

## IMPROVING BROILER PERFORMANCE AND FEED EFFICIENCY BY ADDING ORGANIC ACID TO BROILER DIETS.

Nagla K. Soliman<sup>1</sup>; Y.M. Alyoussef<sup>1</sup> and Nevine B.Ghanem<sup>2</sup>

<sup>1</sup> Animal Production and Fish, King Faisal University, Al Hasa, Saudi Arabia

<sup>2</sup> Food and Nutrition Science Department King Faisal University, Al Hasa, Saudi Arabia

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### SUMMARY

The current study aimed to evaluate the effect of dietary supplementation of 3 types of organic acids as citric, lactic, and propionic acid with the 3 different levels, 0.5%, 1.0% and 1.5% on the performance of broiler chicks. As well, the effect on some strains of intestinal bacteria, intestinal histomorphology, and serum biochemistry of were studied. Three hundred day-old Ross broiler chicks were divided into ten treated groups which were fed on control and nine organic acids – supplemented diets as 3 organic acids types by 3 levels for each acids. The chicks were placed in floor pens and fed on two basal diets as starter (22% protein, 3000 k Cal/kg) and grower (20% protein, 3000 k Cal/kg). Body weight gains of broiler chicks improved clearly due to inclusion different types of organic acids, especially at 1.5% propionic acid. Feed consumption values of chicks fed dietary organic acids were higher than control either cumulatively (0-6 weeks of age) or at different intervals (0-3, 3-6 weeks of age). Cumulative feed conversion ratios were improved with inclusion organic acids into broiler diets, the best improvement was recorded for the chicks fed 0.5% lactic acid or 1.5% propionic acid, however the differences lacked significance. The microbiological examination at the both stages of growth (3 .6 weeks of age) indicated that, bacterial total count per gram of ileal content significantly reduced by feeding different organic acids supplemented diets,. Total count of *E. coli* bacteria and Salmonella was inhibited significantly by including organic acids into broiler diets; propionic acid has the most effect on salmonella count. Lactobacilli bacteria increased significantly due to inclusion organic acids into broiler diets, this increment was huge and more pronounced at the second stage of growth. pH values of starter and finisher diets seemed to decline gradually as dietary concentration of organic acids increased. Also the pH values of ileal content were lowered as a result of organic acids supplementation. The mean values of serum cholesterol, triglycerides and total lipids did not differ due to organic acids supplementation. Both of serum total protein and globulins increased significantly, however serum albumin was not affected by treatments. Both of bursa and spleen weights increased significantly due to feeding organic acids supplemented diets.

**Keywords:** broiler chicks; performance; serum components; microbiological examination; organic acids.

### INTRODUCTION

The Efficiency of organic acids feed additives to modify the bacterial pattern of small intestine of poultry has been established two decades ago (Sheikh *et al* 2010, Müjdat *et al* 1999; El Afifi *et al* 2001). The antimicrobial activity of organic acids is pH dependent Galib and Aqeel (2009) due to its ability to pass through the cell membranes of bacteria, producing H<sup>+</sup> ions which lower the pH of bacterial cell, and reducing their growth of bacterial strains Nemcova (1997). Organic acids have an inhibitory effect against acid-intolerant species such as *E. coli*, Salmonella and Campylobacter. However, lactic acid beneficial bacteria are more resistant to organic acids and able to grow at relatively low pH, Paul *et al.* (2007). Several investigators have been proved that, organic acids supplementation into broiler diets improve their performance characteristics. El Afifi *et al* (2001) observed an improvement in broiler performance due to adding citric acids into diets. Similar results were reported by Izat *et al.* (1990) for propionic organic acid supplementation. (Vogt *et al.*, 1981 and Patten and Woldroup 1988) reported an improvement in body weight gain and efficiency of feed utilization of broiler chicks due to adding each of propionic, or lactic organic acids into their diets. The mode of action of organic acids in improving broiler performance is related mainly due to its inhibitory effect against harmful intestinal bacteria (Galib and Aqeel, 2009). In addition to increase the activity of protease enzymes and enhance protein utilization Giesting *et al.* (1991). Increase mineral absorption especially calcium (El Afifi and El Alaily, 2001). Increased intestinal villi length and keep the mucosal health (Sheikh *et al.* 2010). Several types of organic acids have been introduced into the middle east market either as a sole acid or a blend of two or more

organic acids as a growth promoter feed additives for poultry. However the comparative efficacy of these organic acids with different additive levels is still unclear. Therefore, the current study aimed to evaluate the effect of dietary supplementation of 3 types of organic acids as citric, lactic and propionic acid on the performance, intestinal bacteria, intestinal histomorphology, and serum biochemistry of the broiler chickens, as well the proper supplementation ratio will be studied.

## MATERIALS AND METHODS

This study was carried out at experimental poultry farm of animal production department, Faculty of Agriculture science and Nutrition King Faisal University.

The study was conducted to investigate the comparative efficacy of three different organic acids at different three levels. For this purpose three types of organic acids were used as citric acid, lactic acid and propionic acid. Each organic acids were added at the levels of 0.5%, 1.0%, 1.5%, of the experimental starter or grower diets. The experiment includes ten treatments as control group and nine treated groups for 3 types of organic acids with 3 levels each (3 organic acids x 3 levels of each acid).

### **Birds and their Management:**

Three hundred day-old Ross broiler chicks were used in this experiment. The birds were allocated randomly into ten groups of 30 birds which were divided into three replicates of 10 birds each. Basal diets were formulated (Table 1) to meet the nutrients requirements of broiler chicks during the starter (0-21 day) and grower (22-42 day) periods according to NRC (1994). Water and mash feed were provided ad lib. The chicks were placed in floor pens with wood-shavings litter. Electrical heaters were used for warming, Fan and air conditions were used for keeping suitable temperature. Artificial lighting was provided constantly. Body weights were recorded weekly for each chick and the average was calculated for each replicate and treatment group. Feed consumption values were recorded weekly in gram, and feed conversion ratio was calculated as gram feed /gram gain.

**Table (1): Composition and calculated analysis of experimental basal diets.**

Ingredients	Starter (0-3 wks)	Grower (3-6wks)
Yellow corn	55.8	59.82
Soybean meal (44%)	34.32	33.96
Corn gluten	3.33	-
Vegetable oil	2.79	2.79
Dicalcium phosphate	1.94	1.67
Limestone	1.14	1.14
Common salt	0.25	0.25
Vit. & min. premix*	0.25	0.25
DL. Methionine	0.18	0.12
Total	100	100
<i>Calculated composition</i>		
Crude protein%	22.00	20.00
M.E. Kcal/kg	3000	3000
% Calcium	0.97	0.91
%Available phosphorus	0.50	0.45
%Methionine + cystein	0.91	0.78
%Lysine	1.10	1.10

\*Composition of vitamin and mineral premix. Each 2.5 kg of vitamin and mineral mixture contains: 12000000 IU vitamin A; 2000000 IU D3; 10g E; 1g K; 1 g B1; 5g B2; 1500mg B6; 10mg B12; 10g Pantothenic acid; 20g Nicotinic acid; 1g Folic acid; 50mg Biotin; 500 g choline chloride; 4 g copper; 300 mg iodine; 30g iron; 60 g Manganese; 50g Zinc; and 100mg selenium

### **Slaughtered traits and samples collection:**

At the end of each experimental period either 3 or 6 week of age, six chicks per treatment group were slaughtered, allowed to bleed for blood sample, defeathered and eviscerated and internal organs were separated. Liver, spleen and bursa of fabricus weights were recorded. Ileal content samples were collected by pressing the outer wall of cut ileum to push its contents into clean sterile glass bottle. The pH values of diets and ileal contents were determined by mixing 2g of diet or ileal samples with 8ml distilled water and pH meter was used. Small intestine thickness was determined as the procedures described by Stutz *et al* (1983) and calculated as: *small intestine weight (g) / small intestine length (cm)*.

**Blood analysis:**

Blood samples of each experimental stage were collected and centrifuged at 4000 rpm for 15 minutes. Serum total protein was determined according to Biuret method Henery (1964) and albumin according to Doumas *et al.* (1971). Serum globulin was calculated by subtracting albumin from total protein. Serum total lipid was determined according to Knight *et al.* (1972) and total cholesterol according to Watson (1960).

**Cultivation and counting of bacteria:**

Microbiological examination procedure was done for the experimental period as follows: One gram of ileal content was adjustably weighed and transferred into test tube containing 9 ml of 0.1 sterile peptone. The samples were mixed well; tenfold dilutions were prepared and titrated on the following media:

Total aerobic bacteria were cultured on nutrient agar medium composed of (per liter) yeast extract 2.5g; trypton 5g, glucose 1g, agar 15g and distilled water up to one liter.

*Lactobacilli* bacteria were cultured on M.R.S. agar medium which is composed of casein peptone 10g, meat extract 10g, yeast extract 5g, glucose 20g, tween80 1g, K2mpo4 2g, sodium acetate 5g, diammonium citrate 2g., MnSO4 0.2g and distilled water up to 1 liter.

*E. coli* bacteria were cultured on MacChonkey agar medium that is composed of pancreatic digest of gelatin 17g pancreatic digest of casein 1.5g, peptic of animal tissue 1.5g, lactose 10g, bile salts 1.5g, sodium chloride 5g, neutral red 0.03g, crystal violet 0.001g, agar 3.5g, and distilled water up to 1 liter.

*Enterococcus* bacteria were cultured on MacChonkey agar No.2 medium that is composed of (peptone 20g, lactose 10g., bile salt 5g., sodium chloride 5.0g., neutral red 0.075g and agar, 12g per liter).

*Salmonella* bacterial were cultured on S.S. agar.

Bacterial count was determined by microscopic examination of the cultured media.

**Histological examination:**

Histological examination of slaughtered birds were done by using small pieces (2.5cm) from the same area of ileum for slaughtered birds per treatment either at three or six week of age. The ileum samples were placed in 10% buffered neutral formalin for fixation. A microtome was used to make 5µ sections that were mounted on glass slides and stained with hematoxylin and eosin. Villi length was measured from the apical to the basal region which corresponded to the superior portion of the crypts of Lieberkuhn by using light microscope and micrometer slide.

**Statistical analysis:**

Statistical analysis was carried out using statistical program SAS (1988). Duncan's multiple range test (1955) was applied a test for significant differences among means of traits. The following model was used:  $Y_{ij} = \mu + T_i + e_{ij}$

Where  $Y_{ij}$  = observation,  $\mu$  = overall means,  $T_i$  = effect of treatment and  $e_{ij}$  = experimental error.

## RESULTS AND DISCUSSION

Body weight gains of broiler chicks (Table2) for the entire experimental period 0-6 week improved clearly due to inclusion different types of organic acids into their diets, beyond that, the improvement was significant for the birds fed diet supplemented with 1.5% propionic acid. Similar trends were recorded for body weight gain of chicks at different intervals 0-3 week or 3-6 week of age. Feed consumption values of chicks fed dietary organic acids were higher than control either cumulatively (0-6 week) or at intervals (0-3, 3-6 week). Cumulative feed conversion ratios were improved with inclusion organic acids into broiler diets, this improvement was 11% for the chicks fed 0.5% lactic acid or 1.5% propionic acid however, the differences lacked significance.

The improvement in body weight gains (Table 2) coincide with those of El-Afifi *et al* (2001), Muzaffer *et al* (2003), and Sheikh *et al* (2010) who observed an enhancement in body weight of broiler chicks due to adding different organic acids into their diets. The increment in feed consumption values of the current experiment disagreed with the pervious findings of (El Afifi and El Alaily 2001), Furuse and Okumura (1989) showed that, feed intake of poultry can be depressed as a result of low palatability of acidic diets due to organic acid inclusion. The present result of feed conversion efficiency was in harmony with the finding of Alp *et al.* (1999) and Kahraman *et al.* (1999), who reported that feed conversion was generally improved by feeding dietary organic acids.

The mode of beneficial action of organic acids on broiler performance is related due to lowering pH values (Table5) in the gut (mainly upper intestinal tract), inhibiting the proliferation of unfavorable micro-organisms, (Table5) enhancing nutrients digestibility and absorbability by increasing enzyme activity and reducing the passing rate of digesta, Giesting *et al.* (1991), Waldroup *et al.*, (1995), Ghazalah, *et al.* (2011).

Serum constituents of broiler chicks (Table3) fed different levels of organic acid showed no significant differences in the concentration of serum cholesterol, triglycerides or total lipids among all the treatment groups either at 6 week or 3 week of age. Serum constituents of protein and its derivatives at 3 or 6 week of chicks age showed, a significant increments in both of total protein and globulins, however serum albumin was not affected.

The elevation in blood total protein (Table 3) may be related to stimulating effect of organic acids on protease enzymes and enhancing protein absorption (Giesting *et al.* 1991). The increment in serum globulin concentration may suggest tendency to enhance humoral immunity of chicks due to feeding organic acids, because of a part of globulin is responsible for producing immune bodies. The improvement in immunity may be related to the effect of organic acids in increasing the populations of lactobacilli bacteria which would enhance the natural immune competence ability (Lan *et al.* 2005). The blood cholesterol results are in harmony with those obtained by Sheikh *et al.* (2010) who did not observe any effect on blood cholesterol due to supplementing organic acids into broiler diets.

Bursa and spleen weights (Table4) were increased due to feeding organic acids supplemented diets at 6 week of age; however the increment was not significant for spleen weight. Absolute liver weight increased significantly at 3 and 6 week of age due to feeding dietary organic acids.

Small intestine thickness (Table4) reduced significantly due to feeding dietary organic acids at 3 weeks of age; however this reduction was not clear and lacked significant at 6 week of age. Villi heights ( $\mu$  m) of ileal section (Table 4 & Fig 1), were significantly higher in birds fed organic acids supplemented diets than control. pH values (Table5) of starter and finisher diets seemed to decline gradually as dietary concentration of organic acids increased. Also the pH values of ileal content at both stages of experiment were lowered as a result of organic acids supplementation.

The increment in bursa and spleen weight (Table4) prove that addition of organic acid into broiler diets may enhance immunity. El Afifi (2003) found that, feeding citric acid enriched diets cause an increment in bursa and spleen weight. Dafwang *et al.* (1985) proved that, increase of bursa and spleen weight may reflect an improvement in chicks immunity due to feeding anti microbial agents. The reduction in small intestine thickness (Table4) may be related to the great reduction in the count of undesirable bacteria (Table5) which may induce a chronic inflammation resulting in intestinal wall thickening (Krink and Jamroz, 1996).

The significant increase in villi length (Table4) are in harmony with the earlier workers of L. oddi *et al.* (2004) and Sheikh *et al.* (2010) who reported that, the length of intestinal villi of broiler chicks fed organic acidifiers diets were greater than those of control group. They attributed the increment to the fact that organic acids reduce the growth of many pathogenic or nonpathogenic intestinal bacteria, decreasing the intestinal colonization and infectious processes, ultimately decreasing the inflammatory reactions at the intestinal mucosa, which increases the villis height and functions of secretion, digestion, and absorption of nutrients by the mucosa, finally, enhance broiler performance. However, this result disagrees with those of Gunal *et al.* (2006) and Leeson *et al.* (2005), who demonstrated that addition of organic acid to the broilers diet was ineffective on villi height.

The microbiological examination showed that (Table 5), bacterial total count per gram of ileal content of chicks significantly reduced by feeding different organic acids supplemented diets, at the both stages of growth. Total count of *E. coli* (gram-negative) bacteria was inhibited significantly and sharply by feeding organic acids. The ileal content of Salmonella reduced significantly by including organic acids into broiler diets. Propionic acid has the most effect on salmonella count. In contrast Lactobacilli bacteria increased significantly due to inclusion organic acids into broiler diets, this increment was huge and more pronounced at the second stage of growth.

The reduction in pH values (Table5) of ileal content were recorded by Burnell *et al.* (1988) and El Afifi *et al.* (2001) who attributed this reduction due to lowering dietary pH resulting from organic acids addition.

The inhibitory effect of organic acids against gut microflora was reported by Jin *et al.* (1997) Waldroup *et al.* (1995) that the antimicrobial activity of organic acid may be resulted from decrease pH value, in addition to specific antimicrobial effects of the enter undissociated acid molecules into microbial cells and dissociate into anion and protons which decrease the pH of cytoplasm.

Thus inhibiting synthesis of several macromolecules, including cell wall components, DNA, lipids, proteins and RNA. Mújdat *et al.* (1999) and Waldroup *et al.* (1995) reported that, the sharp reduction in the count of *E. Coli* bacteria may be attributed to the direct effect of organic acids in lowering ileum pH value.

Table (2): Effect of different levels of organic acids on Body weight feed consumption and Feed conversion efficiency.

Item	Body gain (g)			Feed intake (g/bird)			Feed Efficiency (g feed/g gain)		
	(0-3 wk)	(3-6 wk)	(0-6 wk)	(0-3 wk)	(3-6 wk)	(0-6 wk)	(0-3 wk)	(3-6 wk)	(0-6 wk)
Citric acid (0.5%)	417.3	1085.3 <sup>abc</sup>	1502.9 <sup>abc</sup>	774.5 <sup>bcd</sup>	2503.5 <sup>bc</sup>	3278.0 <sup>ab</sup>	1.94	2.32	2.18
	4.88	6.58	7.57	19.51	96.62	115.91	0.306	0.189	0.090
Citric acid (1%)	395.4	1032.8 <sup>abc</sup>	1427.7 <sup>abc</sup>	749.1 <sup>d</sup>	2460.6 <sup>cd</sup>	3209.7 <sup>bc</sup>	1.98	2.39	2.55
	3.58	7.57	7.59	9.15	32.96	24.32	0.296	0.126	0.107
Citric acid (1.5%)	418.8	1024.4 <sup>abc</sup>	1443.2 <sup>abc</sup>	799.3 <sup>b</sup>	2377.7 <sup>cd</sup>	3176.9 <sup>bc</sup>	1.98	2.39	2.21
	4.34	5.67	7.89	14.38	56.94	43.15	0.296	0.179	0.113
Lactic acid (0.5%)	432.7	1079.8 <sup>abc</sup>	1502.5 <sup>abc</sup>	844.3 <sup>a</sup>	2301.4 <sup>d</sup>	3145.7 <sup>bc</sup>	2.07	2.14	2.10
	3.98	6.44	8.33	4.29	30.62	26.83	0.301	0.134	0.095
Lactic acid (1%)	377.6	1068.6 <sup>abc</sup>	1446.2 <sup>abc</sup>	805.9 <sup>b</sup>	2456.7 <sup>cd</sup>	3262.7 <sup>b</sup>	2.24	2.31	2.26
	2.34	4.79	7.15	6.21	53.19	46.99	0.360	0.115	0.111
Lactic acid (1.5%)	370.1	1016.4 <sup>bc</sup>	1386.5 <sup>bc</sup>	758.4 <sup>cd</sup>	2292.7 <sup>d</sup>	3051.1 <sup>c</sup>	2.17	2.28	2.20
	4.21	5.76	8.96	19.02	106.30	125.29	0.397	0.223	0.081
Propionic acid (0.5%)	392.5	1082.8 <sup>abc</sup>	1475.2 <sup>abc</sup>	811.1 <sup>b</sup>	2450.1 <sup>cd</sup>	3261.2 <sup>b</sup>	2.16	2.27	2.22
	1.48	3.60	5.69	1.74	54.27	52.54	0.324	0.109	0.077
Propionic acid (1%)	373.9	1191.1 <sup>ab</sup>	1564.9 <sup>ab</sup>	752.3 <sup>d</sup>	2716.8 <sup>a</sup>	3469.1 <sup>a</sup>	2.11	2.29	2.22
	2.50	3.89	4.91	3.57	37.08	33.67	0.335	0.100	0.098
Propionic acid (1.5%)	392.2	1215.2 <sup>a</sup>	1622.7 <sup>a</sup>	792.1 <sup>bc</sup>	2675.7 <sup>ab</sup>	3467.7 <sup>a</sup>	2.12	2.12	2.12
	3.51	5.02	7.48	3.27	37.87	38.67	0.322	0.107	0.076
Control	389.9	978.2 <sup>c</sup>	1368.0 <sup>c</sup>	775.0 <sup>bcd</sup>	2420.2 <sup>cd</sup>	3195.3 <sup>bc</sup>	2.09	2.49	2.34
	3.42	6.39	9.27	11.93	3.14	11.00	0.341	0.145	0.102

a, b Mean values in a column without a common superscript are different ( $P \leq 0.05$ ).

The value  $\pm$  stander error

Table (3): Effect of different organic acids levels on some blood constituents of broiler chicks.

Item	Globulin		Albumin		Total protein		Triglyceride		Total Lipid		Cholesterol	
	3 wks	6 wks	3 wks	6 wks	3 wks	6 wks	3 wks	6 wks	3 wks	6wks	3 wks	6 wks
Citric acid (0.5%)	1.74	1.34	1.71 <sup>bc</sup>	1.81 <sup>b</sup>	3.45 <sup>a</sup>	3.15 <sup>abc</sup>	75.9	79.9	124.4 <sup>ab</sup>	150.2	135.8	124.4
			0.04	0.06	0.10	0.12	3.57	2.54	10.22	14.78	±15.86	±4.39
Citric acid (1%)	1.74	1.36	1.69 <sup>c</sup>	2.02 <sup>ab</sup>	3.43 <sup>a</sup>	3.38 <sup>abc</sup>	77.6	82.1	125.4 <sup>ah</sup>	159.9	138.3	129.1
			0.03	0.04	0.09	0.31	2.97	4.22	7.22	9.15	±12.26	±8.86
Citric acid (1.5%)	1.02	1.31	1.75 <sup>bc</sup>	1.85 <sup>ab</sup>	2.77 <sup>b</sup>	3.16 <sup>abc</sup>	75.3	93.2	136.8 <sup>ab</sup>	141.4	129.3	133.9
			0.07	0.08	0.15	0.12	6.26	10.19	11.27	11.0	5.63	7.03
Lactic acid (0.5%)	0.85	1.49	1.83 <sup>abc</sup>	1.92 <sup>ab</sup>	2.68 <sup>b</sup>	3.41 <sup>ab</sup>	74.1	81.6	139.2 <sup>a</sup>	146.4	140.0	126.3
			0.02	0.09	0.05	0.09	3.99	5.86	11.56	13.3	7.23	9.92
Lactic acid (1%)	1.02	1.75	1.74 <sup>bc</sup>	1.83 <sup>ab</sup>	2.76 <sup>b</sup>	3.58 <sup>a</sup>	69.9	83.7	108.9 <sup>b</sup>	163.9	149.1	112.2
			0.04	0.11	0.09	0.19	4.17	6.49	4.31	2.49	7.74	4.24
Lactic acid (1.5%)	0.81	1.41	1.90 <sup>abc</sup>	1.80 <sup>b</sup>	2.71 <sup>b</sup>	3.21 <sup>abc</sup>	76.69	79.0	135.3 <sup>ab</sup>	177.3	140.9	117.9
			0.07	0.05	0.15	0.14	3.03	5.41	9.79	7.37	8.09	5.93
Propionic acid (0.5%)	0.97	1.41	1.89 <sup>abc</sup>	1.86 <sup>ab</sup>	2.86 <sup>b</sup>	3.27 <sup>abc</sup>	76.21	77.2	138.3 <sup>ab</sup>	175.9	127.6	118.1
			0.04	0.11	0.13	0.11	3.33	8.37	12.04	36.16	11.85	4.97
Propionic acid (1%)	0.87	1.42	2.00 <sup>a</sup>	1.86 <sup>ab</sup>	2.87 <sup>b</sup>	3.28 <sup>abc</sup>	69.11	74.74	123.9 <sup>ab</sup>	156.4	129.7	121.1
			0.07	0.10	0.11	0.07	2.21	8.04	14.93	7.86	4.94	13.31
Propionic acid (1.5%)	0.82	0.81	1.93 <sup>ab</sup>	2.08 <sup>a</sup>	2.75 <sup>b</sup>	2.89 <sup>c</sup>	69.85	76.6	145.2 <sup>a</sup>	163.1	127.2	120.6
			0.07	0.4	0.09	0.13	1.08	3.31	12.52	9.23	8.56	3.95
Control	0.72	0.85	2.04 <sup>a</sup>	2.06 <sup>ab</sup>	2.76 <sup>b</sup>	2.91 <sup>bc</sup>	73.27	81.9	157.5 <sup>a</sup>	173.9	124.9	124.5
			0.07	0.07	0.03	0.06	2.97	1.13	6.67	4.98	3.34	3.39

a, b Mean values in a column without a common superscript are different ( $P \leq 0.05$ ).

Table (4): Effect of different organic acids levels on liver, intestinal thickness and some immunity organs.

Treatments/Item	Villi hights		Intest. Thickness		Bursa		spleen		liver	
	3 wks	6 wks	3 wks	6 wks	3 wks	6 wks	3 wks	6wks	3 wks	6 wks
Citric acid (0.5%)	36.13 <sup>d</sup>	37.29 <sup>dc</sup>	0.069 <sup>b</sup>	0.128	0.381 <sup>ab</sup>	1.50 <sup>a</sup>	0.451	2.04	9.50 <sup>ab</sup>	31.68 <sup>ab</sup>
	0.989	1.17	0.006	0.007	0.06	0.354	0.07	0.163	0.36	2.57
Citric acid (1%)	63.38 <sup>b</sup>	64.25 <sup>ab</sup>	0.078 <sup>b</sup>	0.121	0.514 <sup>ab</sup>	0.940 <sup>b</sup>	0.326	1.97	9.79 <sup>ab</sup>	30.62 <sup>ab</sup>
	3.66	3.65	0.005	0.007	0.12	0.218	0.060	0.275	0.445	1.670
Citric acid (1.5%)	72.44 <sup>a</sup>	66.71 <sup>a</sup>	0.085 <sup>b</sup>	0.208	0.272 <sup>b</sup>	0.860 <sup>b</sup>	0.304	2.44	9.59 <sup>ab</sup>	33.30 <sup>ab</sup>
	2.00	5.49	0.010	0.012	0.05	0.244	0.05	0.244	0.86	1.92
Lactic acid (0.5%)	64.71 <sup>b</sup>	60.86 <sup>ab</sup>	0.103 <sup>b</sup>	0.255	0.631 <sup>a</sup>	0.890 <sup>b</sup>	0.398	1.84	9.57 <sup>ab</sup>	32.66 <sup>ab</sup>
	4.66	5.19	0.002	0.017	0.12	0.402	0.06	0.250	0.275	3.06
Lactic acid (1%)	64.00 <sup>b</sup>	62.89 <sup>ab</sup>	0.091 <sup>b</sup>	0.212	0.338 <sup>b</sup>	0.868 <sup>b</sup>	0.326	1.94	9.25 <sup>ab</sup>	32.26 <sup>ab</sup>
	2.89	2.47	0.007	0.019	0.09	0.075	0.04	0.250	0.613	1.82
Lactic acid (1.5%)	61.30 <sup>b</sup>	63.78 <sup>ab</sup>	0.090 <sup>b</sup>	0.221	0.294 <sup>b</sup>	0.766 <sup>b</sup>	0.514	1.98	9.66 <sup>ab</sup>	32.28 <sup>ab</sup>
	3.22	2.44	0.009	0.029	0.05	0.097	0.17	0.049	1.906	1.33
Propionic acid (0.5%)	36.50 <sup>d</sup>	44.13 <sup>d</sup>	0.096 <sup>b</sup>	0.127	0.474 <sup>ab</sup>	0.760 <sup>b</sup>	0.452	1.82	10.67 <sup>a</sup>	34.70 <sup>ab</sup>
	0.87	1.16	0.005	0.006	0.06	0.093	0.04	0.195	0.773	1.55
Propionic acid (1%)	50.00 <sup>c</sup>	48.83 <sup>cd</sup>	0.081 <sup>b</sup>	0.321	0.287 <sup>b</sup>	0.668 <sup>b</sup>	0.362	1.68	9.09 <sup>ab</sup>	36.96 <sup>a</sup>
	0.52	1.72	0.005	0.075	0.05	0.049	0.06	0.344	0.786	2.86
Propionic acid (1.5%)	58.43 <sup>b</sup>	55.14 <sup>bc</sup>	0.380 <sup>a</sup>	0.247	0.418 <sup>ab</sup>	0.756 <sup>b</sup>	0.456	2.03	10.93 <sup>a</sup>	34.55 <sup>ab</sup>
	1.34	1.88	0.276	0.005	0.05	0.045	0.05	0.18	1.079	1.87
Control	33.10 <sup>d</sup>	34.50 <sup>c</sup>	0.089 <sup>b</sup>	0.225	0.399 <sup>ab</sup>	0.610 <sup>b</sup>	0.372	1.64	7.10 <sup>b</sup>	28.40 <sup>b</sup>
	1.05	3.86	0.009	0.022	0.12	0.058	0.12	0.20	1.834	2.33

a, b Mean values in a column without a common superscript are different ( $P \leq 0.05$ ).

Fig. (1): Height of Intestinal villi of the small intestine in 3 & 6 weeks age with different acids:

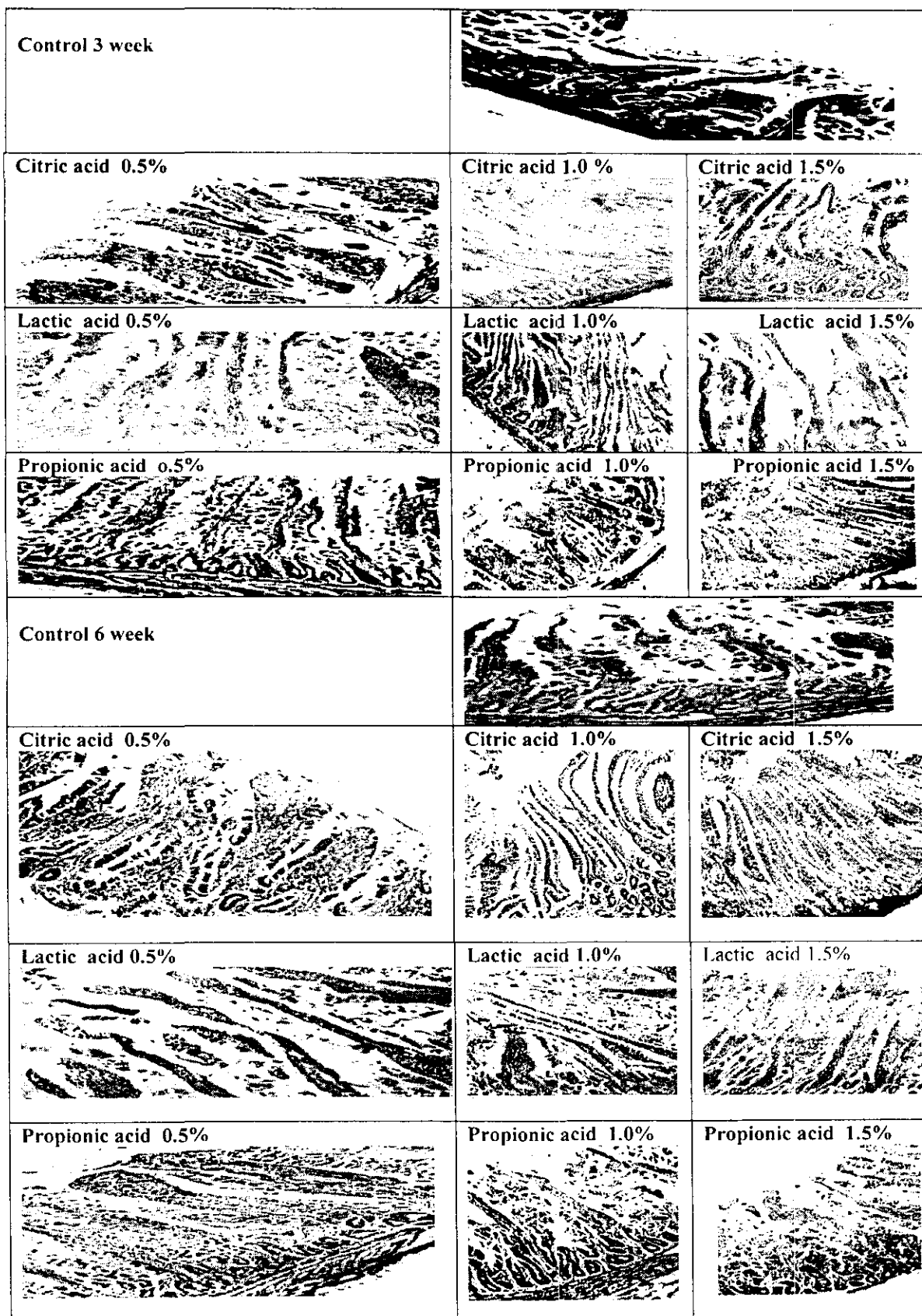




Table (5): Effect of Different organic acids levels on Ileal pH and intestinal bacteria account.

Item	Treatments									
	Propioic Acid			Lactic Acid			Citric Acid			Control
	1.5%	1.0%	0.5%	1.5%	1.0%	0.5%	1.5%	1.0%	0.5%	
<i>3 week Age</i>										
Bacterial total count x 10 <sup>6</sup>	26 <sup>bc</sup>	22.6 <sup>bc</sup>	13.6 <sup>bc</sup>	13.3 <sup>c</sup>	27.1 <sup>b</sup>	16.4 <sup>bc</sup>	22.3 <sup>bc</sup>	14.8 <sup>bc</sup>	16.0 <sup>bc</sup>	52.3 <sup>a</sup>
E. Coli count x 10 <sup>4</sup>	1.73	2.96	1.93	0.88	3.59	3.39	7.36	1.30	4.52	6.74
Salmonella count x 10 <sup>2</sup>	16.3 <sup>bc</sup>	23.7 <sup>bc</sup>	20.7 <sup>bc</sup>	19.7 <sup>cb</sup>	17.7 <sup>bc</sup>	14.3 <sup>cd</sup>	12.7 <sup>cd</sup>	26.3 <sup>b</sup>	4.6 <sup>d</sup>	38.7 <sup>a</sup>
Lactobacilli Sp. count x 10 <sup>5</sup>	0.88	3.67	0.88	2.19	1.67	1.20	1.45	4.06	0.31	8.76
Ileal content pH	2.6 <sup>b</sup>	3.5 <sup>ab</sup>	2.7 <sup>b</sup>	1.7 <sup>b</sup>	3.7 <sup>ab</sup>	2.0 <sup>b</sup>	3.7 <sup>ab</sup>	2.0 <sup>b</sup>	2.5 <sup>b</sup>	6.4 <sup>a</sup>
Dietary pH	1.76	0.76	1.45	0.88	0.33	0.58	0.67	1.15	0.35	0.88
	27 <sup>bc</sup>	62.3 <sup>a</sup>	33.3 <sup>bc</sup>	22 <sup>bc</sup>	22.3 <sup>bc</sup>	23.3 <sup>bc</sup>	11.1 <sup>bc</sup>	13.5 <sup>bc</sup>	14.7 <sup>bc</sup>	6.3 <sup>c</sup>
	14.57	14.68	8.82	4.14	4.63	1.20	2.52	0.74	2.66	1.86
	5.76 <sup>ab</sup>	5.86 <sup>ab</sup>	5.84 <sup>ab</sup>	5.84 <sup>ab</sup>	5.74 <sup>ab</sup>	5.22 <sup>b</sup>	5.78 <sup>ab</sup>	5.14 <sup>b</sup>	5.44 <sup>b</sup>	6.29 <sup>a</sup>
	0.32	0.18	0.20	0.17	0.13	0.20	0.36	0.27	0.22	0.12
	4.22 <sup>d</sup>	5.21 <sup>d</sup>	5.16 <sup>c</sup>	4.89 <sup>c</sup>	5.09 <sup>d</sup>	5.40 <sup>c</sup>	4.61 <sup>g</sup>	4.86 <sup>f</sup>	5.06 <sup>d</sup>	5.90 <sup>a</sup>
	0.03	0	0.04	0.03	0.03	0.04	0	0.25	0.05	0
<i>6 week Age</i>										
Bacterial total count x 10 <sup>6</sup>	50 <sup>bcd</sup>	55 <sup>bcd</sup>	32 <sup>d</sup>	51 <sup>bcd</sup>	42 <sup>bcd</sup>	35.3 <sup>cd</sup>	43.3 <sup>bcd</sup>	58 <sup>bc</sup>	61.3 <sup>b</sup>	93.6 <sup>a</sup>
E. Coli count x 10 <sup>4</sup>	5.77	10.41	3.51	5.57	4.93	4.91	5.21	7.57	6.33	11.97
Salmonella count x 10 <sup>2</sup>	6 <sup>b</sup>	12.6 <sup>b</sup>	15.3 <sup>b</sup>	7.3 <sup>b</sup>	9 <sup>b</sup>	12.4	16.3 <sup>b</sup>	11.7 <sup>b</sup>	12.9 <sup>b</sup>	68 <sup>a</sup>
Lactobacilli Sp. count x 10 <sup>5</sup>	1.53	1.45	3.53	2.85	2.08	6.98	2.03	3.20	2.03	6.43
Ileal content pH	ND <sup>c</sup>	ND <sup>c</sup>	ND <sup>c</sup>	2.0 <sup>bc</sup>	2.0 <sup>bc</sup>	ND <sup>c</sup>	2.3 <sup>bc</sup>	3.6 <sup>b</sup>	ND <sup>c</sup>	12.0 <sup>a</sup>
Dietary pH	1.0	1.00	1.0	1.00	1.00	1.20	1.45	1.45	1.73	1.73
	47.6 <sup>bc</sup>	56.6 <sup>abc</sup>	51.3 <sup>abc</sup>	46.3 <sup>bc</sup>	47 <sup>bc</sup>	86.6 <sup>a</sup>	73.3 <sup>ab</sup>	56.3 <sup>abc</sup>	81.6 <sup>ab</sup>	23.3 <sup>c</sup>
	4.33	1.67	5.93	6.33	4.93	24.04	17.64	13.57	10.14	1.76
	5.15 <sup>ab</sup>	5.13 <sup>ab</sup>	5.20 <sup>ab</sup>	4.75 <sup>b</sup>	5.13 <sup>ab</sup>	5.35 <sup>ab</sup>	5.03 <sup>ab</sup>	5.15 <sup>ab</sup>	5.60 <sup>a</sup>	5.58 <sup>a</sup>
	0.24	0.20	0.28	0.16	0.13	0.10	0.16	0.25	0.17	0.24
	4.98 <sup>d</sup>	4.90 <sup>d</sup>	5.20 <sup>b</sup>	4.73 <sup>c</sup>	4.93 <sup>d</sup>	5.10 <sup>c</sup>	4.40 <sup>g</sup>	4.63 <sup>f</sup>	4.98 <sup>d</sup>	5.70 <sup>a</sup>
	0.03	0	0.04	0.03	0.03	0.04	0	0.25	0.05	0

ND Non detected

a-b within rows, means with no common superscripts differ significantly ( $p \geq 0.05$ )

In addition to the indirect effect of low pH in promoting the growth of lactobacilli bacteria, which have antagonistic action against E. coli bacteria. Jin *et al.* (1997) and Nemcova (1997) stated that, increase lactobacilli causing additional inhibitory effect against coliforms bacteria via secreting bacterocin-like substance termed lactocidine which displays inhibitory activity against coliforms bacteria. As well, Lactobacilli compete with coliforms for sites of attachment which is needed for proliferation and reducing rate of removal of organisms due to movement of digesta. Waldroup *et al.* (1995) observed a reduction number Salmonella due adding propionic acid-containing product into broiler chick diets. The reduction in salmonella counts was more pronounced at the second stage of growth (6weeks of age) than the first stage (3weeks of age). That may be related to the huge increments in lactobacilli bacteria in the second stage compared with first stage of growth (Table5). Terada *et al.* (1994) showed that, Lactobacilli bacteria compete with Salmonella for colonization sites and causing a production of volatile fatty acids which are toxic for salmonella. Fuller (1977) and Waldroup *et al.* (1995) demonstrated that, the slight acidic media created from organic acids addition is responsible for increasing lactobacilli bacterial count. Mathew *et al.* (1991) showed that, lactobacilli bacterial count was not affected by dietary propionic acid of 8 weeks-old piglets and increase at 12 weeks of piglets age.

## REFERENCES

- Alp, M., N. Kocabagli, R. Kahraman and K. Bostan (1999). Effects of dietary supplementation with organic acids and zing bacitracin on ileal microflora, pH and performance in broilers. VIV Poultry Yutav'99 Istanbul S: 496-504.
- Burnell, T.W., G.L. Cromwell and T.S. Stahly. (1988). Effect of dried whey and copper sulfate on the growth response to organic acid in diets for weanling pigs. Journal Animal Science 66: 1100-1108.

- Dafwang, I.Z., Cook, M.E., Sunde, M.L. and Bird, H. Bursal (1985) intestinal and spleen weight and antibody response of chicks fed subtherapeutic levels of dietary antibiotics. , Poultry Science, 64: 634-639.
- Doumas, B.; W. Watson and H. Biggs (1971). Albumin standard and measurements of serum albumin with bromocresol green. *Clinical Chemistry Act* 31: 87 – 88.
- El Afifi, Sh. F and H.A. El Alaily (2001). Effect of citric acid supplementation to diets on performance, calcium retention and eggshell quality of aged layers. *Egyptian journal of nutrition and feed (special issue)*, 25-38.
- El Afifi, Sh. F., N. M. Elmedney and Attia (2001). Effect of citric acid supplementation in broiler diets on performance and intestinal microflora. *Egypt. Poultry Science* 21 (2): 491-505.
- El-Afifi, Sh. F., (2003) Comparative studies on garlic bulb, verginiamycin and citric acid as growth promoters in broiler chicks diet. *Journal of Agriculture Science. Mansoura University*, 28 (5); 3371 – 3381.
- Fuller R. (1977). The importance of lactobacilli in maintaining normal microbial balance in the crop. *British Poultry Science*, 18: 85-94.
- Furuse M. and J. Okumura. (1989) Effect of dietary acetic acid levels on protein and energy utilization in chicks. *Poultry Science* 68: 795-798.
- Galib A. Al-Kassi and Aqeel M. Mohssen (2009). Comparative Study Between Single Organic Acid Effect and Synergistic Organic Acid Effect on Broiler Performance. *Pakistan Journal of Nutrition* 8 (6): 896-899.
- Duncan, D.B. (1955). Multiple range and Multiple F tests. *Biometrics*, 11: 1-42.
- Ghazalah, A.A., Atta, A.M., El Moustafa, Kout Elkloub and F.H. Shata (2011). Effect of dietary supplementation of organic acids on performance, nutrients digestability and health of broiler chicks. *International Journal Poultry Science* 10 (3): 176-184.
- Giesting, W.D., A.M. Roose and A. R. Ester. (1991). Evaluation of the effect of Fumaric acid and sodium bicarbonate addition on performance of starter pigs fed diets of different types. *Journal of Animal Science* 69: 2489-2496.
- Gunal M., G. Yayli and O. Kaya (2006).The effect of antibiotic growth promoter, probiotic or organic acid supplementation on performance, intestinal microflora and tissue of broilers. *International Journal of Poultry Science* 5:149-155.
- Henery R.J.A. (1964). Calorimetric method for the determination of the total protein. *Clinical Chemical*, Harper and Row Publisher, New York.
- Izat, A.L., Tidwell, N.M., Thomas, R.A., Reiber, M.A., Adams, M.H., Colberg, M., Waldroup, P.W. (1990). Effects of a buffered propionic acid in diets on the performance of broiler chickens and on microflora of the intestine and carcass. *Poultry Science* 69:818-826.
- Jin, L.Z., Y.W Ho, N. Abdullah and S. Jalaludin.(1997). Probiotics in poultry: modes of action. *World's Poultry Science Association* 53: 355-368.
- Kahraman, R., I. Abas, K. Bostan, M. A. Tanör, N. Kocabagli and M. Alp (1999). Effects of organic acids and yeast culture on performance ileum pH and Enterobacteriaceae population of broilers. *VIV Poultry Yutav'99 Istanbul S*: 515-522.
- Knight, J., A. Andersonis and J. M. Rawal (1972). Chemical basis of the sula-phosphate vanillin reaction for estimating total serum lipid. *Journal of Biology and Chemistry*, 226-497.
- Krinke, A.L. and D. Jamroz (1996). Effect of feed antibiotic avoparcine on organ morphology in broiler chickens. *Poultry Science* 75: 705-710.
- Lan, Y., M.W.A. Versteegen, S. Tamminga and B.A. Willams (2005). The role of commensal gut microbial community in broiler chickens. *World's Poultry Science Association* 61: 95-104.
- Leeson S, Namkung H, Antongiovanni M, Lee EH. (2005). Effect of butyric acid on the performance and carcass yield of broiler chickens. *Poultry Science* 84: 1418-1422.
- Loddi, M.M.V., M.B. Maraes, I.S.O. Nakaghi, F. Tucci, M.I. Hannasand J.A. Arika (2004). Mannan oligosaccharide and organic acids on performance and intestinal morphometric characteristics of broiler chickens. In *Proceedings of the 20<sup>th</sup> Veterinary Medicine International 7 Annual Symposium on Computational Geometry*, supplement 1, p. 45, Brooklyn, NY, USA.
- Mathew, A.G., Sutton, A.L., Scheidt, A., Bforysyth, D.M., Patterson, J.A. and D.T. Kelly (1991). Effects of a propionic acid containing feed additive on performance and intestinal microbial fermentation of teh weanling pig. In: *Proceedings of the V<sup>th</sup> International Symposium on Digestive Physiology in pigs*. Wageningen, Netherlands, 24-26 April 1991. Eaap Publication No. 54, pp.464-469.
- Müjdat A., N. Kocabagli, R., Kahraman and K., Bostan (1999). Effects of Dietary Supplementation with Organic Acids and Zinc Bacitracin on Ileal Microflora, pH and Performance in Broilers. *Turky. Journal of Veterinary and Animal Sciences* 23: 451–455.

- Muzaffer D., F. Okan and K. Çelik. (2003). Effect of Dietary Probiotic, Organic Acid and Antibiotic Supplementation to Diets on Broiler Performance and Carcass Yield. *Pakistan Journal of Nutrition* 2 (2): 89-91.
- National Research Council (1994) Composition of poultry feed stuffs. Nat Acad. Sci., Washington, D. C.
- Nemcova R. (1997). Criteria for selection of lactobacilli for probiotics use. *Vet. Medical. (Praha)* January: 42 (1): 19-27, 1997.
- Patten J. D. and Waldroup (1988) Use of organic acids in broiler diets. *Poultry Science* 67: 1178-1182.
- Paul S.K, G. Samanta, G. Halder and P Biswas, (2007) Effect of a combination of organic acid salts as antibiotic replacer on the performance and gut health of broiler chickens. *Livestock Research for Rural Development*, 19 (11).
- S.A.S. Institute, SAS user's Guide. (1988) SAS inst. Inc. Cary. N.C.
- Sheikh A., T. Banday, G. Ahmad, B. Masood S. Mir and M. Rehman (2010) Effect of Dietary Supplementation of Organic Acids on Performance, Intestinal Histomorphology, and Serum Biochemistry of Broiler Chicken. *Hindawi Access to Research Veterinary Medicine International*, Volume Article ID 479485.
- Stutz, M. W., S.L. Johnson and F.R. Judith (1983). Effect of diet bacitracin and body weight restriction on the intestine of broiler chicks. *Poultry Science* 62: 1626- 1632.
- Terada, A., H. Hara, J. Sakamoto, N. Sato, S. Takagi and T. Mjsooka (1994). Effect of dietary supplementation with lacto-sucrose (4G-B-D-galctoslsucrese) on cecal flora, cecal metabolites and performance in broiler chickens. *Poultry Science* 73: 1663-1672.
- Vogt, H., S. Maths and S. Harhsch (1981). The effect of organic acid in the ratios on the performance of broiler and laying hens. *Archive. Geflugelkd* 45: 221-232.
- Waldroup, A., S. Kaniawati and A. Mauromoustakos (1995). Performance characteristics and microbiological aspects of broilers fed diets supplemented with organic acids. *Journal of Food Protection* 58: 482-487.
- Watson D. (1960) A simple method for the determination of serum cholesterol. *Clinical Chemistry*, 5: 637.

## تحسين الأداء الإنتاجي والكفاءة الغذائية بإضافة الأحماض العضوية في علائق بداري التسمين.

نجلاء كمال سليمان<sup>1</sup> يوسف محمد اليوسف<sup>1</sup> نفين بهاء الدين غانم<sup>2</sup>

<sup>1</sup> قسم الإنتاج الحيواني والسمكي- جامعة الملك فيصل الاحساء- الهفوف- المملكة العربية السعودية.

<sup>2</sup> قسم علوم الغذاء والتغذية- جامعة الملك فيصل الاحساء- الهفوف- المملكة العربية السعودية.

أجري هذا البحث بهدف دراسة تأثير أنواع مختلفة من الأحماض العضوية هي الستريك- اللاكتيك - البروبونيك وبمستويات 0.5 - 1.5% على الأداء الإنتاجي لكثاكت اللحم وبعض العترات الميكروبية بالأمعاء والشكل الهستولوجي لقطاع في الأمعاء الدقيقة وكذلك دراسة التأثير على بعض مكونات الدم.

استخدم في الدراسة عدد 300 كتكوت لحم من سلالة Ross عمر يوم وحتى 42 يوم من العمر حيث تم تقسيمها إلى عشرة مجموعات تجريبية - كترول وتسع معاملات تجريبية تشمل ثلاث أنواع من الأحماض العضوية ممثلة في ثلاث مستويات من كل حامض عضوي.

تم تربية الكتاكيت في أعشاش أرضية وغذيت على علية بادئ (من صفر إلى 3 أسابيع) تحتوي على 22% بروتين و 3000 كيلو كالورى طاقة ممثلة /كجم وعليقه نامي (من 3 - 6 أسابيع من العمر) تحتوي 20% بروتين و 3000 كيلو كالورى طاقة ممثلة /كجم.

أوضحت النتائج أن هناك تحسن في وزن الجسم مع إضافة الأحماض العضوية وكانت التحسن معنويا مع مستوى 1.5 حمض بروبونيك.

سجلت الكتاكيت المغذاة على الأحماض العضوية قيم استهلاك غذائي عالية مقارنة بالكتكوت عند حسابه خلال مرحلة البادئ (0 - 3) أسابيع و النامي (3 - 6) أسابيع وكذلك خلال كامل فترة التجربة من (0 - 6) أسابيع .

تحسنت كفاءة التحويل الغذائي بإضافة الأحماض العضوية إلى علائق كتاكيت اللحم وكان أعلى مستوى للتحسن في الكتاكيت المغذاة على 0.5% حمض اللاكتيك أو 1.5% حمض البروبونيك بالرغم أن الاختلافات لم تكن معنوية بين المجاميع المختلفة .

أوضحت الدراسة الميكروبيولوجية لمحتويات الأمعاء أنه في كلا المرحلتين العمريتين من (0 - 3) أسابيع و (3 - 6) أسابيع حدث انخفاض معنويا للعدد الكلى للبكتريا لكل جرام من محتويات الأمعاء عند التغذية على الأنواع المختلفة من الأحماض العضوية كذلك انخفض العدد الكلى لبكتريا الايكولاى والاسلامونيلا معنويا نتيجة لإضافة الأحماض العضوية في العلائق وكان حمض البروبونيك هو الأكثر تأثيرا في خفض العدد الكلى للاسلامونيلا. زاد العدد الكلى لبكتريا اللاكتوباسيلس معنويا نتيجة إضافة الأحماض العضوية وكان التأثير أكثر وضوحا خلال المرحلة الثانية من الدراسة (من 3 - 6 أسابيع).

انخفضت درجة pH علائق البادئ والنامي وكذلك محتويات الأمعاء تدريجيا مع زيادة نسبة الأحماض العضوية المضافة للعلائق .

لم تختلف قيم محتوى الدم من الكولسترول و الجلوسريدات الثلاثية والدهون الكلية للكتاكيت المغذاة على علائق مضاف لها الأحماض العضوية مقارنة بالكتكوت. زادت قيم البروتين الكلى ونسبة الجلوبيولين معنويا بينما لم تتأثر نسبة البيومين في الدم معنويا بإضافة الأحماض العضوية للعلائق.

زاد وزن كلا من غدة Bursa والطحال معنويا بالتغذية على علائق تحتوي على نسب مختلفة من الأحماض العضوية .

نستخلص من هذه الدراسة أن إضافة الأحماض العضوية إلى علائق كتاكيت اللحم أدى إلى تحسين الأداء الإنتاجي وانخفاض محتوى الأمعاء من البكتيريا الضارة وزيادة النافعة مما قد يودى إلى تحسين الاستفادة من العناصر الغذائية.