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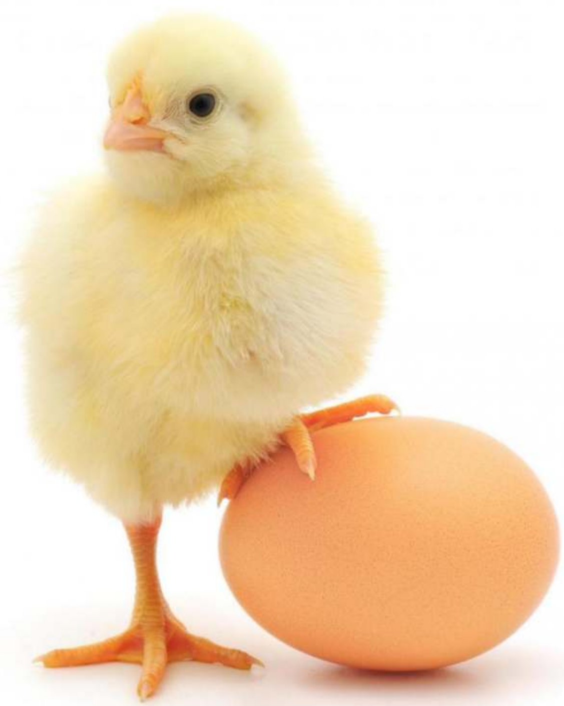

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Effect of Dietary Organic Acid Supplementation on Egg Production, Egg Quality and Some Blood Serum Parameters in Laying Hens

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Abstract: The present study was carried out to determine the effect of dietary organic acid mixture supplementation on performance (body weight changes, feed intake, feed efficiency, egg production) egg quality and concentration of some blood serum parameters in laying hens. A total 53 week old 44000 white Lohmann LSL laying hens which were allotted into 4 groups, each containing 11000 hens. The hens were fed with diets (17% CP% and 2795 Kcal Kg^{-1} ME). Supplemented with 0% (control group No. 1), 260 ppm (group, 2), 520 ppm (group 3) and 780 ppm (group, 4) of organic acid mixture (formic acid and salt of butyric, propionic and lactic acids). The dietary organic acids significantly modify live body weight in groups 2 and 3 and non significantly improved in group 4 when compared with control one. Organic acids supplementation at 780 ppm (group 4) significantly increased egg production by about 5.77% compared with the untreated group, while the lower levels (groups 2 and 3) showed non significant effect. On the other hand dietary organic acid had no effect on the average egg weight, while showed a higher percentage of small, medium and X-large egg size and reduction in large egg size compared with control group. The results revealed that no effect of organic acid on feed intake but improvement egg mass and feed efficiencies was observed. Higher inclusion level of organic acids (780 ppm) improved egg shell quality and non significantly increased egg shell calcium and protein content, improved yolk index, while slightly reduce albumen index. The data revealed that linear increase of serum calcium and concentration of total serum protein. From the obtained data can be concluding that organic acid supplementation at level 780 ppm of laying hens diet improve live body weight, improve persistence of lay and from economical point of view we can concluded that organic acid addition (780 ppm) amazing increase the economical efficiency of layer production.

Key words: Laying hens, organic acids, egg quality, egg production, performance, serum parameters and economic efficiency

Introduction

The poultry sector is continuously searching for new feed additives in order to improve the feed efficiency and chicken health. The use of feed additives has 2 objectives, the first one is to control of pathogen microorganisms such as salmonella and Coliforms and the second one is to enhance the digestive microflora with beneficial microorganisms (Shane, 1999).

Although antibiotics possess these beneficial effects, their use as growth promoters in the poultry industry has been intensively controversial because of the development of bacterial resistance and potential consequences on the human health. That the reason why alternative materials to antibiotics are researched. Among these compounds, organic acids are promising alternatives (Hyden, 2000).

Health of the gut is one of the major factors governing the performance of birds and thus, the economics of poultry production (Samik *et al.*, 2007) and the profile of intestinal microflora plays an important role in gut health (Dhawale, 2005). Dietary organic acids and their salts are able to inhibit microorganism growth in the food and consequently to preserve the microbial balance in the gastrointestinal tract. In addition, by modifying intestinal pH. Organic acids also improve the solubility of the feed ingredients, digestion and absorption of the nutrients (Vogt *et al.*, 1981; Patten and Waldroup, 1988; Owings *et al.*, 1990; Skinner *et al.*, 1991; Adams, 1999).

Previous studies reported that organic acids such as fumaric, propionic and butyric acids and their salts have shown variable effects on egg production and egg quality parameters. These discrepancies would be related to the source, the amount of organic acids used, location, environmental condition and the composition of the diets (Jensen and Chang, 1976; Gama *et al.*, 2000). On the other hand, only few data on blood parameters in laying hens supplemented with organic acids are available. This study was conducted to through light on the effect of different doses of organic acid mixture (Fumaric acid and calcium salt of butyric, propionic and lactic acids) on egg production, some egg quality parameters and some serum constituents during the late production stage of laying hens.

Materials and Methods

This research was conducted at Zainah Commercial Layer Farm in the Kingdom of Saudi Arabia to investigate the effect of organic acid mixture product “ProviMax”

Table 1: Ingredient composition and chemical analysis of the basal diet

Items	Basal Diet (BD)
Ingredients:	
Yellow Corn	54.0
Soybean meal (48%)	24.25
Barley	9.23
Soybean oil	1.0
Limestone	9.8
DCP	1.15
DL-Methionine	0.07
Lysine	0.02
Choline chloride	0.03
Salt	0.25
Mineral premix (0.1%)[1]	0.1
Vitamin Premix (0.1%)[2]	0.1
Chemical Analysis %	
Moisture	11.16
Crude protein	16.95
Ether Extract	3.74
Ash	12.37
Calcium	3.92
Phosphorus (Total)	0.63
ME Kcal Kg[3]	2795

1: Layer vitamin premix produced by Centraly's Co (France) and contain the following vitamins per Kg premix (vitamin A, 10000000, IU; Vitamin D, 2000000 IU; Vitamin E, 20000 mg; Vitamin K, 2000 mg; vitamin B12, 10 mg; Biotin, 200 mg; Folacin, 1000 mg; Niacin, 30000 mg; pantothenic acid, 10000 mg; pyridoxine, 4000 mg; ribroflvin, 5000 mg; thiamin, 2000 mg and proper dose of antitoxidant), 2: Mineral premix produced by Centraly's Co. (contain the following minerals per Kg, Cobalt, 100 mg; Copper, 10000 mg; Iron, 50000 mg; Iodine, 500 mg; manganese, 85000 mg; zinc, 65000 mg and selenium, 200 mg). 3: Metabolized energy calculated according to NRC, 1994)

which contain a mixture of (calcium butyrate-calcium propionate-calcium Lactate and Fumaric acid) on egg production, egg quality and some blood serum biochemical parameters in laying hens.

Birds: Forty four thousand 53 weeks old, Lohmann LSL white layer hens were used in this study. The hens were housed in clean well ventilated house. During the experiment, feeds and water were supplied ad libitum, hens were exposed to 16 hours artificial light and the other to very dim light especially during the night. The housing temperature was maintained at 24°C

Experimental design and feeding program: Laying hens were allotted into 4 groups (11000 hens per each house). The laying hens were fed with a basal diet containing 17% crude protein and other nutrients according to the requirements (NRC, 1994). The ingredient composition and chemical analysis of the basal diets (BD) are presented in Table, 1. The applied experimental design is presented in Table 2. All the experimental diets as well as fresh water were constantly available throughout the experimental period (16 weeks).

Table 2: The experimental design during experimental periods

		Organic acid supplementation	
Group No.	Diet	(ProviMax commercial product)*	Organic acids level
1	Basal diet (Control)	-----	----
2	BD	+ 0.5 Kg Ton^{-1} BD	260 ppm
3	BD	+ 1.0 Kg Ton^{-1} BD	520 ppm
4	BD	+ 1.5 Kg Ton^{-1} BD	780 ppm

* "ProviMax" is a combination of (Fumaric acid, 2%, salt of organic acids "calcium butyrate-calcium propionate and calcium Lactate' 50% and carrier "sepioliite and corn" until 100% produced by Centraly's Co. France

Data Collection, chemical and biochemical measurements: Fifty hen as a random sample were weighed at the beginning and at bimonthly interval (8 weeks) during the experiment were recorded. Egg production was recorded daily for computing the average daily egg production. Egg weight, egg size classification (small, medium, large, X-large and jumbo egg) was separated for computing each class percent from the total egg production, moreover egg mass, feed consumption and feed efficiency (gram feed: gram egg) were recorded daily.
Diet samples were collected once for crude protein, total ash, calcium and phosphorus analysis according to (Randhir and Pradhan, 1981; AOAC, 1985; David, 1976; Cockerell and Holliday, 1975) while, ether extract was determined according to Bligh and Dryer (1959) technique as modified by Hanson and Olly (1963). Diets pH was measured using electric digital pH meter Orion research model 201, after adding 1 g of the diet to 9 mL of dist. Water and mixing for 10 min and B-value measured as milliters (mL) of 0.1 M HCl acid needed to reach a pH of 5 in feed/water mixture (10 g of material in 100 mL of water).
Blood sample were collected by venipuncture from the sub-wing vein and placed in non-additives blood collection tubes to produce serum from a sub-sample of 5 randomly selected laying hens from each treatment groups at the end of the experimental period. Serum was separated by centrifugation at 3000 rpm for 10 min and analyzed for concentration of blood serum protein, albumin, globulin and calcium according to Doumas *et al.* (1981), Reinhold (1953), Coles (1974) and David (1976), respectively.

Egg quality measurements: The percent of thin shell and broken shell were recorded daily. At start and at the end of the experiment a sample of 60 eggs from each group was collected to estimate: Egg shape index (egg width/egg length), yolk index (yolk height/yolk width), albumen index (albumen height/(albumen length + albumen width) according to Card and Nesheim (1972). Shell thickness and weight were recorded also;

Table 3: Effect of dietary organic acid supplementation on feed pH and B-value

Parameters	Groups No. 1 (Control)	2 (0.5 Kg/Ton)	3 (1.0 Kg/Ton)	4 (1.5 Kg/Ton)
Feed pH	6.53±0.01[a]	6.45±0.01[a]	6.41±0.01[ab]	6.31±0.01[b]
Feed B-value	20.1	18.2	16.9	16.2

Values are means ± standard error. Means values with different letters at the same row differ significantly at p = 0.05

Table 4: Effect of dietary organic acid on hen's body weight (BW) changes and mortality during the experimental period (54-70 weeks)

Items	Groups No. 1 (Control)	2	3	4
Hen's weight (Kg):				
53 weeks*	1.86±0.02[a]	1.87±0.02[a]	1.83±0.02[a]	1.86±0.03[a]
62 weeks	1.65±0.02[a]	1.70±0.02[a]	1.63±0.02[a]	1.64±0.03[a]
70 weeks	1.74±0.02[b]	1.82±0.01[ac]	1.81±0.02[ac]	1.79±0.03[bc]
BW change (g)	-120	-50	-20	-70
Mortality %:				
54-58 weeks	0.89	1.56	1.66	1.25
59-62 weeks	0.84	2.59	1.27	1.72
63-66 weeks	1.51	6.03	1.59	2.28
67-70 weeks	4.8	8.41	4.17	3.69
54-70 weeks	7.86	17.48	8.44	8.67

Values are means ± standard error. Means values with different letters at the same row differ significantly at p = 0.05. * body weight before starting the experiment

moreover egg shell was analyzed to determine crude protein, ash, calcium and phosphorus content. Economical efficiency of production: Total production cost was calculated including, feeding, heating, veterinary care, management and housing. Selling price of the produced eggs commonly offered in the market. Economical Efficiency (EE) was estimated as: EE = (Net revenue/Total production cost) × 100.

Statistical analysis: The analysis of variance for the obtained data was performed using Statistical Analysis System (SAS, 1987) to assess significant differences.

Results and Discussion

Feed pH and B-values: Effect of organic acids inclusion in laying hen diet on pH and B-value of the diets are presented in Table 3. Statistical analysis of the obtained data indicated that dietary inclusion of organic acid blend of the diet non significantly reduced pH when added at 260 ppm (group No. 2) when compared with untreated diet, while significantly reduced at 520 and 780 ppm (groups 3 and 4, respectively). On the same way supplementation of organic acids reduced the B-value of the diets according to the inclusion levels.

The pH reducing and antimicrobial effects of the organic acid may assist in gut acidifiers and appear to be good solution for poor egg shell quality (Dibner and Butin, 2002 and Dhawale, 2005). Moreover, the B-value of poultry feeds is particularly important because it affects the course of digestion. When using feeds with high B-values, the pH in the digestive tract may rise to levels unsuitable for optimal digestion and for controlling the bacterial population in the stomach (Langhout, 2000; Yesilbag and Colpan, 2006).

Live body weight change and mortality %: Effect of dietary organic acid mixture on hen's live body weight changes and mortality percent are presented in Table 4. Statistical analysis of the obtained data revealed that dietary organic acids significantly modify live body weight by about 4.59 and 4.02% in groups No. 2 and 3 which fed on the basal diet supplemented by 260 and 520 ppm of organic acids, respectively when compared with the control, while non significantly improved by about 2.87% in group 4 which fed the basal diet supplemented by organic acid at 780 ppm. The results are in agreement with those obtained by (Gama *et al.*, 2000) they reported that dietary addition of 0.05% organic acid increased the body weight of laying hens.

It was clear that group 2 which fed on the basal diet supplemented by organic acid mixture at lower level (260 ppm) showed an increase of mortality in the laying hens, which may be related to the environmental condition of the layer house No. 2 more than the treatment factor. Moreover the other inclusion rates have no effect on the mortality of laying hens during the experimental period.

Production characteristics "Egg production% and egg weight": Effect of dietary organic acid supplementation of egg production of laying hens during the period between 54-70 weeks of age is presented in Table 5. The obtained data revealed that egg production declined within increase of hens age, but organic acid supplementation at 780 ppm (group, 4) maintain the production at the end of the experimental period (70 week of hens age) closely to the starting time of the experiment while it is clearly declined than starting egg

Table 5: Effect of dietary organic acid supplementation on egg production (%) of laying hens during the experimental period (54-70 weeks of age)

Production periods (weeks)	Groups no. 1 (control)	2	3	4
Preliminary(53-54)	82.91±0.46[a]	83.02±0.46[a]	82.56±0.59[a]	82.43±0.43[a]
Period 1 (54-58)	81.63±0.31[b]	83.47±0.37[ab]	82.93±0.48[b]	85.27±0.23[a]
Period 2 (59-62)	82.48±0.08[c]	83.42±0.32[b]	80.77±0.17[c]	87.02±0.05[a]
Period 3 (63-66)	81.48±0.21[b]	79.80±0.20[c]	79.13±0.13[c]	86.31±0.07[a]
Period 4 (67-70)	76.98±0.34[b]	77.03±0.33[b]	78.10±0.79[b]	82.58±0.30[a]
Average (54-70)	80.64±0.24[b]	80.93±0.33[b]	80.24±0.44[b]	85.29±0.19[a]

Values are means ± standard error. Means values with different letters at the same row differ significantly at p = 0.05

Table 6: Effect of dietary organic acid supplementation on average egg weight (g) of laying hens during the experimental period (54-70 weeks of age)

Production periods (weeks)	Groups no. 1 (control)	2	3	4
Period 1 (54-58)	56.83±1.39[c]	60.25±1.25[ab]	59.14±1.25[bc]	61.50±1.37[ab]
Period 2 (59-62)	61.62±1.31[a]	61.60±1.19[a]	63.17±1.26[a]	63.11±1.20[a]
Period 3 (63-66)	69.33±1.32[a]	69.54±1.27[a]	68.86±1.63[a]	65.51±1.03[b]
Period 4 (67-70)	72.85±0.94[a]	73.82±0.86[a]	71.41±0.94[a]	72.51±0.82[a]
Average (54-70)	64.57±1.26[a]	66.05±1.11[a]	65.02±1.13[a]	65.40±1.00[a]

Values are means ± standard error. Means values with different letters at the same row differ significantly at p = 0.05

production percentage of the control and slightly improved in both 260 and 520 ppm organic acids in laying hens diet (group 2 and 3, respectively). Statistical analysis indicated that average egg production% for the whole periods significantly increased by about 5.77% of laying hens fed on the basal diet supplemented by 780 ppm of organic acid (group, 4) when compared with the untreated one (group, 1) while the lower levels of organic acid supplementation (groups, 2 and 3) showed non significantly effect compared with the control.

These results are in agreement with Jensen and Chang (1976), Gama *et al.* (2000) they concluded that organic acid supplementation has positive effects on egg production in laying hens, Moreover, Yesilbag and Colpan (2006) They observed that dietary organic acid supplementation has accelerated the egg production increase in 24-28 weeks old laying hens compared to control birds and extended the period of egg production, indeed, 36-38 week old supplemented hens exhibited a highest laying capacity than non treated hens. On the other hand those obtained results disagree with data obtained by Nollet *et al.* (2004) they reported that a positive effect of sodium butyrate on laying performance increasing from 83.1% (control) to 83.8, 84.3, 84.8 and 86.1% (50, 100, 250 and 500 ppm of sodium butyrate supplementation), respectively, that is conform the variable effect of the additives may be confounded by variations in gut flora and environmental conditions (Miles *et al.*, 1989; Mahdavi *et al.*, 2005).

Effect of dietary organic acid supplementation of average egg weight during the different experimental periods and on different egg classification size (as % from the total egg production) are presented in Table 6 and 7, respectively. Statistical analysis revealed that organic acid supplementation had no effect of the average egg weight compared with the untreated one. On the other hand egg classification size showed a significant effect with organic acid supplementation and the statistical analysis revealed increase of small egg percentage and medium egg production within organic acids supplementation by about (54.21, 84.18 and 54.21%) and (57.7, 103.53 and 40.59%) in groups 2, 3 and 4, respectively when compared with the control, while large and jumbo egg size showed a decreased percentage and X large egg size exhibited a surprising increase of its production with the higher organic acid supplementation (group, 4) when compared with the control one.

Those data agreed with those obtained by (Jensen and Chang, 1976; Gama *et al.*, 2000; Yalcin *et al.*, 2000; Yesilbag and Colpan, 2006) they indicated that the addition of different organic acids into laying hens diet has not significantly modify the egg weight in laying hens. Moreover additives that contain lactic acid producing bacteria in laying hens diet did not influence the egg weight significantly which has already been reported by (Cemiglia *et al.*, 1983; Mohan *et al.*, 1995; Haddadin *et al.*, 1996; Chen and Chen, 2004; Mahdavi *et al.*, 2005).

Feed intake and feed efficiency: Effect of dietary organic acid supplementation on egg mass, feed intake and feed conversion ratio are presented in Table 8. Statistical analysis of the obtained data showed that organic acid supplementation significantly improved egg mass by about 2.29 and 6.67% in laying of groups 2 and 4, respectively when compared with the untreated one, while showed a non significant improvement of group 3

Table 7: Effect of dietary organic acid supplementation on egg classification size (% from the total production) of laying hens during the experimental period (54-70 weeks of age)

Egg classification size	Groups no. 1 (control)	2	3	4
Small Egg:				
Period 1 (54-58)	6.14±0.81[a]	12.20±0.60[a]	12.36±0.67[a]	9.98±0.17[a]
Period 2 (59-62)	4.36±0.26[a]	5.94±0.79[a]	5.44±0.91[a]	5.50±0.36[a]
Period 3 (63-66)	1.38±0.36[a]	0.03±0.03[a]	3.57±0.41[a]	0.15±0.15[a]
Period 4 (67-70)	0.0[a]	0.17±0.17[a]	1.59±1.59[a]	0.15±0.15[a]
Average (54-70)	2.97±0.32[c]	4.58±0.54[bc]	5.47±0.87[ab]	4.58±0.39[bc]
Medium Egg:				
Period 1 (54-58)	15.61±1.56[d]	20.53±0.98[b]	26.41±0.99[a]	18.52±0.76[c]
Period 2 (59-62)	9.25±0.57[d]	16.09±0.77[b]	18.87±0.99[a]	11.71±0.42[c]
Period 3 (63-66)	5.43±0.42[c]	11.89±0.85[b]	15.17±0.73[a]	13.31±0.40[ab]
Period 4 (67-70)	4.77±0.35[c]	6.80±0.43[b]	10.96±0.48[a]	5.76±0.36[bc]
Average (54-70)	8.77±0.59[d]	13.83±0.83[b]	17.85±0.64[a]	12.33±0.49[a]
Large Egg:				
Period 1 (54-58)	39.89±1.25[a]	33.86±0.51[b]	31.14±0.62[c]	30.69±0.81[c]
Period 2 (59-62)	34.75±0.95[a]	37.64±1.11[b]	29.06±0.53[c]	23.81±0.52[d]
Period 3 (63-66)	26.33±0.53[b]	29.59±0.89[a]	28.27±0.65[a]	20.72±0.35[c]
Period 4 (67-70)	22.64±0.45[b]	27.08±0.47[a]	26.37±0.53[a]	14.29±0.57[c]
Average (54-70)	30.90±0.77[b]	32.04±0.54[a]	28.71±0.33[c]	22.38±0.63[d]
X Large Egg:				
Period 1 (54-58)	31.35±1.60[b]	27.48±1.09[c]	22.67±1.09[d]	36.49±0.72[a]
Period 2 (59-62)	43.84±1.33[b]	31.43±1.79[d]	36.19±2.11[c]	54.24±0.79[a]
Period 3 (63-66)	58.15±0.65[b]	49.53±1.30[c]	44.08±1.16[d]	60.12±0.70[a]
Period 4 (67-70)	63.19±0.66[b]	54.14±0.58[c]	51.21±0.51[c]	73.08±0.80[a]
Average (54-70)	49.13±1.31[b]	40.65±1.25[c]	38.54±1.21[d]	55.98±1.30[a]
Jumbo Egg:				
Period 1 (54-58)	6.95±0.31[a]	5.95±0.43[ab]	7.41±0.44[a]	4.70±0.36[b]
Period 2 (59-62)	7.79±0.44[b]	8.89±0.53[b]	10.62±0.53[a]	4.73±0.34[c]
Period 3 (63-66)	8.69±0.46[a]	9.02±0.68[a]	8.91±0.58[a]	3.18±0.30[b]
Period 4 (67-70)	9.22±0.51[b]	12.32±0.60[a]	10.99±0.84[a]	6.72±0.43[c]
Average (54-70)	8.16±0.23[b]	9.05±0.35[a]	9.48±0.33[a]	4.83±0.21[c]

Values are means ± standard error. Means values with different letters at the same row differ significantly at p = 0.05

Table 8: Effect of dietary organic acid supplementation on egg mass (EM), feed intake (FI) and feed conversion ratio (FCR) of laying hens during the experimental period (54-70 weeks of age)

Parameters	Groups no. 1 (control)	2	3	4
EM g/hen/day):				
Period 1 (54-58)	46.35±1.22[b]	50.88±1.04[a]	49.04±0.96[b]	52.44±1.16[a]
Period 2 (5-62)	50.84±1.11[b]	51.39±0.93[b]	51.01±1.01[b]	54.93±1.06[a]
Period 3 (63-66)	56.49±1.07[a]	55.46±0.96[a]	54.45±1.25[a]	56.53±0.88[a]
Period 4 (67-70)	56.08±0.79[b]	56.85±0.65[b]	55.74±0.56[b]	59.87±0.72[a]
Average(54-70)	52.44±0.63[c]	53.64±0.81[b]	52.57±0.56[c]	55.94±0.54[a]
FI (g/hen/day):				
Period 1 (54-58)	108.56±0.37[a]	107.35±0.28[a]	107.58±0.38[a]	107.59±0.37[a]
Period 2 (59-62)	107.56±0.24[a]	108.48±0.32[a]	109.33±0.23[a]	109.27±0.27[a]
Period 3 (63-66)	108.56±0.3[a]	108.62±0.27[a]	108.92±0.36[a]	109.06±0.28[a]
Period 4 (67-70)	108.54±0.43[a]	108.49±0.36[a]	110.08±0.52[a]	108.67±0.33[a]
Average(54-70)	108.77±0.59[a]	108.23±0.16[a]	108.98±0.21[a]	108.65±0.17[a]
FCR:				
Period 1 (54-58)	2.39±0.06[a]	2.13±0.04[b]	2.19±0.05[a]	2.08±0.05[b]
Period 2 (59-62)	2.14±0.05[a]	2.11±0.04[ab]	2.17±0.05[a]	2.01±0.05[b]
Period 3 (63-66)	1.93±0.04[a]	1.97±0.04[a]	2.03±0.05[a]	1.94±0.03[a]
Period 4 (67-70)	1.95±0.03[ab]	1.92±0.02[bc]	1.97±0.04[a]	1.82±0.02[c]
Average(54-70)	2.10±0.03[a]	2.03±0.02[b]	2.08±0.02[a]	1.97±0.02[c]

Values are means ± standard error. Means values with different letters at the same row differ significantly at p = 0.05

by about 0.57%. Feed intake was comparable in the control group and in the organic acid groups. Feed conversion ratio showed significant improvement in laying hen groups which fed on the basal diet

Table 9: Effect of dietary organic acid supplementation on physical character of egg shell (ES) in laying hens

	Groups no.			
Items	1 (Control)	2	3	4
Thin ES (%of TEP)				
54 weeks*	1.55±0.06[a]	1.60±0.07[a]	1.55±0.06[a]	1.54±0.06[a]
70 weeks	1.45±0.05[ab]	1.51±0.005[a]	1.34±0.06[b]	0.72±0.05[c]
Broken ES(% of TEP)				
54 weeks*	0.53±0.03[a]	0.54±0.05[a]	0.56±0.05[a]	0.51±0.04[a]
70 weeks	0.33±0.026[a]	0.34±0.03[a]	0.31±0.02[a]	0.25±0.02[b]
ES thickness (mm)				
54 weeks*	0.31±0.002[a]	0.32±0.008[a]	0.29±0.002[a]	0.29±0.01[a]
70 weeks	0.32±0.006[b]	0.32±0.004[b]	0.35±0.014[b]	0.36±0.016[a]
ES weight (g)				
54 weeks*	8.26±0.21[a]	8.16±0.09[a]	8.07±0.24[a]	8.37±0.24[a]
70 weeks	7.92±0.28[a]	7.56±0.23[a]	8.23±0.47[a]	8.22±0.19[a]

*54 weeks (age of hens at the start of the experimental period). TEP = total egg production. Values are means ± standard error. Means values with different letters at the same row differ significantly at p = 0.05

Table 10: Effect of dietary organic acid supplementation on chemical composition (On DM basis) of egg shell (ES) in laying hens

	Groups no.			
Items	1 (Control)	2	3	4
Crude Protein (%);				
54 weeks*	3.99±0.04[a]	4.10±0.58[a]	4.12±0.15[a]	4.08±0.16[a]
70 weeks	3.19±0.07[a]	3.24±0.04[a]	3.43±0.04[a]	3.44±0.27[a]
Ash (%):				
54 weeks*	90.70±0.41[a]	90.07±0.03[a]	90.59±0.42[a]	90.34±0.08[a]
70 weeks	93.43±1.69[a]	92.11±0.21[a]	91.62±0.68[a]	91.28±1.14[a]
Calcium (%):				
54 weeks*	19.69±0.29[a]	19.46±0.12[a]	19.49±0.19[a]	19.24±0.20[a]
70 weeks	18.40±0.16[a]	18.89±0.16[a]	19.50±0.72[a]	19.38±0.54[a]
Phosphorus (%):				
54 weeks*	0.16±0.01[a]	0.19±0.002[a]	0.17±0.01[a]	0.19±0.01[a]
70 weeks	0.15±0.003[a]	0.14±0.003[a]	0.16±0.008[a]	0.16±0.008[a]

*54 weeks (Age of hens at the start of the experimental period). Values are means ± standard error. Means values with different letters at the same row differ significantly at p = 0.05

supplemented by 260 and 780 ppm organic acid by about 3.3 and 6.19% when compared with the untreated one, while group 3 which fed on diet supplemented by 520 ppm organic acid exhibited a non significant improvement by about 0.95%.

Results regarding feed intake is in agreement with those obtained by Yesilbag and Colpan (2006) who indicated that dietary organic acids in laying hens did not significantly feed consumption, while in contrast with those obtained by Samik *et al.* (2007) they noticed that use of single organic acid salt (ammonium formate or calcium propionate) in the broiler diet lowered feed intake when compared with control birds, that differences may be attributed to the type of organic acid salt used in the study as in our experiment the main salt was butyrate in which Sakata (1987), Pinchasov and Jensen (1989), Leeson *et al.* (2005) they reported that butyric acid, unlike other acids such as propionate, did not depress feed intake. The improvement of feed efficiency were already demonstrated in broiler and quail chickens (Vogt *et al.*, 1981; Patten and Waldroup, 1988; Yalcin *et al.*, 1997; Vesteggh, 1999; Denli *et al.*, 2003). In laying hen Jensen and Chang (1976) have reported a beneficial effect of dietary 0.4 and 0.8% calcium propionate supplementation on feed efficiency.

Egg shell quality: Effect of dietary organic acid supplementation on egg shell quality, egg shell thickness and egg shell weight at the start and end of the experimental period are presented in Table 9. Statistical analysis of the obtained data indicated that no significant differences between groups at the start of the experiment, while at the end of the experimental period data showed that significantly decrease of thin egg shell, broken shell percentage of the total egg production and improvement of egg thickness in laying hens groups fed on the basal diets supplemented by 780 ppm of organic acids (groups No. 4) by about 50.34, 24.24 and 12.5% of the tested parameters respectively when compared with the control one. On the other side supplementation of organic acids at 520 and 780 ppm (groups 3 and 4 respectively) non significantly increased egg shell weight by about 3.91 and 3.79% when compared with the control.

Effect of dietary organic acid supplementation on chemical composition of egg shell at the start and end of the experimental period are presented in Table 10. Statistical analysis of the obtained data indicated that there is no differences of egg shell chemical composition at the start of the experiment (54 weeks of hen's age), while at the end of the experiment observed

Table 11: Effect of dietary organic acid supplementation on internal egg quality in laying hens

	Groups no.			
Items	1 (Control)	2	3	4
Egg Index				
54 weeks*	0.76±0.01[a]	0.76±0.01[a]	0.75±0.01[a]	0.75±0.01[a]
70 weeks	0.76±0.01[a]	0.77±0.01[a]	0.77±0.01[a]	0.77±0.01[a]
Yolk Index				
54 weeks*	44.25±1.13[a]	43.45±1.12[a]	43.17±1.13[a]	42.80±1.13[a]
70 weeks	39.56±1.12[b]	44.69±1.12[ac]	42.26±1.15[bc]	45.68±1.12[a]
Albumen Index				
54 weeks*	6.23±0.78[a]	5.69±0.78[a]	6.43±0.70[a]	5.36±0.70[a]
70 weeks	5.58±0.38[a]	5.19±0.40[a]	5.07±0.39[a]	5.31±0.38[a]

*54 weeks (Age of hens at the start of the experimental period). Values are means ± standard error. Means values with different letters at the same row differ significantly at p = 0.05

Table 12: Effect of dietary organic acid supplementation on the concentration of some blood serum parameters of laying hens

	Groups no.			
Parameters	1 (Control)	2	3	4
Calcium	19.38±1.15[c]	21.43±0.31[bc]	22.09±0.55[b]	24.45±0.37[a]
Total protein	3.98±0.06[b]	3.82±0.05[b]	4.37±0.12[a]	4.72±0.05[a]
Albumin	1.86±0.03[b]	1.89±0.02[b]	2.08±0.03[a]	2.19±0.01[a]
Globulin	2.25±0.11[b]	1.93±0.07[c]	2.26±0.07[b]	2.52±0.07[a]
A/G ratio*	0.83±0.05[c]	0.99±0.05[a]	0.92±0.03[ab]	0.87±0.03[bc]

Values are means ± standard error. Means values with different letters at the same row differ significantly at p = 0.05.

* A/G ratio = Albumin/Globulin ratio.

non significantly increase in egg shell protein content by about 1.57, 7.52 and 7.84% and non significantly increased of egg shell calcium content by about 2.66, 5.98 and 5.33% in groups 2, 3 and 4 which fed on the basal diet supplemented by 260, 520 and 780 ppm organic acid respectively when compared with control group, while egg shell ash content showed a reverse direction.

It was observed that the egg shell quality has improved which is a consequence of the increased mineral and protein absorption. This phenomenon of increased absorption reflected on the increased calcium and protein deposition of the shell and contributes to improve the quality which may result in reduced breaking of the shells. The results are in agreement with those obtained by (Zeidler, 2001; Rodriguez-Navarro *et al.*, 2002; Chen and Chen, 2004) but disagree with those obtained by Mahdavi *et al.* (2005) they reported that addition of lactic acid producing bacteria to the laying hen diet had no significant effect on shell hardness and shell thickness. The results are unharmonious with Yesilbag and Colpan (2006) they reported that supplementation of organic acid mixture in laying hens diet were not improved shell thickness and shell breaking strength and that differences may be attributed to that organic acid mixture of their trail not including butyrate which play a role in the development of intestinal epithelium more than other organic acids (Pryde *et al.*, 2002; Brons *et al.*, 2002).

Egg internal quality: Effect of dietary organic acid supplementation on egg index, yolk index and albumen index at the start and end of the experimental period are presented in Table 11. The statistical analysis of the obtained data revealed that there were no significant differences of the different parameters at the start of the experimental period, while at the end of the experiment (70 week of hen's age) noticed that non significant difference in egg index and non significantly deterioration of albumen index in layer hens groups fed on the basal diet supplemented with different inclusion levels of organic acids when compared with the control group. While, organic acids supplementation improved egg yolk index. These results are in agreement with (Yalcin *et al.*, 2000) they reported significant differences in albumen index, yolk index in layer hens fed on diet supplemented with, 1% lactic acid (Gama *et al.*, 200), stated that a slight decline of haugh unit in hens receiving 0.05% organic acid supplementation. By contrast (Yesilbag and Colpan, 2006) they stated that dietary organic acid have no significant effect on internal egg quality parameters.

Blood serum parameters: Effect of dietary organic acid supplementation on the concentration of some blood serum parameters in laying hens at the end of the experimental period (70 weeks of hen's age) are presented in Table 12. Statistical analysis of the obtained data revealed that linear increase of serum calcium concentration with the inclusion levels of organic acids in laying hen diets and significantly increased the serum total protein and albumin concentration in laying hens of groups 3 and 4 when compared with the control one. This could be attributed to the favorable environment in intestinal tract due to feeding of organic acid, which might have helped to digest and absorb more nitrogen and calcium. In contrast, slight but not significant decrease or increase

Table 13: Economic efficiency of production as affected by dietary organic acid supplementation of laying hens

	Groups no.			
Items	1 (Control)	2	3	4
No. of laying hens	11000	11000	11000	11000
Feed Cost (USD)	39308	39646	40458	40870
Other Costs USD/house)*	34100	34100	34100	34100
Total Costs (USD)	73408	73746	74558	74970
Returns (USD)	89756	89394	88260	95619
Net Income (USD)	16348	15648	13702	20649
T. Return/T. cost ratio (%)	122.27	121.22	118.38	127.54
Net Income/ T. cost %	22.27	21.22	18.38	27.54

*Costs including Vet. Drugs, Egg trays, Electric, water, farm rent, labor and vet. Doctor salary and the breeding period cost.

of total protein and albumin concentration, respectively were observed in the group 2 which fed on the basal diet supplemented by 0.5 Kg organic acid /ton diet. The other serum parameters showing some variation of serum globulin concentration between control and treated birds. The results are in agreement with those obtained by Arslan and Saatci (2004), Who concluded that serum total protein concentration significantly increased in Japanese quail receiving bacteria (Lactobacillus bulgaricus) able to produce lactic acid as probiotics, (Yesilbag and Colpan, 2006) they reported that dietary organic acid supplementation (1 and 1.5%) significantly increased total serum protein and albumin concentration in laying hens (Arun *et al.*, 2006) stated that the serum concentration of protein and calcium increased significantly due to dietary supplementation of lactobacillus sporogenes to broiler diet and (Yesilbag and Colpan, 2006) they reported that dietary organic acid supplementation (1 and 1.5%) significantly increased total serum protein and albumin concentration in laying hens.

Economical efficiency of production: Feeding costs, total production costs, net income (USD/Group) and economic efficiency of production in different laying hen groups shown in Table 13. It was clear from the obtained data that the total costs of laying hens production 73408, 73746, 74558 and 74970 USD for groups 1-4, respectively. These results revealed the possibility of increasing economic efficiency by organic acid supplementation of laying hens diet. The greatest economical efficiency was obtained by group 4 (which fed on the basal diet supplemented 780 ppm of organic acid) by about 23.66% when compared with the control group, while lower level of organic acids (groups 2 and 3) exhibited a decrease of economical efficiency of production by about 4.71 and 17.47%, respectively.

Conclusion: From the obtained data can be concluding that organic acid supplementation at level 780 ppm of laying hens diet improve live body weight, improve persistence of lay and increase daily egg mass output, food conversion ratio, egg shell quality and blood serum protein and calcium concentration during the period of 54-70 week of hens age. The variable effect of organic acids inclusion of laying hen diets may be confounded by variations in gut flora and environmental condition. From economical point of view we can concluded that organic acid addition (780 ppm) amazing increase the economical efficiency of layer production.

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