Short communication

Effect of wheat bran inclusion in barley-based diet on villus morphology of jejunum, serum cholesterol, abdominal fat and growth performance of broiler chickens

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Abstract This research was conducted to investigate the effect of inclusion of wheat bran (as a source of insoluble fiber) in a barley-based diet, fed from 11 to 42 d of age, on villus morphology of jejunum, serum cholesterol level, abdominal fat pad and growth performance of broiler chickens. Three hundred and thirty six 10-d-old female Ross 308 chicks were allocated to six diets with four replicates of 14 birds per diet. The diets were a corn-based diet (CN); barley-based diet without (BL) or with multi-enzyme (BL+E, 500 mg/kg of the diet, Rovabio Excel 10%); and barley based-diet that contained 4 (BL+WB4), 8 (BL+WB8) or 12 (BL+WB12) percent wheat bran. Average daily gain, average daily feed intake and feed conversion ratio (FCR) were measured from 11 to 42 d of age. Serum cholesterol level was measured on d 24. Villus height (VH) and villus surface area (VSA) of jejunum, and relative weight (% of body weight) of abdominal fat pad were measured at 42 d of age. The birds receiving CN, BL+E, BL+WB4 and BL+WB8 had significantly (P < 0.01) lower FCR than those feeding on BL. VH (P < 0.01) and VSA (P < 0.001) in the jejunum increased in birds receiving CN, BL+E and BL+WB12 compared with BL birds. Serum cholesterol level in birds fed with BL+WB12 diet decreased (P < 0.05) compared with CN birds. The birds fed with BL, B+WB4, B+WB8, B+WB12 diets showed lower (P < 0.05) relative weight of abdominal fat pad compared with CN diet. In conclusion, the results of this study showed, when broiler chickens fed barley-based diet, the inclusion of lower levels of wheat bran in diet could have a positive effect on feed efficiency, whereas serum cholesterol level, VH and VSA were influenced with the inclusion of highest level of wheat bran.

Keywords: barley, broiler, villus morphology, performance, cholesterol, wheat bran

Introduction

One of the major challenges feeding cereals especially barley, wheat, oats and rye, to chicks is increase viscosity in gut digesta and subsequent slowing down of the digesta passage rate, leading to proliferation of unfavorable fermentative organisms in the small intestine, which is detrimental especially to the villus of the small intestine (Choc et al., 1996). In addition, the increasing the digesta viscosity reduces the diffusion rate of nutrients and digestive tract enzymes and hampers their interaction at the mucosal surface (Ikegami et al., 1990).

There are some evidences that hulls and brans (as a source of insoluble fiber) speed up feed passage rate (Kirwan et al., 1974; Rogel, 1985). Rogel et al. (1987) showed the beneficial effect of oat hulls inclusion in a wheat-based diet on starch digestion. On the other hand, birds raised on diets diluted with hulls or bran have exhibited improved performance (Sacranie et al., 2012) which may be due to increased gizzard function (Hetland and Svihus, 2001; Sacranie et al., 2012), hydrochloric secretion of gizzard and proventriculus (Peron et al., 2007; Svihus, 2011), digesta reflux between the gizzard and duodenum (Hetland et al., 2003) and pancreatic secretions (Hetland et al., 2003).

Wheat bran is one of the important sources of fiber in poultry nutrition. Wheat bran has high fiber and low bulk density. Leeson and Summers (2005) claimed that wheat bran has a growth promoting effect in poultry nutrition which is related to the modification of bacterial population in the digestive tract.

With regard to the general effects of dietary inclusion of brans or hulls on digestive traits of broiler (Hetland and Svihus, 2001; Hetland et al., 2003), favorable modification of the microflora population in the gut by feeding wheat bran in broilers (Leeson and Summers,
2005) and shortening the digesta transit time by feeding the hulls or brans (Rogel et al., 1987), it was hypothesized that, in the current study, wheat bran inclusion may improve the performance of broilers given barley-based diet. Villus height (VH) and villus surface area (VSA) were measured as a possible mechanism affecting the feed efficiency. Concerning the brans and hulls properties, it was assumed that, in barley-based diet, wheat bran inclusion might affect lipid metabolism. Hence, the serum cholesterol level and abdominal fat pad were measured in the present study.

Materials and methods

Birds and husbandry

Day-old female Ross 308 broiler chicks were housed in an environmentally controlled room and given a commercial starter diet and water ad libitum. At 11 d of age, three hundred and thirty six chicks were randomly distributed in groups of 14 birds in 24 litter-floored pens (1.5×1.5 m). Mean body weight of the chicks in all pens was similar (225±3.5 g). The birds were given ad libitum access to water and diets and exposed to a 23:1 light:dark cycle. Room temperature was kept at 32°C during the first 3 d of life and then, it was reduced gradually according to age until reaching 22°C at 24 d of age. This temperature was kept during 25 to 42 d of age.

Experimental diets

All diets were fed in mash form and met or exceeded the nutritional recommendations of Aviagen (2009) for Ross 308 female broilers. Six diets were used during 11-24 (Table 1) and 25-42 d of age (Table 2) which were a corn-based diet (CN); barley-based diet without (BL) or with multi-enzyme (BL+E, 500 mg/kg of the diet, Rovabio Excel 10%); and barley-based diet that contained 4 (BL+WB4), 8 (BL+WB8) or 12 percent wheat bran. Each treatment was replicated four times, and the experimental unit was the pen.

Table 1. Ingredient and chemical composition of the diets1 (g/kg, unless otherwise indicated) from 11-24 d of age

<table>
<thead>
<tr>
<th>Item</th>
<th>CN</th>
<th>BL</th>
<th>BL+E</th>
<th>BL+WB4</th>
<th>BL+WB8</th>
<th>BL+WB12</th>
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<tr>
<td>Ingredient</td>
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<td>Corn</td>
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<td>275.8</td>
<td>275.8</td>
<td>230.6</td>
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<td>Soybean meal</td>
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<td>310.0</td>
<td>300.0</td>
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<tr>
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<td>-</td>
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<td>300.0</td>
<td>300.0</td>
<td>300.0</td>
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<tr>
<td>Wheat bran</td>
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<td>-</td>
<td>-</td>
<td>40.0</td>
<td>80.0</td>
<td>120.0</td>
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<tr>
<td>Corn oil</td>
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<td>70.0</td>
<td>85.0</td>
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<td>Calcium carbonate</td>
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<td>16.5</td>
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<tr>
<td>Dicalcium phosphate</td>
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<td>10.4</td>
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<td>2.5</td>
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<td>1.1</td>
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<tr>
<td>Multi-enzyme Rovabio</td>
<td>-</td>
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<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Calculated analysis</td>
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<td></td>
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<td>Crude protein</td>
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<td>199.0</td>
<td>197.0</td>
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<tr>
<td>Methionine+Cystine</td>
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<td>9.3</td>
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<td>9.3</td>
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<td>12.1</td>
<td>12.1</td>
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<td>Threonine</td>
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<td>8.2</td>
<td>8.2</td>
<td>8.2</td>
<td>8.2</td>
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<tr>
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<td>9.0</td>
<td>9.0</td>
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<td>4.5</td>
<td>4.5</td>
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</tr>
</tbody>
</table>

1The diets were: a corn-based diet (CN); a barley-based diet without (BL) or with (BL+E) multi-enzyme (500 mg/kg of the diet, Rovabio Excel 10%); the barley-based diets that contained 4, 8 or 12% wheat bran (respectively, for groups BL+WB4, BL+WB8 and BL+WB12).
2The vitamin premix supplied the following per kilogram of complete feed: vitamin A, 9,000 IU (retinyl acetate); cholecalciferol, 2,000 IU; vitamin E, 18 IU (dl-a-tocopheryl acetate); vitamin B12, 0.015 mg; menadione, 2 mg; riboflavin, 6.6 mg; thiamine, 1.8 mg; pantothenic acid, 30 mg; niacin, 10 mg; choline, 500 mg; folic acid, 1 mg; biotin, 0.1 mg; pyridoxine, 3 mg.
3The mineral premix supplied the following per kilogram of complete feed: manganese (MnSO4·H2O), 80 mg; zinc (ZnO), 80 mg; iron (FeSO4·7H2O), 80 mg; copper (CuSO4·5H2O), 10 mg; selenium (Na2SeO3), 0.3 mg; iodine (Iodized NaCl), 0.8 mg; cobalt (CoCl2), 0.25 mg.
The multi-enzyme was a NSP-hydrolyzing enzyme multicomplex (Rovabio Excel 10%, Adisseo, France) of \( \beta \)-xylanase, \( \beta \)-glucanase, pectinase, cellulase and protease. It was a commercial supplement of *Penicillium funiculosum* product with 2200 units visco (equivalent to 1400 units AXC/g) of endo-1,4 \( \beta \)-xylanase and 200 ACL units of endo-1,4 \( \beta \)-glucanase/g of the supplement as the major enzymes.

### Growth performance

Body weight of chicks and feed consumption were determined by pen at 11 and 42 d of age, and average daily gain (ADG), average daily feed intake (ADFI), and feed conversion ratio (FCR) were determined from 11 to 42 d of age. Feed intake was adjusted for all mortalities, and their ADG was included in the calculation of FCR.

### Serum cholesterol level

At 24 d of age, two chicks per replicate (eight chicks/treatment) were randomly selected to measure their serum cholesterol level. The concentration of total cholesterol was analyzed in duplicate, using an automatic biochemical analyzer (Clima, Ral. Co, Spain), following the kit instructions (Pars Azmon, Iran).

### Abdominal fat pad and jejunal morphology

One chick per each replicate (four chicks/treatment) was sacrificed on d 42 to measure the relative weight of abdominal fat pad, and jejunal VH and VSA of experimental birds. The following formula was used to calculate the relative weight (% of body weight) of abdominal fat:

\[
\text{Relative weight of abdominal fat} = \left( \frac{\text{abdominal fat weight (g)}}{\text{live body weight (g)}} \right) \times 100
\]
was removed, washed in physiological saline solution, and fixed in 10% buffered formalin. Each segment was embedded in paraffin, and a 2-mm section of each sample was placed on a glass slide and stained with haematoxylin and eosin for examination (Sakamoto et al., 2000). Histological sections were examined with a Nikon phase contrast microscope (Nikon Eclipse 80i, Nikon Corp., Tokyo, Japan). Fifteen fields of view were measured in each intestinal section from one bird; for statistical analysis the average of these values was used. VH was measured from the top of the villus to the top of the lamina propria. VSA was calculated using the formula \((2\pi)(VW/2)(VH)\), where \(VW\) = villus width, and \(VH\) = villus height (Sakamoto et al., 2000).

**Statistical analysis**

Data were analyzed using the GLM procedure of SAS (SAS, 2003). When treatment effects were significant, the means were compared using the Least Significant Difference at \(P = 0.05\). Pen was the experimental unit, except for the relative weight of the abdominal fat pad, VH and VSA in which the individual bird was the experimental unit.

**Results and discussion**

The performance responses are given in Table 3. There was not a significant difference \((P > 0.05)\) of ADFI and ADG among treatments. The birds receiving CN, BL+E, BL+WB4 and BL+WB8 had significantly \((P < 0.01)\) lower FCR than those feeding on BL. The beneficial effects of enzyme supplementation in diets based on cereal with high content of soluble NSP have been proved in the literature (Choct et al., 1995). In the present study, the birds receiving BL+WB4, BL+WB8 and BL+WB12 did not show a significant difference on FCR compared with those fed on BL+E. This finding showed that using low levels \((4 \text{ or } 8\%)\) of the inclusion of wheat bran can be considered as a solution to improve the feed efficiency of broilers given barley-based diets. This phenomenon has been reported firstly by Rogel et al. (1987). Rogel et al. (1987) showed that the inclusion of oat hulls in a low-ME wheat-based diet improved the nutritive value of such wheat. Rogel (1985) revealed that dietary inclusion of oat hulls increased the feed passage rate through the distal part of the gastrointestinal tract. Stephen and Cummings (1979) indicated that brans or hulls absorb large amounts of water and maintain normal motility of the gut. It has been shown that the increasing the dietary inclusion level of hulls or brans reduced the residence time of digesta in the small intestine (Kirwan et al., 1974) and subsequently, may reduce the available time for anaerobic bacteria to colonize in the distal part of the gastrointestinal tract. It was reported clearly in the work of Langhout et al. (2000) that major adverse effect of highly methylated citrus pectin on nutrient digestibility was the result of fermentation and proliferation of bacteria in the gut. In our research, dietary inclusion of 4 or 8% wheat bran might improve the FCR through reduction of the viscosity of digesta, feed transit time and the population of unfavorable bacteria in the gut, although these parameters were not measured. In addition, the improvement in FCR might be related to the general beneficial effects of brans or hulls inclusion, probably due to the increase of gizzard function and development, HCL secretion, digesta reflux between the gizzard and duodenum (Hetland et al., 2003; Sacranie et al., 2012), and the population of favorable microflora (Leeson and Summers, 2005). Also it has been shown that brans or hulls make a spongy form in the digesta and easy to penetrate enzymes into the digesta (Sarikhan et al., 2010). At high level inclusion \((12\%)\) of wheat bran in the diet, FCR was not improved compared with those fed on CN or BL+E. This ineffectiveness may be related to the high increase of feed passage rate. Although too slow passage rate of digesta have a problem with the accumulation of unfavorable bacteria, there is insufficient time for digestion and absorption of nutrients when the feed passage rate is too fast. It seems the solution is to find a balance of feed passage rate that promotes nutrient utilization and at the same time does not result in the proliferation of unfavorable microflora. For example, Jorgensen et al. (1996) reported that the growth performance was decreased in birds fed on diet with high level of wheat bran compared with those contained medium level of its inclusion.

Table 3 represents the results of VH and VSA measurements. The birds receiving CN, BL+E and BL+WB12 had a significant increase in VH \((P < 0.01)\) and VSA \((P < 0.001)\) of jejunum compared with those fed on BL. The increasing villi height enhances the surface area and contact with nutrients (Silva et al., 2009). As shown in piglets by Pluske et al. (1997). VH correlates positively with empty body-weight gain. The presence of high viscous digesta may increase the rate of villus cell losses leading to villus atrophy (Montagne et al., 2003). \(\beta\)-glucans are soluble NSP and viscous, and the grains that are rich in these, such as oats and barley, can therefore increase the viscosity of intestinal contents (Bedford and Classen, 1992). The addition of enzymes to a barley-based diet improved the histological alterations of villus (Viveros et al., 1994). These findings were in agreement with the result of our research. The birds fed on BL showed shortened VH compared with those fed on CN. The birds receiving BL+E showed the
The inclusion of wheat bran at levels 4 or 8% in barley-based diet decreases the serum cholesterol level compared with those fed on BL. The improved FCR of the birds fed diets containing wheat bran, to some extent, might be related to the more digestion and absorption area of the villus. The beneficial effect of fiber on the VH was reported by Sarikhan et al. (2010). They showed that dietary inclusion of insoluble raw fiber concentrate at different levels (0.25, 0.5 and 0.75% inclusion) improved performance and increased the VH of ileum. However, from 1 to 21 d of age, only high levels of the inclusion (0.5 and 0.75%) increased the VH compared with control. The observed beneficial effect of high levels of fiber on the VH in their research was in agreement with our results. Bi and Chiou (1996) showed that chicks fed high dietary fiber had a larger intestinal villi and higher growth rate. The decreased transit time of the digesta in the diets with high brans or hulls might reduce the proliferation and also deleterious products of bacteria into the epithelium of the gut wall.

Table 3 shows the results of serum cholesterol level and the relative weight of abdominal fat pad. All the treatments (except the BL+E) reduced this parameter significantly ($P < 0.05$) compared with those fed on CN. The mechanism that a barley-based diet decreases the serum cholesterol level is the presence of high content of soluble fiber and consequently high viscous digesta and then probably increased proliferation (Hofshagen and Kaldhusdal, 1992) and bile salt hydrolyzing activity of non-favorable bacteria especially *Clostridium perfringenes* (Knarrenborg et al., 2002). The high viscous digesta reduces the efficiency of bile salts to solublize lipids. In addition, bacterial transformations of bile salts in the small intestine may reduce their absorption and the amount returning to the liver. Consequently, more bile acids are excreted into the faeces (Eyssen and van Eldere, 1984). Therefore, more blood cholesterol is used for the synthesis of bile salts. Another mechanism of faecal excretion of cholesterol, bile salts and lipids (Moundras et al., 1997) is their gelling by soluble and binding by insoluble fibers (Vahouny et al., 1980). Adrizal and Ohtani (2002) confirmed NSPs’ binding property with bile acids. Mathlouthi et al. (2002) reported that indigestible polysaccharides can act directly via increasing the bile acid excretion. Qujeq and Gharejeh (2001) reported that reduction in total cholesterol concentration and is probably the effect of enhanced reverse cholesterol transport in response to the intestinal loss of dietary fat. In the literature, a negative correlation exists between dietary fiber content and serum cholesterol level (Sundberg et al., 1995). Shahin and Abdelazim (2006) concluded that carcass fat of broiler had a considerable reduction using...
high fiber diets. Abdominal fat, carcass fat and total body fat yields were greatly depressed by feeding birds on high fiber diets and lead to less abdominal fat depots (Shahin and Abdelazim, 2006). Mourao et al. (2008) reported that birds fed diets containing insoluble fiber resulted in a lighter carcass with a lower level of abdominal fat pad in compare with control. A trend of reduction in serum cholesterol level (Sarikhani et al., 2009) and abdominal fat (Sarikhani et al., 2010) by feeding insoluble fiber were almost in agreement with our results. Their findings showed more reduction of serum cholesterol concentration by feeding high level of insoluble fiber compared with those of low or moderate inclusion. In agreement with our results, all the diets containing wheat bran decreased abdominal fat compared with that of CN. As mentioned above, the birds fed on BL+E had similar serum cholesterol level and relative weight of abdominal fat compared with those fed on CN which may be due to decreasing the digesta viscosity.

In conclusion, the results of this study showed, when broiler chickens fed barley-based diet, the inclusion of lower levels of wheat bran in the diet could have a positive effect on feed efficiency, whereas serum cholesterol level, VH and VSA were influenced with the inclusion of highest level of wheat bran.

References


*Communicating editor: Mohammad Salarmoini*
اثر افزودن سبوس گندم در جیره بر پایه جو روی مورفولوژی پرز زئوژنوم، کلسترول سرم، چربی بطنی و عملکرد رشد جوجه های گوشتی

چکیده
این تحقیق به منظور بررسی اثر افزودن سبوس سبز پرنده گندم (به عنوان منبعی از الافت تامنالو) در جیره بر پایه جو که از ۱۱ تا ۴۲ روزگی تغذیه شده، روي مورفولوژی پرز زئوژنوم، کلسترول سرم، چربی بطنی و عملکرد رشد جوجه های گوشتی طرح رژیم شد. ۳۶۶ جوجه ماده ۱۰ روزه را در دو گروه آزمایشی و کنترلی، بر اساس گروه های آماده پرز زئوژنوم و هر گروه در دو گروه از روز زده اختراعشده شدند. جیره ها شامل ۱) چربی بر پایه جو به مقدار آزمایشی (۳۲) چربی بر پایه جو حاوی مولتی-آنزیم (۱۰٪) ۲) چربی بر پایه جو حاوی ۴) در صد سبوس گندم (۳۲) چربی بر پایه جو حاوی ۵) چربی بر پایه جو حاوی ۶) در صد سبوس گندم و ۷) چربی بر پایه جو حاوی ۸) در صد سبوس گندم بودند. سطح کلسترول سرم در ۲۴ روزگی اندامگیری شد. ارتفاع و فضای سطح پرز زئوژنوم و وزن نسبی درصد از وزن زده، و ۱۲ روزگی اندامگیری شدند. برندهای تغذیه شده با چربی بر پایه جو میالگین اودایش نیس رنیاله، میالگین مصرم خوراک رنیاله ن ضریب تگبی خوراک این (P < 0/01)را نسبت به تغذیه چربی بر پایه جو حاوی ۸) درصد سبوس گندم نسبت به جیره بر پایه جو بیشتر بود. برندهای تغذیه شده با چربی بر پایه جو حاوی ۱۲) درصد سبوس گندم نسبت به جیره بر پایه جو بیشتر بود. در مجموع، نتایج این مطالعه نشان داد که در هنگام تغذیه چربی بر پایه جو، افزودن سطح پرز زئوژنوم سبز گندم به افزودن بالاترین سطح سبوس گندم تحت تأثیر قرار می گیرد.