pH of the crop, duodenum, and jejunum, but Engberg et al. (2004) reported that xylanase de-
creased the pH of duodenal and cecal digesta in broilers fed wheat-based diets.

Neither xylanase nor citric acid supplementation affected the size of the GIT, pancreas, or liver in this study. These results are in agreement with those reported by Brenes et al. (1993) and Engberg et al. (2004) for xylanase, and with those reported by Corduk et al. (2008) and Ozdulven et al. (2009) for citric acid. In contrast with our results, Wu et al. (2004) reported that xylanase supplementation decreased the size and weight of digestive organs. It has been suggested that NSP can prevent effective contacts between digestive enzymes and digesta by increasing digesta viscosity. To overcome this negative effect, GIT secretion and motility may be increased, which may lead to increases in the size of the GIT, pancreas, and liver (Wang et al., 2005).

In conclusion, xylanase supplementation significantly decreased the viscosity of digesta and improved the performance of broilers fed a low-P wheat-based diet. In addition, xylanase enhanced retention of DM, energy, and CP. Inclusion of 40 g/kg of citric acid reduced feed intake and BW gain even in xylanase-supplemented groups, but citric acid significantly improved feed efficiency and retention of P in broilers fed low-P wheat-based diets. Inclusion of citric acid may also decrease the pH of crop contents. No interaction effect between xylanase and citric acid was observed in any of the responses measured.

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