

The Effects of Different Levels of Costmary (*Tanacetum balsamita*) Medicinal Plant on Performance, Egg Traits and Blood Biochemical Parameters of Laying Hens

Research Article

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ABSTRACT

This experiment was conducted to evaluate the effects of different levels of costmary (*Tanacetum balsamita*) medicinal plant on performance, egg traits, and blood biochemical parameters of laying hens. Hundred-eighty of Hi-line laying hens (W 36) from 46 until 58 weeks of age were used in a completely randomized design in five treatments and three replicates (12 birds per replicate). The treatment groups consisted of a control group (1) with no costmary supplementation, and 2, 3, 4 and 5 experimental groups with 0.5%, 1%, 1.5% and 2% of costmary, respectively. Using different levels of costmary significantly affected the performance, egg traits and blood biochemical parameters of laying hens ($P < 0.05$). The highest and lowest change amounts of daily feed intake (113.47 g) and (99.10 g) were obtained in groups 3 and 1, respectively. The best yolk color index (5.5) was observed in group 3 by using 1% of costmary and the highest Haugh unit (98.75), yolk weight (19.35 g) and albumin weight (36.91 g) were observed in group 5 by using 2% of costmary, whereas the highest percent of egg yolk index (44.66%) was observed in control group. The lowest amounts of blood triglyceride (82.05 mg/dL) and cholesterol (617.2 mg/dL) were observed in groups 5 by using 2% of costmary. The overall results showed that the use of 1.5% and 2% of costmary medicinal plant in the diets of laying hens had positive effects on their performance, egg traits, and blood biochemical parameters.

KEY WORDS blood biochemical parameters, egg traits, laying hens, performance, *Tanacetum balsamita*.

INTRODUCTION

Antibiotics as growth promoters in poultry feed are posing serious health risks to human health, because of their residual effects in poultry meat and eggs, as well as result pathogens develop resistance to antibiotics (Cowan, 1999). Currently, poultry scientists are challenged to find out alternatives to antibiotic growth promoters with no side effects for poultry that could be more or as effective against harmful microorganisms in the gastrointestinal tract and to stimulate the growth by increasing the efficiency of feed

utilization and to enhance the immunity (Jaderi *et al.* 2011). There are various compounds and products in nature that have the potential of stimulating growth and combating various diseases by the virtue of being antibacterial, antifungal and etc. Phytobiotics are the substances obtained from the medicinal plants and herbs have wide range of medicinal properties and are the best possible alternatives to antibiotics as growth promoter (Ullah Khan *et al.* 2009). Natural plant products, mainly essential oils, have been evaluated as possible feed additives for animal production, especially considering them for *in vitro* antimicrobial activ-

ity (Cowan, 1999). Moreover, an important propriety which has also been observed in rats (Platel and Srinivasan, 2004). The benefit of some natural substances on the gastrointestinal enzymatic activity includes most likely improving nutrient digestibility.

In laying hens adding 2% of nettle (*Urtica dioica*) dried areal parts powder in their diets significantly increased egg yield, egg mass and eggshell weight, whereas the highest Haugh unit was recorded by using 2% of savory (*Satureja hortensis*) (Jaderi *et al.* 2011). Moreover, different levels of nettle, savory and ziziphora could not significantly affect the blood biochemical parameters and immunity cells. In the other study by using different levels of *Thymus vulgaris*, *Lamiaceae menthapiperita* and *Oreganum vulgare*, the highest egg production percent, egg mass and the best feed conversion, the highest haugh unit and egg yolk color index were resulted by using 2% of *Oreganum vulgare*, whereas the highest amount of feed intake and the lowest level of blood triglyceride were observed by adding 2% of *Lamiaceae menthapiperita*.

The genus *Tanacetum* is one of about 100 genera in the tribe Anthemideae and family Asteraceae. The basic chromosome number of the species in the tribe Anthemideae is $2n=18$. *Tanacetum balsamita* is a perennial herbal plant that can grow up to 80 cm (Keskito *et al.* 1998). Costmary has Eurasian origin showed different habitats (Zarghari 1996; Kubo and Kubo 1995). It has been postulated that the Mediterranean is the primary origin of costmary and in the last three decades it has been naturalized and cultivated in different parts of the world (Hassanpouraghdam *et al.* 2008a; Hassanpouraghdam *et al.* 2008b).

The cultivation of costmary is reported in Iran, Turkey, Romania, Germany, Italy, Spain and England (Bylaite *et al.* 2000). Costmary has a long traditional usage as aromatic water in folk medicine of Iran. Finally we can claim that costmary is distributed in South and South-East of Europe and South-West of Asia but naturalized in most parts of the world.

Costmary contains various has a rich secondary metabolites with diverse biological and therapeutical activities (Kubo and Kubo 1995; Abad *et al.* 2006; Marculescu *et al.* 2001a; Marculescu *et al.* 2001b). These compounds consist of essential oil or volatile oil (monoterpenes and sesquiterpens), phenylpropane derivatives, flavonoids (Flavonols, apigenine derivatives, scutellareine derivatives and luteoline derivatives), tannins and oligo-elements. Essential oil derived from leaves and flowers is the most important active compound of this plant.

Costmary essential oil is extracted by water and steam distillation of aerial parts and is a colorless to pale yellow liquid (Hassanpouraghdam *et al.* 2008a; Mohajjel shoji *et al.* 2008b).

Some studies indicated that major components of costmary essential oil from Iran were included carvone, α -thujone and β -bisabolene (Hassanpouraghdam *et al.* 2008).

This study focused on evaluation the effects of using different levels of costmary (*Tanacetum balsamita*) medicinal plant dried and powdered aerial part on performance, egg traits and biochemical parameters of blood in laying hens.

MATERIALS AND METHODS

Animals and dietary treatments

Hundred-eighty Hi-line laying hens (W 36) from 46 until 58 weeks of age were used in a completely randomized design in five treatments and three replicates (12 birds per replicate).

The treatment groups consisted of a control group (1) with no costmary supplementation, and experimental groups 2, 3, 4 and 5 receiving in 0.5%, 1%, 1.5% and 2% of costmary respectively. The diets were formulated to meet the requirements of laying hen as established by the NRC (1994) (Table 1). Dried aerial parts of costmary were purchased from local market and its compositions were determined according to AOAC (1990). After fine milling, it was mixed with other ingredients. Diets and water were provided *ad libitum*. The lighting program during the experimental period consisted of a period of 16 hours light and 8 hours of darkness. Average environmental temperature was 15 °C.

Performance parameters and egg traits

Birds were individually weighed at the beginning and at the end of the study and body weight gain was calculated. Feed intake, feed conversion, egg production percent, egg mass and egg weight were determined weekly on bird bases. Mortality was recorded if it occurred. The collected eggs were classified as normal or damaged; the latter included the following: fully cracked eggs (an egg with broken shell and destroyed membrane), hair cracked eggs (an egg with broken shell but intact membrane), the eggs without shell (an egg without shell but with intact membrane). For measuring the egg traits, at the end of the experiment, 3 eggs were collected from each replicate. Egg specific gravity was determined by placing them in salty water. Egg shells were cleaned and maintained at environmental temperature for 48 h until dried, and then weighed. The thickness of egg shell was measured by micrometer with accuracy of 0.001(mm) in the middle of egg and in three spots on four eggs. Afterwards, their average was considered as final thickness of egg shell for each experimental unit. Color index of the yolk (Roche color index), yolk index, egg albumin index, Haugh units were also determined (Card and Nesheim, 1972).

Table 1 Composition of experimental diets

| Feeds ingredients | Control group | 0.5% Costmary | 1% Costmary | 1.5% Costmary | 2% Costmary |
|--------------------------------|---------------|---------------|-------------|---------------|-------------|
| Corn | 53.12 | 52.38 | 51.64 | 50.90 | 50.16 |
| Wheat | 20 | 20 | 20 | 20 | 20 |
| Soybean meal-44% | 16.56 | 16.61 | 16.65 | 16.70 | 16.75 |
| Vegetable oil | 0.3 | 0.49 | 0.68 | 0.88 | 1.07 |
| Costmary | 0 | 0.5 | 1 | 1.5 | 2 |
| Oyster shell | 1.09 | 1.09 | 1.09 | 1.09 | 1.09 |
| Dicalcium phosphate | 8.15 | 8.15 | 8.15 | 8.15 | 8.15 |
| Salt | 0.28 | 0.28 | 0.28 | 0.28 | 0.28 |
| Vitamin premix ¹ | 0.25 | 0.25 | 0.25 | 0.25 | 0.25 |
| Mineral premix ² | 0.25 | 0.25 | 0.25 | 0.25 | 0.25 |
| Calculated composition | | | | | |
| Metabolisable energy (kcal/kg) | 2800 | 2800 | 2800 | 2800 | 2800 |
| Crude protein (%) | 14 | 14 | 14 | 14 | 14 |
| Ca (%) | 3.4 | 3.4 | 3.4 | 3.4 | 3.4 |
| Available phosphor (%) | 0.31 | 0.31 | 0.31 | 0.31 | 0.31 |
| Sodium (%) | 0.15 | 0.15 | 0.15 | 0.15 | 0.15 |
| Lysine (%) | 0.64 | 0.64 | 0.64 | 0.64 | 0.64 |
| Methionine + Cysteine (%) | 0.55 | 0.55 | 0.55 | 0.55 | 0.55 |
| Tryptophan (%) | 0.18 | 0.18 | 0.18 | 0.18 | 0.18 |

¹ Vitamin premix per kg of diet: vitamin A (Retinol): 8500000 IU; vitamin D3 (Cholecalciferol): 2500000 IU; vitamin E (Tocopheryl acetate): 11000 IU; vitamin K₃: 2200 mg; Thiamine: 1477 mg; Riboflavin: 4000 mg; Panthothenic acid: 7840 mg; Pyridoxine: 7840 mg; Cyanocobalamin: 10 mg; Folic acid: 110 mg; Choline chloride: 400000 mg.

² Mineral premix per kg of diet: Fe (FeSO₄.7H₂O, 20.09% Fe): 75000 mg; Mn (MnSO₄.H₂O, 32.49% Mn): 74.4 mg; Zn (ZnO, 80.35% Zn): 64.675 mg; Cu (CuSO₄.5H₂O): 6000 mg; I (KI, 58% I): 867 mg; Se (NaSeO₃, 45.56% Se): 200 mg.

Blood biochemical parameters and immunity cells

At the end of this experiment, two hens from each replicate were randomly chosen for blood collection and approximately 5 mL blood samples were collected from the brachial vein. Five ml of collected blood were centrifuged to obtain serum for determination the blood biochemical parameters (glucose, cholesterol and triglyceride Kit package) using Anision-300 auto-analyzer system (Pars Azmoon Company; Tehran, Iran).

Statistical analysis

The data were subjected to analysis of variance procedures appropriate for a completely randomized design using the General Linear Model procedures of SAS, (2005). Means were compared using the Duncan multiple range test. Statements of statistical significance were based on $P < 0.05$.

RESULTS AND DISCUSSION

Performance parameters

The effects of different levels of costmary in feeds on performance of laying hens are summarized in Table 2. Using different levels of costmary had significant effects on feed intake of laying hens ($P < 0.05$). The highest amount of feed intake (113.47 g) and the lowest feed intake (99.10 g) were observed in groups 3 and 1, respectively. Furthermore, there were no significant effects in other parameters of performance in laying hens. The highest percent of egg production (83.92%) and egg mass (52.65 g), and the best feed conversion ratio (2.13) were observed in group 4 by using

1.5% of costmary. Spices and herbs are containing fibers, tannins and saponins in the structure (Vandergrift, 1998) that might lead to decreasing the digestion and absorption of essential nutrients. Consequently, the decrease in digestion and absorption of feeds due to the presence of fibers, tannins and saponins might lead to increasing of feed intake in groups fed diet supplemented with costmary medicinal plant. Phytogetic feed additives (often also called phytobiotics or botanicals) are commonly defined as plant-derived compounds incorporated into diets to improve the productivity of livestock through amelioration of feed properties, promotion of the animals' production performance, and improving the quality of food derived from those animals (Platel and Srinivasan, 2004). Spices and herbs with growth promoting activity increase stability of feed and beneficially influence the gastrointestinal ecosystem mostly through inhibition of pathogenic microorganism's growth. Due to improved health status of digestive system, animals are less exposed to the toxins of microbiological origin (Windisch *et al.* 2008). Consequently herbs and spices help to increase the resistance of the animals exposed to different stress situations and increase the absorption of essential nutrients, thus improving the growth of the animals (Windisch *et al.* 2008). Accordingly, increasing the absorption of essential nutrients due to increase stability of feed beneficially influences the gastrointestinal ecosystem mostly through inhibition of pathogenic microorganism's growth and could be the main causes of improvement of laying hens performance observed in this study (Nobakht and Mehmannaavaz, 2010).

Table 2 Effects of using different levels of costmary (*Tanacetum balsamita*) on performance of laying hens

| Treatments | Egg weight (g) | Egg production (%) | Egg mass (g) | Feed intake (g) | Feed conversion |
|------------|----------------|--------------------|--------------|----------------------|-----------------|
| 1 | 61.14 | 71.54 | 43.75 | 99.10 ^b | 2.28 |
| 2 | 60.69 | 78.73 | 44.75 | 108.85 ^a | 2.47 |
| 3 | 61.82 | 78.89 | 48.86 | 113.47 ^a | 2.36 |
| 4 | 60.79 | 83.92 | 52.65 | 112.19 ^a | 2.13 |
| 5 | 63.42 | 78.56 | 49.84 | 107.45 ^{ab} | 2.16 |
| SEM | 0.97 | 3.63 | 2.88 | 2.72 | 0.16 |

The means within the same column with at least one common letter, do not have significant difference ($P > 0.05$).
SEM: standard error of mean.

Egg traits

The effects of different levels of costmary in feeds on egg traits of laying hens are shown in Table 3. Costmary had significant effects on egg yolk index, egg yolk color index, albumin weight, yolk weight and Haugh unit in laying hens ($P < 0.05$). Adding costmary to the diet of laying hens significantly improved the egg yolk color index, albumin weight, yolk weight and Haugh unit when compared with control group.

The best color index of the yolk (5.5) was observed in group 3 by using 1% of costmary while the highest Haugh unit (98.75), yolk weight (19.35 g) and albumin weight (36.91 g) were recorded in group 5 by using 2% of costmary. In contrast, the highest yolk index (44.66%) was observed in control group. There was a positive correlation between yolk heights with yolk index while a negative correlation between yolk lengths and yolk heights was observed. Increase in yolk height and decrease in yolk length might be behind the increment in yolk index in control group.

The colorants for increasing yolk color in laying hens in intensive production could be caused by the natural carotenoids present in spices and medicinal plants or due to the once of synthetic origin (Nobakht and Mehmannaavaz, 2010).

Consequently, the presence of pigments and carotenoids might lead to increasing the color index of yolk in groups contained costmary especially at the level of 1%. Several studies indicated that increase in availability and utilization of energy of feed ingredients was a major reason that led to increasing of yolk weight in laying hens. In this study, increase availability and utilization of energy of feed ingredients and the presence of plant sterols in costmary might lead to increasing yolk weight in treatments.

Many studies showed that increasing yolk weight was the main reason for the increment in albumin weight and this might explain the increase in albumin weight in groups fed diets supplemented with costmary. Many studies showed a positive correlation between Haugh unit and quality of egg components (yolk and albumin). Egg albumin height and egg weight are indices for evaluation of Haugh unit. Indeed, increase in egg weight is related to increase in albumin weight and yolk weight. Increasing of egg weight due to increase in weight of albumin and yolk in group 5 might be the main cause of improvement in Haugh unit.

Blood biochemical and immunity parameters

It can be seen from Table 4 that using different levels of costmary had significant effects on blood triglyceride and cholesterol ($P < 0.05$). The lowest amounts of blood triglyceride (82.05 mg/dL) and cholesterol (617.2 mg/dL) were observed in group 5 by using 2% costmary. Most of the spices and herbs enhance the synthesis of bile acids in the liver and their excretion in bile, which beneficially effects the digestion and absorption of lipids (Srinivasan, 2005). This might explain the results of the present study in that increase the digestion and absorption of lipids due to enhancing the synthesis of bile acids in the liver and their excretion in bile by costmary, might be the main cause of decreasing the level of blood triglyceride. Plasma cholesterol levels are regulated by the absorption of dietary cholesterol, excretion of cholesterol via fecal sterols or bile acids, cholesterol biosynthesis, and removal of cholesterol from circulation (Byington *et al.* 1995; Delsing *et al.* 2001). Meguro *et al.* (2001) explained several mechanisms for the cholesterol-lowering activity of plant sterols.

Table 3 Effects of different levels of costmary (*Tanacetum balsamita*) on egg traits of laying hens

| Treatments | Specific gravity (mg/cm ³) | Yolk index (%) | Color yolk index | Shell weight (g) | Albumin weight (g) | Weight yolk (g) | Haugh unit | Shell thickness (mm) | Cholesterol | Triglyceride |
|------------|----------------------------------------|---------------------|-------------------|------------------|---------------------|---------------------|---------------------|----------------------|-------------|--------------|
| 1 | 1.064 | 44.66 ^a | 2.67 ^b | 4.83 | 36.75 ^a | 18.85 ^{ab} | 93.23 ^{ab} | 0.314 | 176 | 1430.7 |
| 2 | 1.072 | 39.27 ^b | 3.67 ^a | 5.07 | 33.53 ^b | 16.67 ^c | 87.01 ^b | 0.318 | 194.67 | 1562.3 |
| 3 | 1.073 | 40.63 ^{ab} | 5.5 ^a | 4.88 | 34.38 ^{ab} | 17.85 ^{bc} | 97.96 ^a | 0.279 | 189.67 | 1638.7 |
| 4 | 1.068 | 39.91 ^b | 5 ^a | 5.24 | 35.38 ^{ab} | 17.44 ^c | 98.65 ^a | 0.320 | 178 | 1509.3 |
| 5 | 1.069 | 40.25 ^b | 4.84 ^a | 4.54 | 36.91 ^a | 19.35 ^a | 98.75 ^a | 0.286 | 210.61 | 1749.3 |
| SEM | 0.003 | 1.30 | 0.341 | 0.298 | 0.89 | 0.41 | 3.02 | 0.022 | 10.38 | 201.67 |

The means within the same column with at least one common letter, do not have significant difference ($P > 0.05$).
SEM: standard error of mean.

They reported that plant sterols, which were structurally similar to cholesterol, could displace cholesterol from mixed micelles, since they were more hydrophobic than cholesterol.

Table 4 Effects of different levels of costmary (*Tanacetum balsamita*) on blood biochemical parameters of laying hens

| Treatments | Glucose (mg/dL) | Triglyceride (mg/dL) | Cholesterol (mg/dL) |
|------------|-----------------|----------------------|----------------------|
| 1 | 190 | 113.83 ^{ab} | 1318.2 ^{ab} |
| 2 | 191.33 | 99.67 ^b | 1213.7 ^b |
| 3 | 174.33 | 158.67 ^a | 2039.7 ^a |
| 4 | 161.17 | 105.17 ^{ab} | 1143.8 ^b |
| 5 | 181.67 | 82.05 ^b | 617.2 ^c |
| SEM | 13.96 | 16.99 | 237.39 |

The means within the same column with at least one common letter, do not have significant difference ($P > 0.05$). SEM: standard error of mean.

This replacement caused a reduction of micellar cholesterol concentration and consequently lowered cholesterol absorption. Thus, it was possible that costmary medicine plant could decrease the storage of cholesterol through this mechanism.

CONCLUSION

From the obtained data, it could be concluded that costmary (*Tanacetum balsamita*) at levels 1.5% and 2% of laying hens diet had positive effects on their performance, egg traits, blood biochemical parameters during the period of 46-58 weeks of hens age.

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