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Effect of Supplementing Broilers Diets with Organic Acid and Whole Grain

[1]K. Çelik, [1]K. Uğur and [2]A. Uzatıcı
[1]Department of Animal Science,
Çanakkale Onsekiz Mart University, Çanakkale, 17020, Turkey
[2]Biga Vocational College, Çanakkale Onsekiz Mart University, Çanakkale, Turkey

Abstract: An experiment was conducted with 102, 2 day-old Ross 308, male broilers to investigate the effects of Organic Acid (OA) components and Whole Wheat (WG) on the growth performance and plasma parameters. There were 3 dietary treatments, each consisting of 34 replicates. The three dietary treatments were, (i) control, (ii) basal diet + 0.5% OA and (iii) basal diet + 25% whole wheat. The chicks were offered starter diet from 1 to 21 and grower diet from 22 to 42 as *ad libitum*. Feed intakes and body weight gains of the broilers were measured at the end of the each week during the experiment. Blood samples was collected from 13 birds in each group by branchial vein and analyzed for serum biochemical values and enzyme activities. There was no toxic effect of OA evidence by the absence of any dramatic change in relative organ weights or other telltale signs of serum clinical chemistry that would suggest liver or kidney dysfunction. The result indicated that dietary organic acids had no significant effects on feed intake, body weight gain, feed efficiency, body weight and carcass weight ($p<0.05$) compared to control but were significantly higher than WG. Whereas protein, albumin, glucose, BUN, cholesterol, ALP, ALT, Ca and P significantly different in OA group ($p<0.05$). The relative weights of the hot carcass and length of small intestines was statistically significant ($p<0.05$). Dietary whole grain had significant effects on feed intake, body weight and body weight gain ($p<0.05$). It is therefore concluded that 0.5% OA and 25% WG supplemented diets did not alter the performance and had no adverse effects on health of the broiler chickens.

Key words: Broiler, organic acid, whole grain, performance

INTRODUCTION

Antimicrobial feed additives are so far used world wide in animal husbandry to improve the economy and ecology of animal production by increasing growth rate, decreasing feed expenditure per gain and diminishing the risk of disease (Hays, 1981; Gropp *et al*., 1992). Recently antibiotic resistance entering the chemical drugs to human and animal food chain have been seriously considered. Several ways have been suggested as strategies to limit the antibiotic usage. Generally in poultry breeding, methods to improve efficiency of meat production must be developed as antibiotic use decreases. Any improvements which will serve as a replacement for antibiotics in animal feed, must enhance growth, improve feed efficiency, or decrease mortality at no additional cost to the consumer. Addition of prebiotics, probiotics, organic acids and whole grain to the poultry ration are few of these solutions (Chaveerach *et al*., 2004). Nowadays in poultry production organic acids have maiuly been used in order to improve animal performance. Organic acids are metabolic intermediates produced in pathways of central energy production, detoxification, neurotransmitter breakdown, or intestinal microbial activity as well. Several years ago some integrated poultry companies in the United Kingdom and The

Corresponding Author: K. Çelik, Department of Animal Science, Çanakkale Onsekiz Mart University, Çanakkale, 17020, Turkey

Netherlands have diluted broiler rations with up to 20% whole wheat and less commonly, poultry feed with up to 40% whole wheat to partially tolerance to industrial style animal raising. The claim is that feed processing and ingredient costs have been reduced with little or no effect on performance and improved bird health due to stimulation of the gizzard and drier litter conditions. The present study was conducted to evaluate the effects of OA components (0.5%) and WG (25%) on performance, GIT organs, liver, hearth and serum characteristics of broilers.

MATERIALS AND METHODS

Birds and Management

The animal experiment was carried out in accordance with the guide for care TIPDAM from Çukurova University. Two days old male broiler chicks (Ross 308) were supplied by a commercial hatchery (Banvit Turkey). The chicks were housed in an electrically heated battery brooder and fed water and a commercial starter diet (CP, 24%, 13.3 metabolizable energy (MJ kg^{-1}) *ad libitum* for the first 13 days. The experiment was conducted in a temperature-controlled room at 33°C for the first week and reduced to 3°C every week till 25°C. Relative humidity was kept at 50-70% throughout the experiment.

Experimental Diets and Feeding

A total of 102, 2-day-old Ross 308, male broiler chickens with an initial body weight of 41.5 g were used in a feeding experiment. The study design consisted of 3 treatments with 34 replicate in wired floor cages (102 cages) with 1 chicken in each cage. Corn-soybean-based broiler starter diet was prepared according to National Research Council (1994) specification. The three diets used were (i) Control, (ii) basal diet + 0.5% OA and (iii) basal diet + 25% whole wheat (WG). For the WG group, not only the extra crude-protein and energy of added wheat but also the crude protein and energy of feed which is given to both control and OA group were equally balanced by increasing the content amount (Rephrase). The chicks were fed with starter diet from 1 to 21, grower diet to 22 to 42 day *ad libitum*. Feed Intakes (FI), Body Weight Gains (BWG), Body Weights (BW) and Feed Conversion Ratios (FCR), of the broilers were measured at the end of the each week during the experiment. The OA component was obtained from TOPKIM which contains 17.4% formic acid, 14.1% ammonium formiate, 12.4% propionic acid, 8.4 and 47.7% wheat bran. During the experiment chickens from each group were monitored for mortality and morbidity. All chickens were slaughtered by cervical dislocation at the end of the experiment (42 day). Adjusted feed conversion was calculated by using total feed consumption minus the assumed feed consumption of the dead or removed chickens in a pen divided by the total growth of the surviving birds at the end of the study.

Mortalities

Infectious disease was not found during the experiment. Ascites (right ventricular hypertrophy) was the predominant cause of bird mortality. A small number of birds were culled because of leg abnormalities that occurred across each treatment. Bird mortality was 4, 0 and 2 for Control, WG and OA groups, respectively.

Organ Analysis

The intestinal package and heart was removed mechanically and the remaining carcass was weighed and the contents of the crop, gizzard, ileum and both large intestines were quantitatively collected before further analyses. The ileum was defined as the small intestinal segment caudal to Meckels diverticulum. After dissection, the liver and heart were rinsed in sterile water and weighed. In order to determine the weight of the filled and empty intestines were weighed with their contents and then rinsed in sterile water and weighed again.

Serum Chemistry Analysis

Prior to euthanasia, the birds on 42 day were weighed and the blood from 13 birds in each group was collected by branchial vein in Vacutainer tubes (BD Bioscience, Franklin Lakes, NJ) containing potassium-ethylene diamine tetra acetate (EDTA) for differential count or clot accelerator for serum clinical chemistry. Serum was obtained from blood samples and analyzed for serum albumin, total serum protein, gamma-glutamyltransferase (EC 2.3.2.2) and aspartate aminotransferases (E.C. 2.6.1.1) with a Technicon RAB1000 system (Miles Inc. Diagnostics Division, Tarrytown, NY) according to standard procedures, as described by Technicon-RA of Systems (1994). The differential count was done using a Cell-Dyn blood counter (Abbott Laboratories, Chicago, IL) and clinical chemistry with a Corning clinical chemistry analyzer (Chiron Corporation, San Jose, CA).

Statistical Analysis

All data were analyzed by GLM procedure using Duncan's multiple range test with SAS statistical software SAS (1996). Relative organ weight data were subjected to arcsine transformation, which showed a similar statistical trend. Differences were considered significant at ($p<0.05$).

RESULTS AND DISCUSSION

As Table 2 indicates FI is lower in WG group than control and OA groups. The same trend was observed for BW and BWG. OA group was better and heavier than others but there was no improvement in feed conversion due to OA supplementation. The results are not in agreement with Yeo and Kim (1997), Jin *et al.* (1998) and Rath *et al.* (2006) who did not find improved growth in the OA supplemental groups. It was also noted that feed conversion ratio was poorer for supplemental birds at 0 to 3 week than for control birds. Besides, WG group birds showed a significant decreases in FI, BW and BWG ($p<0.05$) than other groups in this investigation. FCR showed that figure was not statistically important in groups at the end of experiment ($p<0.05$). In this study significant decreases for FI, BW and BWG was found in WG group birds ($p<0.05$). On the contrary, Belyavin (1993)

Table 1: Composition of complete diets

Ingredients (g kg^{-1}) complete diet	Starter (1-21 day)	Grower (22-39 day)
Wheat	380.0	395.0
Maize	126.2	199.6
Soyabean meal	393.0	314.0
Rapeseed oil	53.0	46.0
Organic acid components	5.0	5.0
D-L-methionine	1.6	1.4
Calcium carbonate	13.2	11.5
Dicalcium monophosphate	18.0	17.5
NaCl	4.0	4.0
Mineral mixture[1]	1.0	1.0
Vitamin mixture[2]	5.0	5.0
Calculated analysis		
ME (MJ kg^{-1})	13.3	13.6
Crude protein (g)	240.6	210.6
Methionine + cystine	9.2	8.5
Lysine	13.3	11.0
Calcium	10.6	9.7
Available phosphorus	4.4	4.4

[1]The mineral mixture supplied (mg kg^{-1} diet): Co, 0.39; Cu, 8.2; I, 1.3; Se, 0.26; Fe, 44; Zn, 88; Mn, 101 and Ca, 216. [2]The vitamin mixture supplied the following vitamins (per kg diet): Vitamin A (all-trans-retinol), 20 mg; cholecalciferol, 3 mg; Vitamin E (d,l--tocopheryl acetate), 20 mg; butylated hydroxy toluene, 125 mg; menadione, 1.25 mg; thiamine, 0.5 mg; riboflavin, 3.5 mg; calcium pantothenate, 3.6 mg; niacin, 30 mg; pyridoxine, 1 mg; Vitamin B_{12} 8 g; folic acid, 1.5 mg; biotin, 0.25 mg; choline chloride, 700 mg. These vitamins were mixed with wheat bran (65% of vitamin mixture) before mixing vitamin mixture with other ingredients

Table 2: Performance traits of broilers throughout the experiment

Overall performance characters (0 to 6 week)	Control group	Whole grain group	Organic acid components
Feed intake (g/chick/day)	3442.61±55.21^a	3146.03±49.49^b	3432.06±37.46^a
Body weight	1818.73±35.96^a	1519.71±31.82^b	1829.41±28.14^a
Body weight gain (g/chick/day)	1358.40±15.0^a	1340.40±15.0^a	1366.40±15.0^a
Feed conversion ratio (feed:gain)	1.89±0.0167^a	2.07±0.0231^a	1.87±0.0159^a
Mortality	4 (3.92%)	0 (0.0%)	2 (1.92%)

Value are shown in $\bar{X}\pm S\bar{X}$, ^{a,b}Significant differences (p<0.05) between treatment means are indicated by different superscript letter(s)

Table 3: GI organs, liver and heart characteristics of 6-week-old chickens

Characters	Control	Whole grain	Organic acid components
Hot carcass yield (g)	1644.39±35.83^a	1321.67±27.27^b	1670.29±26.27^a
Gizzard (Filled) (g)	34.80±2.03^a	39.20±2.24^a	39.60±3.32^a
Gizzard (Empty) (g)	28.40±1.11^b	29.80±1.25^{ab}	34.20±2.39^a
Small intestine (Filled) (g)	56.60±1.71^a	56.80±2.62^a	57.60±2.57^a
Small intestine (Empty) (g)	42.20±1.9^a	42.00±1.74^a	41.60±1.97^a
Small Intestine length (cm)	180.80±4.08^a	164.40±2.7^b	169.80±4.2^a
Large intestine (g)	16.60±1.27^a	15.60±1.19^a	16.20±1.09^a
Large intestine length (cm)	8.80±0.63^a	9.40±0.43^a	8.00±0.42^a
Liver (g)	41.40±2.94^a	37.40±2.89^a	40.20±1.47^a
Heart (g)	11.20±0.68^a	10.40±0.4^a	11.40±0.79^a

Value are shown in $\bar{X}\pm S\bar{X}$, ^{a,b}Significant differences (p<0.05) between treatment means are indicated by different superscript letter(s)

Table 4: Effect of organic acid components (OA) on plasma chemistry of 6-week-old chickens (n = 13), mean±SEM

Treatment	Protein (g dL^{-1})	Albumin (g dL^{-1})	Glucose (mg dL^{-1})	BUN[1] (mg dL^{-1})	Cholesterol (mg dL^{-1})	ALP[2] (U L^{-1})	ALT[3] (U L^{-1})	Ca (mg dL^{-1})	P (mg dL^{-1})
Control	3.3±0.1^a	1.4±0.05^{ab}	202.8±5.3^a	1.2±0.1^a	98±02^a	748.3^a	8.7±0.3^a	11.7±0.3^a	6.3±033^a
25% WG	3.2±0.1^a	1.4±0.03^a	197.3±6.5^b	0.9±0.1^b	87±03^a	590.5^a	7.9±1.1^a	10.9±0.2^{ab}	6.2±037^a
0.5% OA	2.9±0.1^b	1.3±0.03^b	183.7±4.3^b	0.6±0.1^b	80±11^b	457.6^b	4.4±0.3^b	9.5±0.2^b	5.6±049^b

^{ab}Dissimilar scripts in a column denote significant differences p≤0.05, [1]BUN: Blood urea nitrogen, [2]ALP: Alkaline phosphatase, [3]ALT: Alanine amino transferase, WG: Whole Grain, OA: Organic Acid

reported reduced mortality in a large study when 150 g kg^{-1} of whole wheat was added as a supplement to grower and finisher feeds. Similarly we did not find mortality in WG supplement group. WG diets increased gizzard size but did not alter carcass yield. Feeding WG supplements caused at least a temporary loss in growth rate and feed efficiency but in some cases improved bird health. The results of carcas weights observed in this study are presented in Table 3 and carcass weights were affected by WG supplementation (p<0.05). There seems to be some evidence that the proportion of whole grain inclusion in the feed mix prior to pelleting of broiler diets leads to alteration of both gut development and other responses that contributed to better overall performance of broilers. Jones and Taylor (2001) has indicated that addition of 200 g kg^{-1} of whole grain into the mix prior to pelleting resulted in consistent differences in gut morphology, which may contribute to an improvement in productive performance by way of health benefits. Besides GI development of the birds was greater when they were fed the WG diet as gizzard weights increased by the incorporation of whole grain into the diet.

Results of the analyzed data for filled gizzard, filled and empty small-large intestines, length of large intestines were not statistically important among groups (p<0.05), however length of small intestines was statistically significant in WG group (p<0.05). The results of large intestine lengths show that birds on whole grain had longer large intestine while the intestine of those on AO group were shorter. Among groups no significant differences were found in liver and heart weight (p<0.05). Similar results was by Taylor and Jones (2004) that GI development of the birds was greater when they were fed the whole grain diet as gizzard weights were increased by the incorporation of WG into the diet. Similarly GI-intestinal development was thought to be related to the presence of large fibre particles in the feed in WG group birds. In this research statistical analyses indicated good results on performance factors for group OA birds. The reason for the observed might be due to dietary acidification that inhibited intestinal bacteria competing with the host for available nutrients and a reduction of possibly toxic bacterial metabolites, e.g., ammonia and amines, thus improving weight gain of the host animal. In an early study, Rahmani and Speer (2005) showed that dietary acidification increases gastric proteolysis and protein and amino acid digestibility. The OA has been shown to complex with Mg, Ca, P and Zn, which results in an improved digestibility of these minerals in this research.

Furthermore, the growth inhibition of potential pathogen bacteria and zoonotic bacteria, e. g., *E. coli* and Salmonella, in the feed and in the GI-tract are of benefit with respect to animal health (Mac Carthy, 2001). This research showed that chickens given OA birds even not statistically different were heavier than whole grain and control chickens. But BW, BWG and hot carcass findings in WG group was lower than control and OA given birds and this agree with Nir and Şenköylü (2000). Similar results has been reported by Huang *et al*. (2006) but, on the contrary they found that small intestines not longer statistically WG group birds. Besides, several studies have emphasized that increasing intestinal viscosity with corresponding reduction in physical potential of mixing diet in gut and low distribution of feed in gastrointestinal tract may effect on endogenous enzyme activity, changing microflora population and also renewal intestinal cells (Almiral *et al*., 1995; Bennett *et al*., 2002). Findings showed that OA affected all serum chemistry mentioned and values at high concentration in this research and there was a trend for decrease in protein, albumin, glucose, blood urea nitrogen, ALP, AST and Ca concentrations.

Feeding OA resulted in reduced levels of cholesterol and P concentrations and differences were significant at ($p<0.05$). A similar preliminary results from an experiment with broiler chickens (Klöcking, 1994) do also indicate that OA at concentrations of 0.2% may have a influence on reduction of the serum concentrations Ca and P due to a metal chelating effects of OA, which is affected by large number of carboxylic acid side chains. Although the decreased values were statistically different than control, they did not reflect any trend that would suggest any toxic effect of OA on kidney, heart, liver, intestines or muscles. The blood chemistry results were concordant with relative organ weight results, which showed no dystrophic eulargement or atrophy as it could happen under maladaptive conditions. This experiment appears to provide evidence that feeding of organic acid components (0.5%) to broiler chicks could improve growth performance and it does not have any adverse health effects on chickens while whole wheat (25%) feeding had no positive effect on the performance of birds.

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