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≪Research Note≫

The Effect of Malic Acid on Performance and Some Digestive Tract Traits of Japanese Quails

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To evaluate the effect of malic acid (MA) on performance and some digestive tract (gut) traits of Japanese quails, 320 quails were divided into 4 treatment groups each with 4 replicates containing 20 birds. While one of groups was fed a mash form basal diet with no additives (0 MA), other groups were fed the diets with malic acid at 0.4 (0.4 MA), 0.8 (0.8 MA) or 1.2 (1.2 MA) g/kg from 7 to 42 d of age. The weight gains of 0.8 MA and 1.2 MA birds and the feed consumption of 1.2 MA were higher than those of 0 MA. Addition of the malic acid to the diets did not affect the feed efficiency. The 0.8 MA birds had a higher carcass weight compared to other groups. The 1.2 MA birds had a higher gut length compared to the 0 MA. The results indicated that feeding diets with malic acid at 0.8 and 1.2 g/kg might increase weight gain and feed consumption without affecting feed efficiency and a significant benefit of feeding diet with malic acid at 0.8 g/kg compared to the control diet was obtained in terms of carcass weight in the Japanese quails.

Key words: digestive tract, growth performance, malic acid, organic acid, quail

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Introduction

Recently, most of antibacterial performance promoters have been banned, because feeding with antibiotics is risky (Neu, 1992; Casewell *et al.,* 2003) due to not only cross-resistance but also to multiple resistances. The ban on the use of antibiotics as growth promoters in the European Union and the potential for a ban in most parts of the world has prompted the search for alternative feed supplements in animal production (Casewell *et al.,* 2003; Decuypere and Dierick, 2003). Prebiotics, probiotics and organic acids are three of the several approaches that have a potential to reduce enteric disease and improve performance in poultry, and reduce subsequent contamination of poultry products (Patterson and Burkholder, 2003; Ricke, 2003).

Although the antimicrobial mechanism(s) of organic acids are not completely understood, organic acids used as antimicrobial feed additives are acetic, propionic, butyric, formic, citric, fumaric, lactic, sorbic, and malic acids or commercial acid blends. Organic acids are often included in diets as mould inhibitors, but they are known to provide protection against pathogens such as some bacteria and yeasts (Szylit *et al.,* 1988; Dhawale, 2005). Therefore, they have made a great contribution to the profitability in the poultry and provided people with healthy and nutritious poultry products (Patten and Waldroup, 1988; Ricke, 2003; Moharrery and Mahzonieh, 2005).

Malic acid is an organic dicarboxylic acid formed in the metabolic cycles in the cells of plants and animals, including poultry, and plays a key role in the energy-producing Krebs cycle. Malic acid is not an antibiotic but, if used correctly along with the nutritional, managerial and biosecurity measures, it can be a powerful tool in maintaining the health of the gastrointestinal tract (gut) of poultry, thus improving their zootechnical performances (Maheswari and Kadirvel, 1996; Moharrery and Mahzonieh, 2005). Maheswari and Kadirvel (1996) reported that the use of malic acid in broiler diets did not have an acidifier effect on the body weight, feed conversion, or mortality during the starter phase. The effects of different levels of malic acid given in a water pan on the live weight, feed consumption and characteristics of carcass and gut in the broiler chickens have been studied (Moharrery, 2005; Moharrery and Mahzonieh, 2005). However, there is incomplete knowledge on the effect of exogenous malic acid supplementation to the compound feed on the growth, and carcass and gut characteristics of poultry. It is not clear also, what the response of quails, which is frequently recognized as a model animal for poultry, especially for chicken or turkey broiler was? Accordingly, the present study was conducted to determine the effects of feeding diets with malic acid on growth performance, carcass and gut characteristics in Japanese quails.

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Materials and Methods

A total of 320 seven-day-old quail chicks were divided into 4 treatment groups each with 4 replicates containing 20 birds (10 male and 10 female quails). The birds were sexed on a basis of the dimorphic morphological traits a week after formation of experimental groups. The quail chicks were reared in floor pens (85×75×75 cm) with wood-shavings during the experimental period. The pens were fitted with feeders and waterers. Lighting was provided for 23 h per day throughout the experimental period by 2 fluorescent bulbs. Mean ambient temperature was reduced by 3℃ per week from 30 to 24℃ and the relative humidity was maintained within a range of 60–70%. While one of groups was fed a commercial basal (control) diet without malic acid (0 MA), other groups were fed the diets with DL-malic acid (CAS No.: 6915-15-7, Doruk Gida ve Kimya Amb. T. A. S., Istanbul, Turkey) at 0.4 (0.4 MA), 0.8 (0.8 MA) or 1.2 (1.2 MA) g/kg from 7 to 42 d of age. All birds were fed *ad libitum* and free access to water. All experimental diets (Table 1) were fed in mash form. The study was approved by the local Ethical Committee of Ondokuz Mayis University for Experimental Animals, and ascertained that the experiment is not an unnecessary repetition of previous experiments.

Table 1. **Ingredient and composition of experimental diets**

Item	g/kg
Ingredients	
Yellow corn	408.7
Soybean meal (45% crude protein)	226.7
Full fat soybean	120.0
Wheat	100.0
Sunflower meal	60.0
Meat and bone meal	30.0
Fish meal	20.0
Vegetable oil	20.0
Dicalcium phosphate (DCP)	5.5
Vitamin and mineral premix†	2.0
Salt	2.5
Limestone	2.5
DL-methionine	1.1
Calculated nutrient content per kg diet	
Metabolizable energy (kcal)	3200.0
Crude protein (g)	239.7
Lysine (g)	18.3
Methionine (g)	5.1
Calcium (g)	9.0
Available phosphorus (g)	4.8

† Vitamins and trace elements included to provide the following per kg premix: 1,200,000 IU vitamin A; 160,000 IU vitamin D_3; 1,600 mg vitamin E; 400 mg vitamin K_3; 200 mg vitamin B_1; 600 mg vitamin B_2; 400 mg vitamin B_6; 8 mg vitamin B_{12}; 4,000 mg vitamin C; 4 mg Biotin; 60 mg Folic acid; 800 mg Ca D-pantotenat; 80,000 mg Choline chloride; 2,000 mg Niacin; 8,000 mg Mn. 3,000 mg Fe; 6,000 mg Zn, 500 mg Cu, 50 mg Co, 200 mg I, 20 mg Se.

Body weight and feed consumption were recorded on a pen bases on 7 and 42 d of age. Feed efficiency (g feed/g gain) was calculated from the body weight and feed consumption data. To determine weights of gut and edible inner organs (heart+liver+gizzard) of Japanese quails, on 42 d of age, 4 quails (2 male and 2 female) from each replicate (a total of 16 quails from each treatment) with body weight within 1 standard deviation of the mean treatment weight were slaughtered. The gut, from the oesophagus to the cloaca, and organs were carefully excised. Any digesta remaining in the whole gut were emptied by gentle pressure. Empty weights of gizzard and weights of heart and liver were recorded as the weight of edible inner organs. Length of the whole gut was recorded. Weights of the gut and edible inner organs (g/100 g body weight), the length of gut (cm/100 g body weight) were expressed as a portion of body weight.

The complete randomized model was used to analyze the data for weight gain, feed consumption, feed efficiency and characteristics of carcass and gut. All data were analyzed according to the ANOVA model, using the GLM procedure of the Statistical Package for the Social Sciences (SPSS, 1999). Data from levels of malic supplementation were analysed as an orthogonal polynomial. Thus linear, quadratic and cubic effects were determined by orthogonal polynomial contrasts. Student-Newman-Keuls multiple-range test was used to compare mean values of treatments at a significance level of $P<0.05$ (SPSS, 1999). All percentage data were converted to arcsines prior to analysis and untransformed means are presented. For performance data, pen means served as the experimental unit for statistical analysis. For data on relative weights and length of the gut, individual birds were considered as the experimental unit.

Results

Mortality has not occurred in any of the groups during the experiment. Body weight gain, feed consumption and feed efficiency in Japanese quails given the experimental diets are presented in Table 2. For daily body weight gain, the effect was both linear and quadratic. Daily weight gains of birds in the 0.8 MA and 1.2 MA groups were higher ($P<0.05$) than those in the 0 MA group. No differences were observed among the 0.4 MA, 0.8 MA and 1.2 MA groups for the daily weight gain and daily feed consumption ($P>0.05$). The daily feed consumption linearly increased with malic acid supplementation. The daily feed consumption of birds in the 1.2 MA was higher ($P<0.05$) than those in the 0 MA. Addition of the malic acid to the diets did not affect the feed efficiency ($P>0.05$).

Means for carcass weight, dressing percentage, the relative weight and length of gut and the relative weight of edible inner organs at 42 d of age are shown in Table 3. For carcass weight, the effect was quadratic. Birds fed the diets with malic acid at 0.8 g/kg had a higher ($P<0.05$) carcass weight compared to other groups. No differences were observed among the 0 MA, 0.4 MA and 1.2 MA groups

Table 2. **Body weight gain, feed consumption and feed efficiency in Japanese quails fed diets without or with malic acid**

Measures	0 MA	0.4 MA	0.8 MA	1.2 MA	S.E.M
Initial body weight, g	25.8	25.7	25.8	25.7	0.07
Body weight gain, g/day/bird	4.0^{a}	4.2^{ab}	4.4^{b}	4.3^{b}	0.03
Feed consumption, g/day/bird	17.1^{a}	17.5^{ab}	17.8^{ab}	18.0^{b}	0.16
Feed: gain, g: g	4.09	4.10	4.12	4.18	0.03

^{a,b}: Values in the same row not sharing a common superscript differ significantly ($P<0.05$).
S.E.M: pooled standard error of the mean.

Table 3. **Carcass weight (g), dressing percentage and weights of gut and edible inner organs (g/100 g body weight), and length of gut (cm/100 g body weight) in Japanese quails fed diets without or with malic acid**

Measures	0 MA	0.4 MA	0.8 MA	1.2 MA	S.E.M
Carcass weight	116.2^{a}	117.2^{a}	121.2^{b}	118.9^{a}	0.68
Dressing percentage	70.09	67.86	67.41	67.48	0.41
Edible inner organs	5.33	5.36	5.27	5.24	0.04
Full gut weight	12.87	12.17	12.09	11.97	0.27
Full gut length	49.73^{a}	50.63^{ab}	52.18^{ab}	53.68^{b}	1.27

^{a,b}: Values in the same row not sharing a common superscript differ significantly ($P<0.05$).
S.E.M: pooled standard error of the mean.

in terms of the carcass weight ($P>0.05$). The relative gut length linearly increased with malic acid supplementation. Birds fed the diet with malic acid at 1.2 g/kg had a higher ($P<0.05$) gut length compared to birds fed the control diet, but the feeding diets with malic acid did not affect dressing percentage, weights of gut and edible inner organs ($P>0.05$).

Discussion

The result of the present study indicates that feeding diets with malic acid at 0.8 and 1.2 g/kg might increase the growth performance and feed consumption without any concomitant reduction or augmentation in the feed efficiency and mortality in Japanese quails. These outcomes are in general agreement with previous studies using organic acids in poultry diets, such as malic acid (Maheswari and Kadirvel, 1996), fumaric acid (Patten and Waldroup, 1988; Skinner *et al.,* 1991) and Lucta'cid (Daskiran *et al.,* 2004). However, they disagree with the previous observations of Moharrery (2005) and Moharrery and Mahzonieh (2005) in broilers. These researchers (Moharrery, 2005; Moharrery and Mahzonieh, 2005) showed that the malic acid given in the waterer pan did not affect body weight gain, feed efficiency and dressing percentage, but that feed consumption tended to be lower in broilers on treatment with 0.15% malic acid concentration to compare with control (0% malic acid). Other studies have also reported that organic acids improved growth performance of growing pigs and piglets (Roth and Kirchgessneer, 1998; Partanen *et al.,* 2002; Hanczakowska *et al.,* 2005; Kim *et al.,* 2006). However, Karaoglu *et al.* (2004) failed to observe any effect on the performance of chickens when humic acid was added to the feeds, which is in contrast to the findings of this experiment with quails. These discrepancies may be attributed to differences in the inclusion level and form (e.g. feed or water), in characteristics, such as chain length, side chain composition, pkA values and hydrophobicity of organic acids and/or in animal species used in the present and previous studies (Ricke, 2003; Moharrery and Mahzonieh, 2005; Jarquin *et al.,* 2007).

In all experimental groups the feed efficiency was more than 4 g feed/g gain which is too high, because of its feeding behavior resulting in a significant wastage of feed (Wang *et al.,* 2003; Ocak and Erener, 2005). The result with respect to feed efficiency does not confirm the argument that organic acids may stimulate digestibility, which can, in turn, improve feed efficiency (Ricke, 2003). This may firstly be explained by the fact that organic acids are capable of exhibiting bacteriostatic and bactericidal properties depending on the physiological status of the organisms and the physicochemical characteristics of the external environment (Ricke, 2003). On the other hand, antimicrobial agents may have more impact when the diet used is less digestible, and the diet contained highly digestible ingredients so that bacterial growth in the intestine probably may be limited (Lee *et al.,* 2003). Therefore, second explanation may be related to the digestibility of the basal diet used in the current study. In fact, it has been shown that dietary antibiotics appear to stimulate the activities of amylase and chymotrypsin in pancreatic tissue from broilers fed on wheat-soybean meal based diet (Engberg *et al.,* 2000). Attia *et al.* (2008) reported that Japanese quail hens can be fed diet containing 10% Nigella seed meal and moreover, this level can be increased to when supplemented with enzyme. The wheat and partially dehulled sunflower meal were used in diets offered in the present study, similarly to diets offered in the study of Enberg *et al.* (2000) or contrary to the diets offered in the study of Moharrery and Mahzonieh (2005). Therefore, the difference between the results of this study and the studies of Moharrery (2005) and Moharrery and Mahzonieh (2005) may be explained by differences in the inclusion level (highest 0.12% vs. 0.15%) and form (diet vs. water) of malic acid, in bird species (quail vs. broiler) and in the digestibility of the basal diet (probably low vs. high). It is cognoscible that some scientific articles on use of the organic acids in animal diets contain also such physiological data as: pH in different parts of alimentary tract (crop, gizzard, duodenum, jejunum, caecum), bacterial activity in intestine and results of balance trial (digestibility coefficients) and the physiological activity of malic acid inside of gut. Unfortunately, these physiological traits were not investigated in the present study.

In the present study, a significant benefit of the feeding diet with malic acid at 0.8 g/kg compared to the control diet was obtained in terms of carcass weight in Japanese quails. However, feeding of malic acid to quails with a view to increase carcass weight, because dressing percentage was decreased, though not significantly, in all the malic treatments, should be treated with caution. The results reported here for full gut weight and edible inner organs in agreement with results of the experiment 2 in the study of Moharrery and Mahzonieh (2005) who has reported that gut and liver weights of broilers were not affected by malic acid, contrary results of the experiment 1 in them study. Thus, the fact that no effect of malic acid treatments on the gut weight in both the present and previous study does not support the idea that the digestibility of the basal diet used in the current study was low. However, the higher feed consumption and gut length of quails fed the diet with 0.12% malic acid to compare with control (0% malic acid) probably explains why the digestibility of the basal diet used in the current study was low. This result indicates that nutritional value of diets used in the present study may be improved by malic acid supplementation by increasing feed intake. Increased relative length of gut may be attributed to adaptation to increased feed consumption.

It was expected that supplementing the antimicrobial (Lee *et al.,* 2003) or acidifier principles (Daskiran *et al.,* 2004) would stimulate growth performance in poultry. In the present study, beneficial effect of malic acid on body weight gain and carcass weight was detected during the growing phase of Japanese quails. On the other hand, the fact that mortality has not occurred in any of the groups during the experiment confirmed the ideas that malic acid is relatively non-toxic in acute toxicity studies using animals (Fiume, 2001) and that malic acid used as an acidifier in broiler rations had not an effect on mortality (Maheswari and Kadirvel, 1996). Therefore, it may be said that malic acid offers advantages in the growing performance due to its effects of carbohydrate on feed consumption (Dunaev *et al.,* 1988; Abraham and Flechas, 1992; Fiume, 2001), of antimicrobial in maintaining the gut health of quail (Ricke, 2003; Gowda *et al.,* 2004; Moharrery and Mahzonieh, 2005) and/or of acidifier in quail diet (Maheswari and Kadirvel, 1996; Daskiran *et al.,* 2004; Dhawale, 2005).

In conclusion, feeding diets with malic acid may increase the growth performance and feed consumption without any concomitant reduction or augmentation in the feed efficiency and mortality in Japanese quails. Furthermore, a significant benefit of feeding diet with malic acid may be gained in terms of carcass weight. Based on the first outcome, feeding diets with malic acid may increase the feed cost per unit weight gain of quail. Consequently, the final body weight should be determined according to the desired carcass weight, taking into account the differences between prices of diet and quail meat in markets. Further research is needed for better understanding of the role of the dietary malic acid in poultry performances and in toxins, colonization and proliferation of microorganisms in the poultry gastrointestinal tract.

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