Small intestinal histology, production parameters, and meat quality as influenced by dietary supplementation of garlic (*Allium sativum*) in broiler chicks

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Abstract

Four hundred day-old male hatchling chicks were obtained from the female line of Lohmann grandparent stock farms and fed rations supplemented with different levels (0%, 0.25%, 0.50%, 1.0%) of dried garlic powder. Each treatment consists of 5 pens. Feed and water were provided as ad libitum throughout the experimental period. Production parameters measured were body weight, feed consumption, feed conversion ratio. Final body weight and feed conversion ratio were not affected (P>0.05) by garlic supplementation. However, feed consumption was the lowest (P<0.05) at 0.5% and 1%. Small intestinal histological measurements were influenced (P<0.05) by garlic powder supplementation. In duodenum, villus length was the highest (P<0.05) in birds fed with diets containing 1% garlic powder, and villus and epithelial width were the lowest (P<0.05) in chicks fed with diets containing 0.5% of dried powder. In jejunum, the villus length was the highest (P<0.05) in birds fed with diets containing 1% garlic powder, while villus and epithelial width were the lowest (P<0.05) in chicks fed with diets containing 0.5% of dried powder. Carcass traits were not affected (P>0.05) by garlic supplementation. All meat quality parameters measured were not affected (P>0.05) by garlic powder supplementation (cooking loss percentage, shear force, lightness, redness and yellowness), except for juiciness percentage and pH, which were the lowest (P<0.05) at 1.0% and 0.25%, respectively. As a conclusion, this study shows that garlic at a 0.5% level might be of beneficial effect on intestinal morphometry parameters, as well as on production parameters.

Introduction

Over the past four decades, a significant progress in the genetic selection of meat-type chicken has been achieved. In return, broiler production became thriftier and registered higher body weights in a shorter period of time. However, such genetic progress made chicken very fragile and more susceptible to diseases. So, attaining high production levels became very challenging without the use of synthetic compounds in feed; for example, subtherapeutic antibiotics supplementation. The safety of such practices has been questioned and their use is becoming more restricted in many regions of the world.

Improving meat quality with feed additives is interesting in broiler production. Garlic (*Allium Sativum*) has been used in animal rations. Several components of garlic, including sulfur-containing compounds, have different activities, such as antibacterial and antioxidiant ones. Also, garlic compounds have a hypcholesterolemic effect in chickens (Qureshi et al., 1983; Konjufca et al., 1997). Van Damme et al. (1991) reported the presence of lectins in the bulbs of garlic (*Allium sativum agglutinin*, ASA). According to their study, Bulbs of garlic are known to accumulate two types of mannose binding lectins, the heterodimeric and the homodimeric. Though these two lectins differ in the lengths of their polypeptide chains, they exhibit marked similarities with respect to their primary sequence, post translatational modifications, serological properties, immunochemical attributes as well as carbohydrate binding properties. Gastrointestinal tract development and health is the key to productivity in all farm animals and poultry. The digestive functions could be considered the most limiting factors in performances. Due to its antimicrobial properties, garlic has been tested as an alternative growth promoter in broiler chickens (Freitas et al., 2001; Demir et al., 2003 and Lewis et al., 2003). Freitas et al. (2001) showed that body weight gain and feed conversion ratio in broiler chickens, that received a diet supplemented with commercial garlic product at concentration up to 45 Kg/ton were not affected by garlic supplementation. However, Lewis et al. (2003) and Demir et al. (2003) reported a trend in improved body weight gain and feed conversion ratio in broiler chickens fed low concentration of commercial garlic product. Tolba and Hassan (2003) found that garlic as a natural feed additive, improved broilers growth, feed conversion ratio, and decreased mortality rate. Demir et al. (2003) indicated that there was a decrease in crypt depth in the ileum of broilers given dietary natural growth promoters such as garlic.

Based on the scientific and medical research on the health properties of various forms of garlic, we propose the use of garlic as a feed additive in broiler rations. In a previous study, garlic supplementation improved laying performance, egg quality characteristics; it also extended the shelf life without affecting the yolk cholesterol level (Qatramiz, 2006). The aim of this study is to investigate the effect of garlic administration on performance, carcass parameters, meat quality and small intestinal histology of broiler chicks.

Materials and methods

A total of 400 day-old male hatchling chicks, obtained from the female line of Lohmann grandparent stock farms, were reared and grown to the market age (42 days) under standard commercial conditions. At one-day of age, chicks were randomly allocated into four different pens, for the first two weeks of age. Each pen was equipped with wood shavings, and an automatic bell-drinker and one tube feeder. A corn-soybean based diet was offered for all groups throughout the experimental period as indicated by Table 1. Ration was prepared according to National Research Council (1994). Group 1 was offered a basal diet served as a control; groups 2, 3 and 4 were offered a basal diet with 0.25%, 0.50%, and 1.0% of dried garlic, respectively. Having chicks at one group for each treatment, through the first two weeks

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Fresh garlic bulbs (Allium sativum) were obtained and prepared as follows: fresh garlic bulbs were weighed, peeled, and garlic cloves were minced using an electrical mincer. Minced garlic was weighed and placed in trays and dried at 40°C for 3 days. All garlic used in this experiment was re-weighed after drying, and dry matter was calculated as (initial weight – final weight ×100)/initial weight. Dried garlic was grounded and stored at room temperature and added to rations at the time of formulation.

At 28 days of age, 20 birds (5 birds/treatment) were randomly selected and bled, then slaughtered to sample the small intestine. Small intestine samples were washed with a solution (formalin 90% and distilled water 10%), then they were stored in a chiller at 4°C. Feed and water were provided as ad libitum throughout the experimental period with a photoperiod of 23 h of light and 1 h of darkness. Feed intake and refusal were recorded daily and body weight was measured weekly to determined growth performance. Feed conversion ratio was calculated as the ratio between total feed consumption to final body weight. Chicks were vaccinated against New Castle and Infectious Bronchitis diseases at the age of 6 days, and against the Infectious Bursal Disease at the age of 14 days. Ration was formulated every week and dried garlic was added to the formulated ration and was properly hand-mixed to ensure homogeneous distribution of the powder.

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of age, eased the brooding management and gave us more control during vaccination. At the beginning of the third week of age, each group of chicks was randomly distributed into five different pens; each pen was considered as an experimental unit and 20 chicks were allocated in each experimental unit.

Table 1. Ingredient of the commercial starter and finisher broiler diet.

| Ingredient      | Composition, g/kg |
|-----------------|-------------------|
|                 | Starter 1-21 day   |
|                 | Finisher 22-42 day |
| Corn            | 624.8             |
| Soybean         | 302               |
| Fish meal       | 30                |
| Oil             | 10                |
| Limestone       | 12                |
| Dicalcium phosphate | 12.5           |
| Methionine      | 1.7               |
| Additives*      | 7                 |
| Total           | 1000              |
| Metabolizable energy, kcal/kg | 3002 |
| Crude protein, %| 21.3              |

*Includes salt, mineral and vitamin premix. (Co 259 mg/kg, Cu 700 mg/kg; I 300 mg/kg; Fe 40,000 mg/kg; Se 200 mg/kg; Mn 100,000 mg/kg; Zn 50,000 mg/kg; Vitamin A 60,000 U; Vitamin K, 3000 mg; Ca; Panthothenate 40,000 mg; Vitamin D3 15,000 U; Vitamin B1 5000 mg; Vitamin B2 250 mg; Folic acid 2000 mg; Vitamin E 40,000 U; Vitamin B6 20,000 mg; Nicotine 75,000 mg; Biotin 150 mg), anti mold and anti coxidian; garlic powder was added as an additional feed additive.

After that, 6 cores from cooked muscles were used to evaluate tenderness using Warner-Bratzler shear blade with the triangular slot cutting edge mounted on Salter model 235 (Warner-Bratzler meat shear, G-R Manufacturing, Inc., Manhattan, KS, USA). The pH values were determined in duplicate samples using the iodoacetate method as described by Jeacocke (1977) and Sams and Janký (1986). One to 1.5 g of raw meat were put into plastic test tubes containing 10 mL of neutralized 5 mM iodoacetate reagent and 150 mM of KCl, and then homogenized using a homogenizer (Ultra-Turrax T8, IKA Labor-technik, Staufen, Germany). The ultimate pH values of the homogenate were measured using a pH meter (pH Spear, Large screen, waterproof pH/temperature/Tester, double injection, model 35634-40, Eurotech Instruments Ltd., Ramaipuram, Malaysia). Color of the muscles was measured by placing the muscles on polystyrene trays, and then covered with plastic oxygen-permeable film and allowed to oxygenate for 3 h at 2°C (Geesink et al., 2000). A colorimeter device (12MM Aperture U123 59730-30, Cole-Parameter Int., Accuracy Microsensors Inc., Pittsford, NY, USA) was used to objectively measure CIELAB (Commission International d’Eclairage) lightness (L*), redness (a*) and yellowness (b*). The water binding properties of the pectoralis major muscles were estimated by measuring the amount of water released from the muscle protein by the application of force (expressible juice), also measuring the ability of muscle to bind water through the expression of the pectoralis major muscle. The water binding properties of the pectoralis major muscles were estimated by measuring the amount of water released from the muscle protein by the application of force (expressible juice), also measuring the ability of muscle to bind water through the expression of the pectoralis major muscle.
protein to retain water present in excess and under the influence of an internal force (juiciness). Juiciness was measured using the method described by Grau and Hamm (1953) modified by Sainudo et al. (1986), using samples of approximately 5 gm of raw meat (initial weight). Each sample was cut into small pieces and the pieces were covered with two filter papers (qualitative, 185 mm φ circles, fine crystalline retention, Whatman Int. Ltd, Maidstone, Kent, UK) and 2 thin plates of quartz material, then pressed with a weight of 2500 gm for 5 min. The meat samples were then removed from the filter paper and their weight was recorded (final weight). Juiciness percentage was reported as the weight lost during sample pressing divided by the initial sample weight and expressed as a percentage.

Data was analyzed as a completely randomized design where all measured variables were classified by their garlic level (0, control; 0.25%; 0.50% and 1.0%). The statistical analysis was attained by the General Linear Model procedure of SAS (1990). A probability level of P≤0.05 or as described in the text was considered statistically significant.

Results and discussion

Broiler growth performance

Performance parameters are presented in Table 2. Final body weight was 2219, 2164, 2188, and 2135 g for 0%, 0.25%, 0.50%, and 1.0% garlic powder groups, respectively. Administered garlic powder did not manifest body weight markedly, however, group of chicken fed 0.25% consumed numerically more (P=0.05) feed compared to control group. Therefore, chicken fed 0.25% was considered to be numerically the least efficient group regarding their feed conversion ratio. Konjufca et al. (1997) reported that feeding dietary garlic from hatching to 21 days of age did not affect body gain or feed conversion ratio of chickens. Qureshi et al. (1983) as well did not report any differences in final body weight and daily feed consumption of pullets fed diets with various garlic products at levels equal to about 50 kg/ton of added garlic bulb. However, Lewis et al. (2003) and Demir et al. (2003) reported improved feed conversion ratio and body weight gain in broilers fed garlic. The variety of garlic preparation and administration methods limits the possibility of comparing results of the current study with previous findings.

Small intestine histology

Histological parameters of villus length and width and epithelial width for the proximal duodenal loop and jejunum are detailed in Table 3. The statistical analysis of the collected data showed that villus of different small intestine sections were elongated (P<0.05) in response to the garlic powder supplementation (Table 3). Villus length increased in a linear fashion with respect to the level of garlic powder. Villus length was higher in garlic-fed birds compared to control-fed birds, with maximum length recorded for birds fed 1.0% garlic powder. Villus width results were not consistent across all small intestine sections. In duodenum, villus width was higher (P<0.05) in garlic-fed birds compared to control-fed group; and widest (P<0.05) villus was observed among those fed a diet with 0.50% powdered garlic. In Jejunum, the control fed group had the widest (P<0.05) villus compared to the garlic-fed groups. The response of epithelial width to the dietary inclusion of powdered garlic was not a consistent cross in different sections of the small intestine. Epithelial width was the highest (P<0.05) in the duodenum of broiler birds supplemented with 0.50% garlic powder when compared with the remaining groups. On the opposite, garlic powder supplementation regressed (P<0.05) epithelial width in jejunum (Table 3). Epithelial width of jejunum in the control fed-group maintained wider (P<0.05) cells when compared with the garlic-fed groups. The results reported in this study are in complete agreement to what had been reported in the literature. Garlic administration stimulated the selective population of intestinal cells. Gupta and Sandhu (1998) reported that feeding garlic agglutinin to rats caused lengthening intestinal villi due to cellular hypertrophy and hyperplasia. They also reported that feeding garlic agglutinin to rats caused thinning and sloughing off of the jejunum villus, lengthening of the villi and an increase in the number of goblet cells. On the other hand, it has been reported that villus length, width and surface were reduced in birds fed barley-based diets (Moharrery and Mohammadpour, 2005). They also reported that villi length and surface gradually decreased from duodenum to ileum sections. Santin et al. (2001) reported that when birds were treated with 0.2% of yeast cell walls, villus height was increased and crypt depth was

Table 2. Least-square means for performance parameters of birds as affected by garlic powder supplementation from 14–42 days.

| Performance parameters                  | Garlic powder percentage | Standard error | P   |
|-----------------------------------------|--------------------------|----------------|-----|
|                                        | 0% | 0.25% | 0.50% | 1.00% |
| Body weight at 14 days, g               | 454b | 458b | 448b | 442d | 0.82 | 0.092 |
| Final weight, g                         | 2219 | 2164 | 2188 | 2135 | 44.2 | 0.481 |
| Total weight gain, g                    | 1764 | 1704 | 1739 | 1692 | 44.0 | 0.541 |
| Average daily gain, g/d                 | 63  | 61   | 62   | 60  | 1.6  | 0.541 |
| Cumulative feed consumption, g          | 3328b | 3514a | 3239b | 3200b | 85.0 | 0.040 |
| Daily feed consumption, g/d             | 119b | 126b | 116b | 114b | 3.0  | 0.040 |
| Feed conversion ratio                   | 1.7 | 1.8  | 1.7  | 1.7  | 0.04 | 0.092 |

*Means in the same row, bearing different superscript letters differ according to the indicated P.

Table 3. Least-square means of Histological parameters of villi length and width and epithelial width for proximal duodenal loop and jejunum of broiler birds as affected by garlic powder supplementation.

| Villi parameters, µm                     | Garlic powder percentage | Standard error | P   |
|-----------------------------------------|--------------------------|----------------|-----|
|                                        | 0% | 0.25% | 0.50% | 1.00% |
| Duodenum                                |                |                |     |      |
| Villi length                            | 991c | 1385b | 1489b | 1613a | 52  | 0.001 |
| Villi width                             | 133e | 155a | 223a | 169b | 4   | 0.001 |
| Epithelial width                        | 56b | 59a | 71a | 59b | 2   | 0.001 |
| Jejunum                                 |                |                |     |      |
| Villi length                            | 751b | 1068a | 858a | 1151a | 66  | 0.001 |
| Villi width                             | 171a | 140b | 117b | 132c | 7   | 0.001 |
| Epithelial width                        | 69a | 54a | 39a | 44a | 3   | 0.001 |

*Means in the same row, bearing different superscript letters differ according to the indicated P.
reduced. It is possible that some garlic constituents by direct interaction with selective population of intestinal cells or by the indirect effect of via gut endocrine cells are able to stimulate gut cellular proliferation (Gupta and Sandhu, 1998). The number of proliferations and the epithelial cells turn over have a great impact on the protein and energy requirements of the small intestinal mucosa (Simon, 1989). Increased crypt depth indicates an increased villus length, as the rate of sloughing is high; thus, the epithelial cells will use more energy to compensate for the sloughed cells, which in turn reduces the energy available for lean tissue mass synthesis. Large crypt indicates a fast turnover and a high demand for new tissue (Yason et al., 1987).

Krinke and Jamroz (1996) reported reduced duodenal cell proliferation and a thinner epithelial thickness in chicks fed antibiotic, which is in agreement with the observed effect of garlic meal in present research. Thinner intestinal epitheliums enhance nutrient absorption and reduce the metabolic demands of the gastrointestinal system (Visek, 1978). Thinning of the gastrointestinal walls tract may be due to the inhibition of the microbial production of polyamines and volatile fatty acids, known to increase enterocyte turnover rate and activity. This increased net energy committed to maintaining the luminal tissue comes at the expense of more productive purposes such as muscle accretion (Bedford, 2000). In the current study, it was clear that garlic administration maintained longer and more slender villi, especially in the duodenum section.

**Carcass characteristics**

Results for carcass weight, and cuts and dressing percentages are presented in Table 4. Results showed that garlic powder had no influence on cold carcass weight or dressing percentage (Table 4). However, values for cold carcass weight and dressing percentage were numerically higher in birds fed diets containing 0.50% garlic powder when compared with the other treatments. Carcass cuts percentages were all comparable among garlic powder percentages for breast, leg, back, neck, and wing cuts, and abdominal fat. Breast percentage were 31.2, 30.7, 31.1, and 30.2 for birds fed diet containing 0%, 0.25%, 0.50%, and 1.0% of dried garlic powder, respectively.

Bampidis et al. (2005) reported that fasted body weight, cold carcass weight and carcass yield were not affected by feeding diets containing different levels of garlic bulb and husks fed to lambs. Payne et al. (2001) reported that addition of isoflavones to corn-soy protein concentrate diet increases carcass leanness and decreases carcass fat and results in carcass traits similar to, or better than, those of barrows fed corn-soybean meal. However, in a second study, Payne et al. (2001) reported that isoflavones supplementation in excess of that present in a typical corn-soybean meal diet does not affect growth performance, carcass traits, or meat quality of growing-finishing gilts. The different responses in barrows versus gilts may be due to the different treatments used or to an actual gender difference. Isoflavones have several biological properties including inhibition of tyrosine kinase and DNA topoisomerase, antioxidant activity, immune system activity, and estrogenic and antiestrogenic activities (Kurzer and Xu, 1997; Zhang et al., 1997). Adibmoradi et al. (2006) reported that small intestinal morphological changes in chickens due to dietary garlic meal supplement demonstrate that absorptive process could be activated by garlic meal as an antibiotics alternative growth promoter.

**Meat quality**

Meat quality measurements are filed in Table 5. Values of cooking loss percentages, shear force values, color coordinates (L*, a*, b*) registered comparable means to control group (Table 5), while juiciness percentage and pH differed (P<0.05) among groups.

Cooking loss represents the percentage of water liberated upon cooking and calculated as a percent of the initial weight. Warner-Bratzler shear force values were measured on cooked meat. Jiang et al. (2007) reported that the shear force values of broiler breast muscles were not affected by isoflavone supplementation. Zhang et al. (2005) reported that the shear force of broiler breast meat decreased when yeast was included in the birds ration.

Color coordinates (L*, a*, and b*) are presented in Table 5. Lightness (L*) represents the degree of lightness in meat color; the higher the value, the brighter the color. Redness (a*) represents the degree of meat redness and yellowness represents the degree of meat yellowness. The collected data showed that the color coordinates were not influenced by different levels of garlic powder supplementation. However, meat redness numerically registered the lowest value in breasts excised from birds fed diet supplemented with 1.0 percentage of garlic powder when compared to their control-fed group. The rate of meat discoloration is believed to be related to the effectiveness of oxidation process and enzymatic reducing sys-

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**Table 4. Least-square means of fasted weight, cold carcass weight, cuts and dressing percentage for broiler birds as affected by garlic powder supplementation.**

| Carcass parameters | Garlic powder percentage | Standard error | P   |
|-------------------|--------------------------|---------------|-----|
|                   | 0% | 0.25% | 0.50% | 1.00% |                |
| Fasted weight, g   | 2116 | 2071 | 2163 | 2010 | 70 | 0.20 |
| Cold carcass weight, g | 1631 | 1593 | 1665 | 1521 | 55 | 0.12 |
| Breasts, %         | 31.2 | 30.7 | 31.1 | 30.2 | 0.50 | 0.33 |
| Legs, %            | 28.2 | 28.4 | 28.3 | 28.1 | 0.30 | 0.92 |
| Wings, %           | 10.3 | 10.2 | 10.0 | 10.1 | 0.14 | 0.41 |
| Neck, %            | 7.4  | 7.4  | 7.2  | 7.2  | 0.14 | 0.42 |
| Back, %            | 20.9 | 21.1 | 21.4 | 21.5 | 0.23 | 0.14 |
| Abdominal fat, %   | 1.3  | 1.6  | 1.5  | 1.5  | 0.11 | 0.08 |
| Dressing, %        | 75.8 | 75.4 | 75.8 | 74.3 | 0.68 | 0.26 |

**Table 5. Least-square means of fasted live weight, cold carcass weight, dressing percentage and cuts’ weights for broiler birds as affected by garlic powder supplementation.**

| Meat quality parameters | Garlic powder percentage | Standard error | P   |
|-------------------------|--------------------------|---------------|-----|
|                         | 0% | 0.25% | 0.50% | 1.00% |                |
| pH                      | 6.08a | 5.94a | 6.10a | 6.04a | 0.04 | 0.02 |
| Cooking loss, %         | 23.7b | 24.60 | 24.30 | 24.30 | 0.52 | 0.51 |
| Juiciness, %            | 33.50b | 33.40b | 31.30b | 30.50b | 0.60 | 0.001 |
| Shear force, kg/cm²     | 2.14 | 2.30 | 2.38 | 2.13 | 0.13 | 0.39 |
| Colour coordinates      |               | L* | L* | L* | L* |        |       |
|                         | 49.62 | 51.04 | 49.28 | 50.29 | 1.00 | 0.38 |
|                         | 2.26  | 2.57  | 2.69  | 2.04  | 0.33 | 0.29 |
|                         | 13.26 | 13.12 | 13.97 | 14.18 | 0.85 | 0.77 |

(a,b) Means in the same row, bearing different superscript letters differ according to the indicated P-value.
tems in controlling the metmyoglobin levels in meat (Faustman and Cassens, 1989). It appears from the current results that garlic supplementation did not affect the oxidation process leading to color alteration except at the redness level. Jiang et al. (2007) reported that lightness increased when isoflavones were added to broiler diets.

The pH value measured after thawing (Table 5) was affected (P<0.05) by garlic powder supplementation. Breast muscles excised from birds fed 0.25% garlic powder registered the lowest (P<0.05) pH value. Jiang et al. (2007) reported that pH increased when isoflavones are added to broiler diets.

Conclusions

In conclusion, there is evidence in this study to show that garlic at a 0.5% level might be of beneficial effect on intestinal morphometry parameters as well as on production parameters. Also, the results reported in the study strongly suggested that garlic maintained taller and sometimes wider villus at different sections of the intestine, especially in the duodenum. We believe that more conclusive results might be obtained using a commercial broiler cross and taking more frequent cuts from the proximal and distal duodenal sites for morphometric parameter evaluation.

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