Effect of probiotic and medicinal plant supplements on the production and egg quality of laying Japanese quail hens

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The present study was conducted to compare the response of laying Japanese quail hens to dietary supplementation with probiotic (Lacto-Sacc) and medicinal plant Thyme flowers (*Thymus vulgaris*) through three-months experimental period. The response was measured in terms of productive performance, egg quality and chemical composition of the edible parts of egg, blood constituents, and digestibility of nutrients. A total of 150 laying Japanese quail hens which had been in production for 12 weeks were, randomly, allocated in a completely randomized design considering five treatments with three replicates and 10 samples in each. A group of them received a layer basal diet with no supplementation and served as control. The second and third groups were fed the control diet supplemented with 1.0 or 2.0 g of probiotic / kg diet, respectively. The fourth and fifth groups were fed the same control diet with a supplement of 1.0 or 2.0 g of Thyme flowers / kg diet, respectively.

The results showed that the supplementation of 1.0 or 2.0 g probiotic / kg diet resulted in significant effect ($p \leq 0.01$) on the improvements of egg production, eggs mass / hen / day, egg weight and feed conversion ratio as compared to control laying hens. However, only slight numerical improvements, but not significant, were observed for means egg production, egg weight, egg mass and feed conversion ratio when birds were fed with 1.0 g Thyme flowers supplemented diet compared with control.

Egg quality measurements were not affected by the dietary treatments. However, yolk cholesterol was significantly ($p \leq 0.01$) decreased by adding 1 g of lacto-Sacc or 1 g Thyme flowers / Kg diet. Neither digestibility of ether extract, nitrogen free extract, crude fiber and organic matter nor energy utilization were affected by dietary treatments. However, probiotic supplementation to diets at 1.0 or 2.0 g / kg diet caused a pronounced increase in protein digestibility as compared to control. Notably, using 2.0 g probiotic Kg diet and 1.0 g Thyme flowers / Kg diet caused significant ($P \leq 0.05$) decrease in plasma cholesterol and plasma total lipids, but they had no significant effects on other blood constituents.

Key word: Quail; probiotic; medicinal plant; egg quality; performance

INTRODUCTION

The chicken industry is one of the most dynamic of world agribusiness trade. The importance of feed supplementation in poultry production has increased in the last years with the aim of improving the economic situation of poultry projects. Nowadays, food safety is more seriously considered than before. On the other hand, economy of food production is also a factor which is not ignorable. Recently, many countries tended to prohibit antibiotics because of their side effects on both bird and human. Researchers are looking to natural means of reducing classical feed additives such as antibiotics in poultry nutrition (Genedy and Zeweil, 2003; Ibrahim *et al.,* 2005). Several researchers

(Kumprecht *et al.,* 1995; Zeweil *et al.,* 1996; El-Sheikh, 2006) have investigated the beneficial effects of feeding microbial culture on poultry as a possible alternative to antibiotics for growth promotion and improvement of feed efficiency. The ideal situation throughout the life of any animal would be to maintain specific numbers of beneficial bacteria in the digestive tract. This would ensure that at all times the animal would have the proper microbial balance. This, of course, cannot be guaranteed under natural field conditions. However, if microorganisms and / or substances, which contribute to the proper microbial balance, were added to the diet then the animal would continually receive a "boost" to establish the proper microbial population. Many investigators gained data in the literature concerning the use of bacterial cultures to control and / or promote the proper environmental conditions for the establishment of an ideal microbial population in an animal's digestive tract. These organisms though will promote metabolism and suppress other undesirable bacteria. The mode of action of probiotics (Sissons, 1988; Makled, 1991) may operate by producing antibiotic substances, inhibiting harmful bacteria, altering microbial metabolism, decrease intestine pH and simulating the immune system. Genedy and Zeweil (2003) used the medicine plant Thyme flowers in growing Japanese quail diets as a growth promoter; they found that Thyme flowers at level of 1 g / Kg diet improved the nutritive value of the diet by increasing body weight, improved feed conversion ratio and along with better economic efficiency.

The present experiment was designed to study the possibility of using probiotic Lacto-Sac and medicinal plant Thyme flowers as supplements to Japanese quail diets to evaluate its effect on productive performance, egg quality, blood constituents, digestibility, and economical benefit.

MATERIALS AND METHODS

The present study was carried out at the Poultry Research Laboratory, affiliated to the Department of Animal and Fish Production, Faculty of Agriculture (Saba Basha), Alexandria University, Egypt.

A total of 150 laying Japanese quail hens which had been in production for 12 weeks were, randomly, allocated in a completely randomized design considering five treatments with three replicates and 10 samples in each. The birds were selected on the basis of more than 70 % egg production rate after a two-week pretest period. A group of them received a layer basal diet with no supplement and served as control. The second and third groups were fed the control diet supplemented with 1.0 or 2.0 g of probiotic (Lacto-Sacc) / Kg diet, respectively. The fourth and fifth groups were fed the same control diet with a supplement of 1.0 or 2.0 g of Thyme flowers / Kg diet, respectively. The basal diet was formulated according to NRC (1994). It contained 19.9 % crude protein and 2717 Kcal / Kg metabolizable energy. Lacto-Sacc was obtained from Nicholasville Kentuky, Altech, Biotechnology Center, USA. It composed, mainly, of dried *S. faecium* fermentation product, dried *L. Acidophilus* fermentation product, Yeast culture (live *Saccharomyces Cerevisiae* grown on media of ground yellow corn, diastatic malt and cane molasses), dried *A. oryzae* fermentation extract, dried *A. niger* fermentation extract and betaglucan. Fresh feed was mixed weekly and not stored for more than one week. The hens were reared in wire batteries under similar environmental conditions. All birds had full access to feed and water. The photoperiod was 16 hours of light per day throughout the experimental period, which lasted for three months. Records were kept for egg production, feed consumption, egg weight, average body weight change, and mortality. Measurements of egg quality were taken on average of 21 eggs from each treatment and were performed through two consecutive days per month. At the last week of the experiment, 18 eggs were collected from each treatment-group and divided into 3 replicates of 6 eggs each. Yolks and albumins of 3 eggs from each replicate were separated, while those of the remaining 3 eggs were blended together (i.e. total edible parts of the egg), and their chemical composition was determined according to the methods of A. O. A. C. (1990). Yolk cholesterol was extracted by the method of Folch *et al.,* (1956) as modified by Washburn and Nix (1974) from three eggs of each replicate. Blood samples from the brachial vein of 3 hens in each treatment were drawn and plasma were obtained by centrifugation of blood at 3500 r.p.m. for 15 min. and kept at – 18º C until analyzing. Plasma total protein, albumin, alanine aminotransferase (ALT), total lipids, and cholesterol were, calorimetrically, determined using commercial kits (from Biomerieux, Poains, France). The digestibility coefficients of nutrients of the experimental diets were determined using six male birds from each treatment. Faecal nitrogen was determined following the procedure outlined by Jakobsen *et al.* (1960). The proximate analysis of feed and excreta was carried

out according to A. O. A. C. (1990). Efficiency of energy utilization was calculated according to the following equation:

Energy utilization % = [GE intake – GE excreted / GE intake] x 100

Data were, statistically, analyzed according to Snedecor and Cochran (1980). The significant differences between means were tested using Duncan's Multiple Range test (1955). Finally, the relative economical efficiency of the products was calculated from the input-output analysis as described by Zeweil and Ismail (1998).

RESULTS AND DISCUSSIONS

The effect of probiotic Lacto-Sacc and medicinal plant Thyme flowers on the productive performance of laying Japanese quail hens is presented in Table 1. Body weight change of birds fed different diets was not significantly affected by different treatments as compared to control. It should be pointed out that any gain in body weight after commencement of egg production should be minimal as the hen is essentially, at its mature body weight. The results showed that the supplementation of 1.0 or 2.0 g probiotic / kg diet brought about a significant ($p \leq 0.01$) improvements in egg production by the value of 6.6 %, eggs mass / hen / day ($p \leq 0.01$) by 12.0 and 11.3 %, egg weight ($p \leq 0.01$) by 5.1 and 4.3 % and feed conversion ratio ($p \leq 0.01$) by 14.8 and 12.1 %, respectively, as compared to control laying hens. However, a slight numerical improvements, but not significant, were observed in means egg production, egg mass, egg weight and feed conversion ratio when birds were fed with 1.0 g Thyme flowers supplemented diet compared with, the control treatment. The addition of Lacto-Sacc or Thyme flowers did not demonstrate any significantly effect on feed consumption. It is of interest to note that egg weight did not decline by the increase of egg production in Lacto-Sacc-fed hens during the experimental period. These results are in good agreement, more or less, with those of Zeweil and Ismail (1998) and Abdel-Azeem (2005), who indicated that egg production, egg weight and egg mass were improved in laying hens fed on probiotic-supplemented diets. The positive effective of Lacto-Sacc supplementation to Japanese quail layers diet could be due to decrease the proliferation of pathogenic bacteria resulting from change in gut environment and enhanced nutrient utilization (Miles, 1993). Lacto-Sacc may, also, enhance enzymatic activity in the digestive tract resulting in improving nutrient utilization (Table 5). The positive effect of probiotic supplementation, observed in the present study, is in agreement, more or less, with that reported by El-Sheikh (2006).

The results presented in Table (1) showed that Lacto-Sacc supplementation at 1 g / Kg diet improved relative economic efficiency than those received diets supplemented with 2 g Lacto-Sacc, 1 or 2 g Thyme flowers or un-supplemented (control) diets. This improvement could be due to improving egg weight, egg mass, and feed conversion ratio. These results agreed with Osman (2003) who reported that, probiotic inclusion 1, 2, 3 and 4 g / Kg diet improved economic efficiency as compared with that of the control diet. Mortality percentage was not affected by different treatments.

Table (1): Performance of laying Japanese quail hens fed on experimental basal diets supplemented with Lacto-Sacc or Thyme flowers.

Parameters	Control	Lacto-Sacc		Thyme flowers	
		1 g / Kg	2 g / kg	1g / Kg	2 g / Kg
Body weight change, g.	14.32 ± 5.81	14.13 ± 5.62	13.71 ± 6.94	15.47 ± 8.11	14.88 ± 7.43
Egg production %, hen-day	74.31±1.50^{b}	79.21± 0.78^{a}	79.23 ± 1.71^{a}	75.71 ± 0.50^{b}	74.78 ± 1.07^{b}
Mean egg weight, g.	10.85 ± 0.18^{b}	11.40 ± 0.11^{a}	11.32 ± 0.12^{a}	10.94 ± 0.18^{b}	10.88 ± 0.10^{b}
Egg mass / hen / day, g.	8.06 ± 0.10^{b}	9.03 ± 0.14 ^{a}	8.97 ± 0.19 ^{a}	8.28 ± 0.11^{b}	8.14 ± 0.17^{b}
Feed consumed / hen day, g.	26.20 ± 1.04	25.57 ± 0.77	26.00 ± 0.88	25.80 ± 0.65	25.97 ± 0.94
Feed conversion, g feed / g egg	3.25 ± 0.09 ^{a}	2.83 ± 0.07 ^{b}	2.90 ± 0.12 ^{b}	3.11 ± 0.10 ^{a}	3.19 ± 0.06 ^{a}
Relative economic efficiency (EE)	100	105.4	97.92	99.96	95.34

$^{a-c}$ Means in the same raw bearing different superscripts differ significantly ($P< 0.05$)

No significant differences were detected concerning egg quality measurements as assessed by weights of egg, yolk, albumen, and shell or by shell thickness, egg specific gravity, albumen index,

and yolk index (Table 2). Abdel-Azeem *et al.,* (2005) showed that laying hens fed on diets containing Yea-Sacc or Lacto-Sacc supplementation did not affect egg quality traits. Also, Zeweil (2003) reported that Japanese quail laying hens fed on diet supplemented with 2 g canella / Kg diet did not affect egg quality trait.

Table (2): Egg components and egg quality for laying Japanese quail hens fed on experimental basal diets supplemented with Lacto-Sacc or Thyme flowers.

Parameters	Control	Lacto-Sacc		Thyme flowers	
		1 g / Kg	2 g / kg	1g / Kg	2 g / Kg
Egg weight, g	10.83± .24	11.26 ± 0.17	11.19 ± 0.10	10.95 ± 0.20	10.89 ± 0.10
Egg specific gravity	1.071 ± 0.002	1.073 ± 0.002	1.072 ± 0.002	1.069 ± 0.004	1.071 ± 0.002
Albumen weight, g	5.54 ± 0.08	5.83 ± 0.21	5.78 ± 0.11	5.74 ± 0.12	5.56 ± 0.10
Albumen %	51.83 ± 1.31	51.79 ± 1.08	51.63 ± 0.52	52.43 ± 0.17	51.07 ± 0.43
Yolk weight, g	3.44 ± 0.16	3.60 ± 0.11	3.61 ± 0.14	3.59 ± 0.09	3.57 ± 0.03
Yolk %	31.79 ± 0.79	31.97 ± 0.46	32.22 ± 0.95	32.81 ± 0.18	32.82 ± 0.05
Egg shell weight, g	0.97 ± 0.07	0.99 ± 0.06	1.02 ± 0.08	0.98 ± 0.04	0..97 ± 0.06
Egg shell %	8.95 ± 0.45	8.79 ± 0.47	9.08 ± 0.66	8.91 ± 0.25	8.91 ± 0.46
Egg shell thickness, mm	0.231 ± 0.003	0.234 ± 0.004	0.229 ± 0.006	0.229 ± 0.005	0.230 ± 0.003
Albumen index	23.03 ± 0.45	23.83 ± 0.58	24.13 ± 0.58	23.61 ± 0.91	22.93 ± 0.51
Yolk index	5.40 ± 0.24	5.30 ± 0.36	5.67 ± 0.37	5.51 ± 0.44	5.34 ± 0.18

[a-c] Means in the same raw bearing different superscripts differ significantly (P< 0.05)

It was notably, that insignificant differences were existed among dietary treatments in the chemical composition of egg, except yolk cholesterol which was significantly ($p \leq 0.01$) decreased by adding 1 g of lacto-Sacc or 1 g Thyme flowers / Kg diet (Table 3). These results were in agreement, more or less, with those of Haddadin *et al.,* (1996) and Mahdavi *et al.,* (2005), who reported that inclusion of *lactobacillus acidophilus* or two strain of bacilli *Bacillus subtilis* and *Bacillus licheniformis* decreased egg cholesterol of laying hens at 40 weeks of age.

Table (3): Chemical composition for the edible parts of eggs of Japanese quail fed on experimental basal diets supplemented with Lacto-Sacc or Thyme flowers.

Criteria *	Control	Lacto-Sacc		Thyme flowers	
		1 g / Kg	2 g / kg	1g / Kg	2 g / Kg
Egg yolks:					
Dry matter %	52.55 ± 0.97	52.88 ± 0.46	52.68 ± 0.43	52.78 ± 0.29	52.94 ± 0.36
Crude protein %	29.64 ± 0.35	29.67 ± 0.38	29.25 ± 0.24	29.37 ± 0.19	29.28 ± 0.34
Ether extract %	63.13 ± 0.75	63.45 ± 0.67	63.34 ± 0.38	63.12 ± 0.47	63.31 ± 0.24
Yolk cholesterol (mg /g yolk)	11.20 ± 0.23 [a]	10.54 ± 0.06 [c]	10.97 ± 0.13[ab]	10.58 ± 0.13[bc]	11.22 ± 0.09[a]
Egg albumen :					
Dry matter %	11.75 ± 0.17	11.84 ± 0.16	11.73 ± 0.09	11.70 ± 0.17	11.80 ± 0.03
Crude protein %	83.68 ± 0.46	83.71 ± 0.43	83.85 ± 0.41	84.09 ± 0.38	83.94 ± 0.18
Ether extract %	8.70 ± 0.07	8.73 ± 0.05	8.73 ± 0.06	8.75 ± 0.05	8.71 ± 0.04
Edible parts:					
Dry matter %	25.96 ± 0.24	26.11 ± 0.42	26.05 ± 0.47	26.10 ± 0.38	26.16 ± 0.31
Crude protein %	47.31 ± 0.66	47.38 ± 0.45	47.20 ± 0.54	47.63 ± 0.27	47.24 ± 0.22
Ether extract %	46.61 ± 0.73	46.47 ± 0.27	46.59 ± 0.37	46.66 ± 0.25	46.48 ± 0.22

a-b Means in the same row bearing different superscripts differ significantly (P < 0.05)
* All criteria were calculated on a dry matter basis.

The effect of probiotic Lacto-Sacc and medicinal plant Thyme flowers on some blood constituents of laying Japanese quail hens are presented in Table (4). The treatments with either Lacto-Sacc or Thyme flowers did not, significantly, affect the plasma total protein, albumin, globulin, albumin /

globulin ratio or ALT, except plasma cholesterol and plasma total lipids, where birds fed 2.0 g lacto-Sacc / Kg diet and 1.0 g Thyme flowers / Kg diet had significantly ($p \leq 0.01$) lower value than the other groups. These results were in agreement, more or less, with those of Abdulrahim *et al.,* (1996) and Mahdavi *et al.,* (2005) who confirmed the important roles of gastrointestinal tract microorganisms in recycling of lipids. Likewise, primary bile salts in the presence of specific microorganisms such as *S. faecium*, *L. Acidophilus* and *Saccharomyces Cerevisiae* are prevented from the re-absorption and have more chance to be converted to second type and this inhibits their absorption. On the other hand, Mahdavi *et al.,* (2005) indicated that microorganisms such as *Bacillus subtilis* and *Bacillus licheniformis* are able to synthesize estrase enzymes alongside with lipase enzymes, which the former converts free fatty acids to esterified form different from triglyceride to intestinal content and finally less chance for triglyceride absorption into the plasma. Genedy and Zeweil (2003) reported that total lipids and cholesterol concentrations tended to decrease in the blood of growing Japanese quail group fed medicinal plant-diets (containing Thyme flowers or Chamomile flower heads) as compared to the control group.

Table (4): Effect of Lacto-Sacc and Thyme flowers on some blood constituents of Japanese quail hens

Parameters	Control	Lacto-Sacc		Thyme flowers	
		1 g / Kg	2 g / kg	1g / Kg	2 g / Kg
Total protein (g %)	4.92 ± 0.12	5.10 ± 0.09	5.06 ± 0.18	5.15 ± 0.09	5.11 ± 0.14
Albumin (g %)	2.68 ± 0.08	2.84 ± 0.10	2.75 ± 0.10	2.73 ± 0.11	2.69 ± 0.10
Globulin (g %)	2.24 ± 0.05	2.26 ± 0.04	2.31 ± 0.08	2.42 ± 0.17	2.42 ± 0.04
Albumin / globulin ratio	1.20 ± 0.01	1.26 ± 0.05	1.19 ± 0.01	1.14 ± 0.11	1.11 ± 0.02
Cholesterol (mg / dl)	204.67 ± 12.00[a]	177.40 ± 7.90[a]	130.43 ±12.47[b]	138.20±15.76[b]	186.10±15.26[a]
Total lipids(mg / dl)	374.22 ± 3.13[a]	368.73 ± 3.53[a]	328.68 ± 3.64[b]	333.90 ± 4.44[b]	371.88 ± 5.53[a]
ALT (IU/L)	51.70 ± 1.22	48.00 ± 2.50	46.97 ± 0.17	48.73 ± 3.88	51.33 ± 3.84

[a-c] Means in the same raw bearing different superscripts differ significantly (P< 0.05)

The results presented in Table (5) showed that neither digestibility coefficient of ether extract, crude fiber, nitrogen free extract and organic matter nor energy utilization was affected by dietary treatments. However, Lacto-Sacc supplementation to diets at 1 or 2 g / Kg diet caused a pronounced increase in protein digestibility as compared to control. Also, a slight increase in protein digestibility was observed due to Thyme flowers supplementation but not reached the significant level. The positive response in protein digestibility due to supplementing the probiotic Lacto-Sacc could be explained by regulation the microbial environment of the intestines, decrease digestive disturbances, inhibit pathogenic intestinal microorganisms and improve feed conversion efficiency.

Table (5): Nutrient metabolizability and digestibility coefficients of mature Japanese quail cocks fed on experimental basal diets supplemented with Lacto-Sacc or Thyme flowers.

Measurements	Experimental diets				
	Control	Lacto-Sacc		Thyme flowers	
		1 g / Kg	2 g / kg	1g / Kg	2 g / Kg
Energy metabolizability, %	79.18 ± 1.70	79.79 ± 1.82	79.21 ± 2.32	78.97 ± 1.72	79.07 ± 0.31
Digestion coefficient, %					
Crude protein	84.2 ± 1.5 [b]	89.4 ± 0.9 [a]	89.1 ± 2.5 [a]	87.1 ± 1.1 [ab]	86.0 ± 1.3 [ab]
Ether extract	73.0 ± 1.6	75.3 ± 1.0	74.8 ± 1.2	73.9 ± 2.1	74.4 ± 0.4
Crude fiber	19.7 ± 2.5	19.3 ± 4.7	22.14 ± 5.7	22.9 ± 3.4	18.7 ± 2.3
Nitrogen free extract	78.3 ± 0.9	79.2 ± 0.8	79.5 ± 1.1	78.4 ± 1.4	77.5 ± 1.1
Organic matter	84.71 ± 0.27	84.52± 0.22	84.51 ± 0.32	84.77 ± 0.21	84.32 ± 0.43

a-b Means in the same row bearing different superscripts differ significantly (P < 0.05)

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