THE EFFECT OF USING DIFFERENT LEVELS OF CINNAMON AND THYME POWDER ON EGG CHARACTERISTICS AND FATTY ACIDS PROFILE IN JAPANESE QUAILS

*Nasrollah Vali¹ and Sasan Mottaghi²

¹Department of Animal Science, Shahrekord University, Shahrekord, Iran ²Department of Animal Sciences, Shahrekord, Islamic Azad University, Shahrekord, Iran *Author for Correspondence

ABSTRACT

The present study was conducted to investigate the effect of using different levels of Cinnamon (Cinnamomum verum) and Thyme (Thymus vulgaris) powder supplementation on egg characteristics and fatty acids profile in Japanese quails (Coturnix japonica). A total of one hundred 42 days old Japanese quailswere distributed randomly into five groups as a completely randomized design. The groups were subdivided into five replicates with 4 quails each. The experiment was continued 42 to 105 days. Quails were fed by basal diet as control; basal diet contained 1 and 2 percentage turmeric and thyme powder. In 56, 84 and 105 days a total of 10 eggs from each cage were collected and egg characteristics such as egg mass weight, egg yolk weight, egg shell weight, egg white weight and egg shell thickness were evaluated. Also the fatty acids profiles of eggs were determined. Data from this study showed that there were no significant effects between treatments for egg mass weight. The result showed that egg shell weight and egg shell thickness increased significantly compared to the control ($p \le 0.05$). As result revealed myristic acid (C14:0), palmitic acid (C16:0) decreased significantly but stearic (C18:0) didn't changed by cinnamon and thyme supplementation (p≤0.05). Data obtained from this study showed that palmitoleic (C16:1), oleic (C18:1), linoleic (C18:2), arachidonic acid (C20:4), eicosapentaenoic acid (C20:5) acids increased (p≤0.05) and linolenic acid (C18:3) and docosahexaenoic acid (C22:6) didn't change numerically compared to the control. Result also showed that saturated fatty acids (SFA), monounsaturated fatty acid (MUFA) and polyunsaturated fatty acids (PUFA) differed by using thyme and cinnamon powder compared to the control. In conclusion we could suggested that dietary inclusion of cinnamon and thyme powder at 1 and 2 percentage could have decrease saturated fatty acids and increase unsaturated fatty acids in the eggs of experimental Japanese quails.

Keywords: Cinnamon, Fatty Acids Profile, Japanese Quails, Thyme

INTRODUCTION

The use of antibiotic as growth promoters in livestock has been banned according to concern about their residues in animal tissue and induction of bacterial resistance (Vali, 2009). It have evaluated alternative growth promoter such as probiotics and prebiotics, which benefits the health of digestive tract (Jin, 1998; Fuller, 1989; Mulder et al., 1997). Herbal, as feed additives are possible natural alternatives to the use of antibiotics and probiotics as growth promoters in broiler diets. The importance of feed additives especially of plant origin has greatly increased in recent years and a number of studies have reported that the beneficial effect of herbals or their active components on digestion process and better performance in birds (Al-Kassie, 2009; Hashemi and Davoodi, 2011). Also herbals have received increased attention as possible antibiotic growth promoter replacements (Denli et al., 2004, Faghani et al., 2014). Thyme (Thymus vulgaris) has been reported to possess strong antioxidants properties (Bolukbasi et al., 2006). The compounds which comprise the essential oil of thyme have been identified as phenoylic compounds such as tymol (44.4 up to 58.1%), carvacrol (2.4 up to 4.2%) and γ -terpipene (6.9 up to 18.9%). Thyme may stimulates digestive enzymes such as amylase, protease and lipase and consequently improve digestibility of food elements (Bolukbaşi and Erhan, 2008). Cinnamon (Cinnamomum verum) originates from tropical Asia, especially Sri Lanka and India. Cinnamon is a spice with an old history in human nutrition which it can be used as feed additive in poultries (Vali et al., 2013). Cinnamaldehyde is the

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important component of cinnamon, creating about 65 percent of the extracted essential oil. Immune system stimulating effects has been reported for cinnamaldehyde (Faghani *et al.*, 2014). Also, considerable antibacterial and antifungal properties have been found for cinnamon essential oil. The cinnamon extract potentially significantly inhibits fatty acid oxidation and lipid per oxidation in vitro. In addition different flavonoids isolated from cinnamon have free-radical-scavenging activities and antioxidant properties (Mathew and Abraham, 2006). A number of studies have been conducted to evaluate thyme and cinnamon effects on broiler performance, and laying hens, however, the results have not been consistent (Bolukbasi *et al.*, 2006). The current study was designed to investigate the effect of using different levels of thyme and cinnamon powder on egg characteristics and fatty acids profile in Japanese quails.

MATERIALS AND METHODS

The Birds and Diets

A total of one hundred Japanese quails were used and distributed randomly into five groups a completely randomized design. The groups were subdivided into five replicates with 4 quails (one male and 3 female per each). The experiment period lasted at 63 days (from 42 until 105 days). Quails were fed by basal diet as control; basal diet contains 1 and 2 percent of Cinnamon (*Cinnamomum verum*) and Thyme (*Thymus vulgaris*) powder respectively. Required amounts of the herbs were ground, measured, and added to the base feed for each group. The diets were balanced as recommended by nutrition research council for birds (NRC, 1994). The compositions of basal diet are shown in Table 1. During the breeding period no vaccine and no drug were used. The condition of the experiment was the same for all groups. Each group was exposed to 16 hours of light and 8 hours of darkness per day, the temperature was controlled day and night. Also, water and feed were provided ad libitum.

Ingredients % Control Thyme Thyme Cinnamon					Cinnamon
-		1%	2%	1%	2%
Corn	44.48	44.48	44.48	44.48	44.48
Soybean meal	45.85	45.85	45.85	45.85	45.85
Oil	5.25	5.25	5.25	5.25	5.25
Methionine D-L	0.14	0.14	0.14	0.14	0.14
DCP	1.62	1.62	1.62	1.62	1.62
Oyster shells	1	1	1	1	1
Salt	0.25	0.25	0.25	0.25	0.25
Bicarbonate soda	0.15	0.15	0.15	0.15	0.15
Vitamin Premix*	0.30	0.30	0.30	0.30	0.30
Mineral Premix*	0.30	0.30	0.30	0.30	0.30
Vitamin E	0.30	0.30	0.30	0.30	0.30
Turmeric	0	0	0	1	2
Thyme	0	1	2	0	0
Calculated Nutrients Conter	nt				
ME(Kcal.kg)	2828	2828	2828	2828	2828
CP (%)	23.60	23.60	23.60	23.60	23.60
Ca (%)	1.34	1.34	1.34	1.34	1.34
Available Phosphorus (%)	0.67	0.67	0.67	0.67	0.67

Table 1: The Composition and	Calculated Nutrients	Content of the I	Experimental Diets
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*Supplied per kilogram of feed: 900000 IU of vitamin A, 200000IU vitamin D3, 18000Mg vitamin E, 1800 mg vitamin B12, 30000mg B6, 2000 mg Vitamin K, 6600 Mg vitamin B2, 1800 mg vitamin B1, 3000 Mg B6, 2000mg B12 and 50000 mg Choline chloride.

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Data Collection

In 56, 84 and 105 days a total of 10 eggs from each cage were collected and egg mass weight, egg yolk weight, egg shell weight, egg white weight and egg shell thickness were evaluated. The thickness of the shell was measured by micrometer with the accuracy of 0.001mm in the middle and 3 points of egg shell and the average was considered as the shell thickness.

Also, at the age of 105 days five eggs from each cage were selected and marked in the special boxes and fat separation and extraction of yolk were performed according to the method described by Folch *et al.*, (1957).

After the separation and purification of fats to investigate the fat fatty acids profiles they were methylated and the type and amount of fatty acid in each sample was determined individually.

Statically Model and Data Analysis

The statically model was: Yijk = μ + $\dot{\alpha}$ i+ β j+ eijk

Whereas: Yijk = average effect observed, μ = total average, $\dot{\alpha}$ i= effect of treatments, β j= effect of replications, eijk = effect of errors.

The SAS software process (SAS, 2002) was used for data analysis. The significant difference among the mean were calculated by Duncan's multiple range tests (1995).

RESULTS AND DISCUSSION

The result of egg mass weight, egg yolk weight, egg shell weight, egg white weight and egg shell thickness are summarized in table 2. Data showed that egg mass weight values weren't significantly different. Egg shell weight and egg yolk weight increased significantly by using herbals compared to the control ($p \le 0.05$).

As result revealed from this study egg white weight andegg shell thickness tended to increase by using thyme and cinnamon powers ($p \le 0.05$). Thyme and cinnamon group of the present study had higher egg mass weight, and egg shell weight. This deterioration might be resource from synergic or antagonist effect of mixture of the cinnamon and thyme powder. According to the Semsik *et al.*, (2015) result, cinnamon had positive effects on egg production and egg shell quality. An egg has more than eleven percent fat of in the yolk and most of it is unsaturated, its dual band is sensitive to oxidation and spoilage that causes per oxidation and change in odor, texture (Hargis *et al.*, 1991). During the metabolism and interaction with oxygen, free radicals are created that attack the electrons causing spoilage of molecules and erosion of egg cellular walls.

The antioxidants neutralize these radicals preventing spoilage (Feng *et al.*, 2010; Babu and Srinivasan, 1997). Thyme and cinnamon are both from the labiates family and are herbal plants with antioxidant properties. Use of cinnamon and thyme increases the length of intestine, depth and width of the villis of the intestine and improves absorption of nutrients resulting in improved egg production, egg quality, and hatchability and improve egg production and egg quality (Siimsek *et al.*, 2015). Vali *et al.*, (2014) results indicated that experimental treatments by cinnamon and thyme significantly improved egg quality parameters in Japanese quali (P<0.05) except for yolk weight.

Zeweil *et al.*, (2006) demonstrated that using of 1.0 and 2.0 g of thyme showed no significant improvements in means egg production, egg weight, egg mass in Japanese quail. Orhan and Eren (2011) reported that the addition of essential oil mixture including *Origanum vulgare*, *Thymus vulgaris*, thyme oil, origanum oil, garlic oil, anise oil and fennel oil into laying hen diets had no significant effect on egg weight, egg production.

Aghdam-Shahryar *et al.*, (2011) suggested that there was an insignificant increased in egg yolk percentage in group with 3% thyme in result of increase in egg weight. They noted that percentage of yolk formation depends on nutrients intake. Thymus increases digestion and absorption of nutrients because of having menthol (Alcicek *et al.*, 2003).

Ali *et al.*, (2007) reported that the addition of 0.25% thyme in the diet of laying hens did not change yolk (weight, height, diameter, index and color), shell (weight and thickness), egg shape index and Hough units.

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However, they observed that the addition of thyme increased insignificantly the percentage of egg shape index, shell weight and shell thickness.

Kaya *et al.*, (2013) showed that supplementation of a mixed herbal product containing *Origanum vulgare*, *Thymus vulgaris*, thyme oil, origanum oil, garlic oil, anise oil and fennel oil to diets of laying hens can be beneficial to improve egg quality traits especially such as shell stiffness and thickness. They mentioned that the plant extract mixture could increase quadratically shell stiffness and linearly shell thickness, but did not affect other egg traits.

The effects of using thyme and turmeric on saturated fatty acids in quail's egg are shown in table 3. The results showed that dietary supplementation with thyme and cinnamon led to significant differences between saturated fatty acids in treated groups compared to the control. Data showed that although myristic acid and palmitic acid decreased significantly ($p \le 0.05$), but alsostearic acid didn't changed by using cinnamon and thyme powder.

Table 2. Eggs Characteristics of Experimental Japanese Quans						
Ages (Days)	Egg Mass Weight _(g)	Egg Shell Weight _(g)	Egg Yolk Weight _(g)	Egg White Weight _(g)	Egg Shell Thickness (mm)	
56	$11.66 \pm 0.08^{a^*}$	1.17 ± 0.01^{b}	3.74±0.03	$6.69{\pm}0.05^{b}$	$0.240 \pm 0.01^{\circ}$	
84	$11.85{\pm}0.08^{a}$	1.99 ± 0.01^{b}	3.76±0.03	$6.90{\pm}0.05^{a}$	0.260 ± 0.01^{a}	
105	$11.59{\pm}0.08^{b}$	1.22 ± 0.01^{a}	3.77±0.03	$6.59 {\pm} 0.05^{b}$	0.250 ± 0.01^{b}	
Treatments						
Control	11.55±0.11	1.16±0.01 ^b	3.80 ± 0.04^{ab}	$6.59{\pm}0.07^{b}$	0.240±0.01 ^c	
Thyme (1%)	11.83±0.11	$1.20{\pm}0.01^{a}$	$3.78{\pm}0.04^{ab}$	$6.74{\pm}0.07^{a}$	$0.260{\pm}0.01^{a}$	
Thyme (2%)	11.80 ± 0.11	$1.19{\pm}0.01^{ab}$	$3.81{\pm}0.04^{a}$	$6.80{\pm}0.05^{ab}$	0.253 ± 0.01^{b}	
Cinnamon (1%)	11.51±0.11	$1.20{\pm}0.01^{a}$	3.66 ± 0.04^{b}	$6.64{\pm}0.05^{ab}$	$0.258{\pm}0.01^{ab}$	
Cinnamon (2%)	11.71 ± 0.11	1.21 ± 0.01^{a}	$3.73 {\pm} 0.04^{ab}$	$6.77 {\pm} 0.05^{ab}$	$0.259{\pm}0.01^{a}$	

Table 2. Eggs	Characteristics	of Ex	nerimental	Jananese	Onails
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*Means within row with no common on letter are significantly different ($p \le 0.05$).

 Table 3: The Effect of Using Thyme and Cinnamon on Average Percentage of Saturated Fatty

 Acids Profile in Quails Eggs (150 Days Old)

Fatty Acids(%)	Control	Thyme (1%)	Thyme (2%)	Cinnamon (1%)	Cinnamon (1%)
(C14:0) ^{**}	$0.82{\pm}0.02^{a^*}$	$0.79{\pm}0.01^{a}$	$0.79{\pm}0.02^{a}$	0.70 ± 0.01^{b}	0.70 ± 0.02^{b}
(C16:0)	28.82 ± 0.46^{a}	$25.84{\pm}0.47^{b}$	24.78 ± 0.18^{b}	24.72 ± 0.33^{b}	24.76 ± 0.32^{b}
(C18:0)	6.73 ± 0.28^{a}	5.55 ± 0.35^{b}	5.66 ± 0.19^{b}	6.57 ± 0.23^{a}	6.49 ± 0.23^{a}

*Means within row with no common on letter are significantly different ($p \le 0.05$). **Myristic (C14:0), Palmitic (C16:0) Stearic acid (C18:0)

Cinnamon and thyme powder increased palmitoleic, oleic, linoleicarachidonic and eicosapentaenoic unsaturated fatty acids levels ($p \le 0.05$) compared to the control. The highest values of linolenic (0.37 ± 0.04) and docosahexaenoic (0.60 ± 0.04) were related to the groups n which fed by thyme powder. Gerzilov *et al.*, (2015) concluded that phospholipids, triglycerides and linoleic acid in the egg yolk was higher in the chicks fed by thyme and cinnamon (P<0.05).

Changes in body fat deposition among treatment groups may be related t o consumption of different dietary fatty acid profiles that affected different rates of lipid synthesis or lipid oxidation (Crespo and Esteve Garcia, 2001).

Table 4: The Effect of Using Thyme and Cinnamon on Average Percentage of Unsaturated Fatty
Acids Profile in Quails Eggs (150 Days Old)

Fatty Acids (%)	Control	Thyme (1%)	Thyme (2%)	Cinnamon	Cinnamon
				(1%)	(2%)
(C16:1) ^{**}	$6.80 \pm 0.24^{b^*}$	7.47 ± 0.37^{ab}	8.00 ± 0.22^{a}	7.56 ± 0.40^{b}	7.54 ± 0.36^{ab}
(C18:1)	43.31 ± 0.42^{a}	45.31 ± 0.44^{ab}	45.17 ± 0.33^{a}	45.17 ± 0.35^{b}	45.55 ± 0.38^{ab}
(C18:2)	$11.11\pm0.17^{\circ}$	12.78 ± 0.20^{ab}	13.14 ± 0.17^{a}	12.56 ± 0.25^{ab}	12.48 ± 0.15^{b}
(C18:3)	0.31±0.006	0.33 ± 0.007	0.37 ± 0.04	0.34 ± 0.01	0.35 ± 0.01
(C20:4)	1.27 ± 0.06^{b}	$1.48{\pm}0.04^{a}$	1.55 ± 0.03^{a}	$1.55{\pm}0.09^{a}$	$1.54{\pm}0.05^{a}$
(C20:5)	0.03 ± 0.005^{cb}	0.05 ± 0.004^{ab}	0.06 ± 0.006^{a}	0.03 ± 0.005^{cb}	$0.3 \pm 0.007^{\circ}$
(C22:6)	$0.34{\pm}0.03^{b}$	$0.50{\pm}0.03^{a}$	$0.60{\pm}0.04^{a}$	$0.35 {\pm} 0.02^{b}$	0.38±0.04 ^b

*Means within row with no common on letter are significantly different ($p \le 0.05$). **(C16:1), Oleic (C18:1), Linoleic (C18:2), Arachidonic (C20:4), Eicosapentaenoic (C20:5) Linolenic (C18:3) and Docosahexaenoic (C22:6).

The effect of using thyme and cinnamon powder on SFA, MUFA and PUFA showed that there were significant differences between treatments ($p\leq0.05$). This result mentioned that SFA decreased but MUFA and PUFA increased significantly ($p\leq0.05$) compared to the control. Dalkilic *et al.*, (2009) showed that supplementation of cinnamon oil to diets could be considered as a natural alternative for producing poultry meat with more desirable PUFA.

Table 5: The Effect of Using Thyme and Cinnamon on Average Percentage of SFA, MUFA and
PUFA Profile in Quails Eggs (150 Days Old)

Fatty Acids (%)	Control	Thyme (1%)	Thyme (2%)	Cinnamon (1%)	Cinnamon (2%)
SFA ^{**}	$36.37 \pm 0.30^{a^*}$	32.19±0.59 ^b	31.28 ± 0.28^{b}	31.99 ± 0.49^{b}	31.96±0.54 ^b
MUFA	50.11 ± 0.20^{b}	52.78 ± 0.65^{a}	53.17±0.24 ^a	52.73±0.56 ^a	53.09 ± 0.47^{a}
PUFA	13.7±0.12 ^c	15.4 ± 0.21^{b}	15.75 ± 0.14^{a}	14.85 ± 0.23^{b}	14.80 ± 0.16^{b}

*Means within row with no common on letter are significantly different ($p \le 0.05$). **SFA: Saturated fatty acids, MUFA: monounsaturated fatty acid and PUFA: Polyunsaturated fatty acids.

Bolukbasi et al., (2006) showed that the addition of thyme oil to the broiler feed caused to a significant reduction in the saturated (SFA) and polyunsaturated fatty acid (PUFA) concentrations of the leg and breast tissues. They showed that the monounsaturated fatty acid (MUFA) concentrations in these tissues increased by using experimental diets compared to the control. Youdim and Deans (2000) also found that the palmitic and stearic acid content of a rat's brain fed thyme oil was lower than that from the control group. Lee et al., (2003) also found that the linoleic acid levels increased in adipose tissue with dietary thymol supplementation. Youdim and Deans (2000) investigated the effect of thyme oil on the fatty acid composition of rat brains and also found that the arachidonic acid level decreased with the addition of dietary thyme oil. Case et al., (1995) showed that 150 ppm concentration of thymol and carvacrol decreases serum cholesterol in leghorn hens. In Kaya et al., (2013) study feeding of laying hens with diet containing combination of plant extract mixture and copper increased MUFA level of yolk, but did not decrease the cholesterol level of egg yolk in laying hens. In Gerzilov et al., (2015) study the fatty acid analysis of phospholipids and triacylglycerols showed no statistically significant differences with respect to saturated, monounsaturated and polyunsaturated fatty acids ratio in chicks fed by thyme and cinnamon. Ciftci et al., (2013) suggested that total saturated fatty acid (SFA) ratio decreased and totals unsaturated fatty acid (PUFA) ratio, omega-6 fatty acids increased significantly in serum and thigh meat in cinnamon groups. These results showed that cinnamon oil had hypocholesterolaemic and antioxidant characteristics, and it also improved meat quality. Also, many studies have shown that the phenolic components are responsible for its antioxidant activity (Ali et al., 2007; Bolukbaşi et al., 2006; Lee et al., 2004). It was

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hypotheses that that antioxidant property of cinnamon blocked lipid per oxidation of tissue lipids especially PUFA (Kaya and Macit, 2012). For this reason, PUFA and MUFA of quail's egg increased significantly in the study.

Conclusion

We could be explained by the facts that cinnamon and thyme could benefit acts on egg characteristics and fatty acid profile in Japanese quails. This improvement may be due to the biological functions of these herbal to improve growth or that may be due to its role as stimulant, carminative, enhanced digestibility and antimicrobial and antioxidant properties. Little information has been published concerning the fatty acid profile of quails feed with supplemental herbs just like thyme and cinnamon in the diets. Therefore, further studies are needed for more explanation.

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