

Canola oil and organic selenium in quail diets: fatty acid profile, cholesterol content and external egg quality

Óleo de canola e selênio orgânico na dieta de codornas: perfil de ácidos graxos, colesterol e qualidade de ovos

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Abstract

The effects of partial or total substitution of soybean oil (SO) with canola oil (CO) supplemented or not with organic selenium (SeO) in the diet of quails (*Coturnix coturnix coturnix*) on external egg quality, yolk cholesterol and fatty acid profile were studied. A total of 252 quails were fed throughout three 28-day periods with six treatments: Control, (SO); SO + 0.3 ppm SeO; canola Oil (CO); CO + 0.3 ppm SeO; SO (50%) + CO (50%); SO (50%) + CO (50%) + 0.3 ppm SeO. A completely randomized design was used and treatment means were compared by orthogonal contrasts and Duncan's multiple range test with $\alpha = 0.05$. A significant, 4% increase in oleic acid levels in egg yolk was observed with canola oil supplementation, relative to the control and soybean oil treatments. SeO supplementation did not change the yolk fatty acid profile. Neither canola oil nor SeO supplementation affected yolk cholesterol. Egg weight was higher in birds fed CO supplemented with SeO compared with CO alone, but did not differ significantly from that of other treatments. In conclusion, the substitution of SO with CO in the quail's diet changes the fatty acid profile of the yolk, and increases the concentration of oleic acid while decreasing that of linoleic acid without affecting other egg quality traits. Birds fed diets containing CO supplemented with 0.3 ppm SeO also show increased egg weight.

Key words: Canola oil, cholesterol, eggs, fatty acids, organic selenium

Resumo

Foram estudados os efeitos da substituição parcial ou total do óleo de soja (SO) por óleo de canola (CO), suplementado ou não com selênio orgânico (SeO) na dieta de codornas (*Coturnix coturnix coturnix*) sobre o perfil de ácidos graxos, o colesterol na gema e a qualidade externa dos ovos. Um total de 252 codornas foram alimentadas durante três períodos de 28 dias cada, utilizando seis tratamentos: Controle (SO); SO + 0,3ppm SeO; Óleo de canola (CO); CO + 0,3ppm SeO; ½ SO + ½ CO; ½ SO + ½ CO + 0,3ppm SeO. O delineamento foi inteiramente casualizado e as médias dos tratamentos foram comparadas por contrastes ortogonais e teste de Duncan a 5%. Observou-se um aumento significativo de 4% nos níveis de ácido oleico na gema dos ovos com a suplementação de óleo de canola, em comparação com os demais tratamentos. A suplementação de SeO não alterou o perfil de ácidos graxos

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da gema. Nem o óleo de canola nem a suplementação de SeO afetaram o colesterol da gema. O peso do ovo foi maior nas aves alimentadas com CO suplementado com 0,3ppm SeO comparado com CO não suplementado, mas não diferindo dos demais tratamentos. Conclui-se do experimento que a substituição de SO por CO na dieta das codornas muda o perfil de ácidos graxos na gema, aumenta a concentração de ácido oleico e diminui o ácido linoleico, sem afetar outras características de qualidade do ovo.

Palavras-chave: Ácidos graxos, colesterol, ovos, óleo de canola, selênio orgânico

Introduction

The use of a lipid source in animal diets aims to increase their energy levels (ROWGHANI et al., 2007), improve performance, improve upon the absorption of fat-soluble vitamins (KÜÇÜKERSAN et al., 2010) and enrich products (LEESON; SUMMERS, 2005).

During the last two decades, canola has surpassed peanut, sunflower and cottonseed oils in worldwide production. Furthermore, canola has a very high monounsaturated fatty acid concentration, contains intermediary quantities of the polyunsaturated fatty acids, such as linoleic acid and alpha-linolenic acid, and has a very low concentration of saturated fat (FOULADI et al., 2012).

The replacement of soybean oil with canola oil in the diets of birds is intended to change the fatty acid profile of their eggs, making them healthier for the consumer. Therefore, attempts have been made to decrease the cholesterol content of eggs and enrich them with polyunsaturated fatty acids (PUFA) in order to improve nutritional quality (FAITARONE et al., 2013; GÜL et al., 2012; PITA et al., 2010).

In contrast, the use of proportionally less saturated oil increases the PUFA content of the yolk, amplifying the oxidation potential (SCHRAUZER, 2000). Monogastrics are very responsive to changes in the dietary fatty acid profile. Soybean and canola meal are both PUFA sources, but with different fatty acid profiles (LEESON; SUMMERS, 2005). Birds will deposit in their tissues and eggs the type fatty acids they consume in their diets. This may bring about a downgrade in their products because these fatty acids may undergo oxidative

challenges that can be alleviated by antioxidants. Selenium, as a component of glutathione and other enzymes, protects tissues from oxidative damage (SCHRAUZER, 2000). Selenium is also involved in the prevention of protein oxidation, which could be the mechanism behind the positive effect of Se on the Haugh units of eggs. In addition, selenium in its organic form is more bioavailable than the inorganic form (CANTOR et al., 1997). The organic form of selenium produced by *Saccharomyces cerevisiae* is a nutritional additive composed of trace elements (EFSA, 2011). For this reason, its inclusion in laying diets could be of interest in reducing the oxidative potential of yolks containing fatty acids with a high degree of unsaturation.

In comparison with other species of birds, not much information is available on quail nutrition and its association with fatty acid profile, cholesterol and egg quality in the literature. Therefore, this study aimed to examine the possibility of changing the fatty acid and cholesterol profiles of quail eggs by partially replacing soybean oil with canola oil with or without the addition of organic selenium.

Materials and Methods

The experiment was carried out at the poultry farm of the Federal University of Pelotas, Brazil. A total of 252 quails (*Coturnix coturnix coturnix*) were evaluated during an 84-day trial (3 periods of 28 days). Birds were raised on litter up to 5 weeks of age. At that age, they were allocated to individual cages, where they were kept at 23 ± 1 °C and fed 50 g daily up to 19 weeks of age.

Birds were fed a pre-rearing diet until they reached sexual maturity (approximately 45 days). Following an adaptation period (42 to 53 days of age), birds were fed the experimental diets for 12 weeks (Table 1). The experimental diets consisted of T1 = Control, (SO); T2 = SO + 0.3 ppm SeO; T3 = Canola Oil (CO); T4 = CO + 0.3 ppm SeO; T5 = SO (50%) + CO (50%); and T6 = SO (50%) + CO (50%) + 0.3 ppm SeO. Organic selenium (30 g SeO/100 kg, total of 0.3 ppm Se) was added on top.

Table 1. Dietary composition and analyzed fatty acid composition (% of total fatty acids) of the experimental quail diet.

<i>Ingredients</i>	T1	T2	T3	T4	T5	T6
Maize	48.72	48.72	48.72	48.72	48.72	48.72
Soybean meal-45%	40.20	40.20	40.20	40.20	40.20	40.20
Soybean oil	2.4	2.4	-	-	1.2	1.2
Canola oil	-	-	2.4	2.4	1.2	1.2
Limestone	5.2	5.2	5.2	5.2	5.2	5.2
Salt	0.4	0.4	0.4	0.4	0.4	0.4
Vitamin+mineral premix*	3.0	3.0	3.0	3.0	3.0	3.0
Inert	0.1	0.1	0.1	0.1	0.1	0.1
Organic Se “on top”	-	0.03	-	0.03	-	0.03
<i>Fractional composition of fatty acids</i>						
Arachidonic acid	0.47	0.55	0.69	0.61	0.57	0.57
Behenic acid	0.49	0.56	0.51	0.39	0.47	0.44
Cis-eicosenoic acid	0.36	0.19	0.77	0.84	0.44	0.56
Cis-eicosadienoic acid	0.11	0.04	0.09	0.15	0.05	0.09
Stearic acid	3.59	4.17	3.57	2.92	3.62	3.30
Linoleic acid	49.38	45.51	32.38	35.89	40.70	40.47
Linolenic acid	3.43	2.65	2.79	3.64	2.80	3.16
Oleic acid	26.16	28.15	40.18	40.61	33.66	35.07
Palmitic acid	11.73	13.32	10.99	9.41	11.86	11.23
Palmitoleic acid	0.09	0.11	0.18	0.17	0.17	0.15

*Composition per Kg of Premix (Brastec): Vit A - 86000µg of retinyl acetate; D3 - 50.000 UI; Vit. E - 175mg of dl- α -tocopheryl acetate; K3 - 37mg; B1 - 40mg; B2 - 110mg; B6 - 75mg; Vit B12 - 300mcg; Niacin - 650mg; Folic acid - 17mg; Pantothenic acid 10.000mg; Choline - 250mg; Biotin - 50.000mg; Methionine - 25g; Manganese - 1.750mg; Zinc - 1.250mg; Iron - 1500mg; Copper - 250mg; Iodine - 9mg; Selenium - 7,6mg. T1= Soybean oil, T2= Soybean oil + SeO, T3= Canola oil, T4= Canola oil + SeO, T5= 50% soybean oil+50% canola oil, T6= 50% soybean oil+50% canola oil + SeO.

+The nutritional composition of all diets (Table 1) was identical: ME (kcal/kg): 2,780; C.P. (%): 22.0; Ca (%): 2.7; available P (%): 0.46; total SAA (%): 0.74; total methionine (%): 0.38; total lysine (%): 1.28; total cystine (%): 0.36; choline (mg/kg): 2.04; linoleic acid (%): 2.6; fat (%): 4.78; crude fibre (%): 3.8; Na (%): 0.2 in addition to a commercial vitamin and trace mineral supplement.

Analysis of fatty acid profile

A total of 60 eggs (10 per treatment) were analyzed at the Chemistry Institute, UFPel. Each egg was considered a replicate. Lipid extraction was performed using the Bligh and Dyer (1959) method and esterification was carried out according to the Hartman and Lago (1973) protocol. Samples were analyzed by gas chromatography, using the

Shimadzu GC-2010 chromatographer, with an auto-injector AOC-20i (Shimadzu) and 2560 SPTM column (Supelco), with dimensions of 100 m x 0.25 mm I.D. x 0.2 μ m. The standard used was frame mix 100 m SP-2560 from Supelco, with an injection volume of 1 μ L and split 100:1, to identify up to 37 fatty acids.

Direct saponification and cholesterol extraction

The methodology for extraction and dosage of cholesterol in the yolk was described by Bragagnolo and Rodriguez-Amaya (2003). Our decision to use the enzymatic method for cholesterol determination was based on Mazalli et al. (2003), who found no differences in cholesterol quantification between the enzymatic and chromatographic methods (FAITARONE et al., 2013).

A total of 0.25 g yolk was added to a 50 mL polypropylene vial. An alkaline saponification was performed, using 10 mL KOH (2%) in ethanol. Later, vials were placed in a water bath at 50 °C with shaking for 2 h. Then, 5 mL distilled water was added and the bath allowed to cool. The extraction of unsaponifiable material in the sample was performed in three successive extractions. A total of 10 mL hexane (P.A.) was added with shaking in a vortex tube for 1 min. After separation, all of the organic phase was transferred to a new 50 mL vial.

For cholesterol determination, a commercially available kit was used (LABTEST®). A sample of 0.5 mL organic extract was added to a 5 mL glass vial and evaporated under nitrogen in a 37 °C water bath. A total of 0.5 mL isopropanol chromatographic substance was added with shaking by vortex until complete solubilisation occurred. A total of 3 mL enzymes were added to the vial kept in the water bath at 37 \pm 2 °C for 10 min of thermal treatment.

Absorbance at 499 nm was read in a spectrophotometer and corrected by comparison with a blank. Six samples ranging from 0.04 mg/mL to 0.24 mg/mL were used to generate a standard cholesterol curve with an $r^2 = 0.991$.

Egg quality

A total of 360 eggs were harvested in each period to evaluate egg weight, albumen height, Haugh units, yolk weight, shell weight, shell thickness and specific gravity. At the end of each experimental period, each egg was identified, individually weighed and analyzed at the Animal Nutrition Laboratory, UFPEL. Fresh eggs were placed in a perforated plastic basket and immersed in buckets with different salt concentrations for the determination of specific gravity.

Egg shells were washed and dried in a furnace at 60 °C, for 24 h. Later, shells were weighed and their thickness evaluated with a micrometer. The albumen height was determined with a specific ruler. The Haugh unit was obtained using the formula: $UH = 100_{\log} (H + 7.57 - 1.7W^{0.37})$, in which H = albumen height (mm); W = egg weight (g) (SILVA et al., 2000).

Statistical analysis

In the present study, we were not interested in exploring the pattern of relations between quail age and the response to treatments, or in detecting overall differences between related means of age effect on response variables. Therefore, a completely randomized design was used with the following statistical model: $y_i = \mu + \alpha_i + E_i$, where μ is the mean, α_i are fixed effects of the treatment and E_i is the random error associated with the treatment.

Treatment means were compared using Duncan's test and polynomial orthogonal contrasts with $\alpha = 0.05$.

Results and Discussion

The main fatty acids found in egg yolk were oleic (43%), palmitic (24%), linoleic and stearic (>10%) acids. Linolenic, arachidonic and palmitoleic acids were present at less than 3% each. The lowest amount of oleic acid was obtained in the soybean oil + SeO (40%) group, while the highest amount was observed in the group fed canola oil (45%) ($P < 0.05$).

Conversely, the highest (13.9%) and lowest amounts of linoleic acid were found in the soybean and canola oil groups, respectively. These results are in agreement with those of Gül et al. (2012), from a study conducted in laying hens.

High levels of monounsaturated fatty acids (MUFA) and low levels of PUFA were observed in the canola oil group. This was due to the differences in fatty acid profiles of the diets brought about by the presence of canola oil (Table 1).

The quantities of PUFA and MUFA in egg yolk observed in this trial were in agreement with those found by other researchers in hens (GÜL et al., 2012; MILINSK et al., 2003; PITA et al., 2010) and quails (GÜÇLÜ et al., 2008).

Monounsaturated fatty acids (MUFA) increased in the groups with increasing levels of canola oil (GÜL et al., 2012). Milinsk et al. (2003) reported that supplementation with canola oil increased the amount of oleic acid compared with other feed-grade oil sources (soybean oil, flax oil, sunflower oil). Güçlü et al. (2008) found the highest level of oleic acid in the group fed rapeseed oil due to the high amount of oleic acid in rapeseed (73.2%) in

comparison with various other oil sources.

However, Pita et al. (2010) examined commercial layers supplemented with different oil sources and found a higher MUFA composition in the egg yolk of birds fed linseed oil, a mixture of industrial sardine and tuna oil, crude salmon oil and canola oil, respectively, in comparison with soybean and corn oils. These results are in agreement with the present study, in which treatments including canola oil increased the incorporation of MUFA in egg yolks, in comparison with soybean oil.

Canola oil prevents the accumulation “bad” low density lipoprotein cholesterol (LDL-C) by enriching the MUFA (oleic acid) content of the diet (GÜL et al., 2012). Monounsaturated fatty acids, such as oleic acid, play an important role in human health, by preventing dyslipidemia, which brings about cardiac diseases. In this regard, oleic acid has been associated with decreasing serum levels of total cholesterol and LDL-C and increasing levels of high density lipoprotein cholesterol (HDL-C) in individuals with hypercholesterolemia, as well as reducing blood sugar levels in diabetics (SOARES; ITO, 2000). Therefore, quail egg enrichment with MUFA from sources such as oleic acid can improve the quality of the egg for human nutrition. Replacing soybean oil with canola oil in the diet significantly changed the concentrations of the MUFA, oleic acid and the PUFA, linoleic acid in the yolk. The analysis of these results using Duncan’s multiple range test are shown in Table 2. The amount of oleic acid significantly increased by 4% with the inclusion of canola oil in the diet, relative to the control and soybean oil treatments.

Table 2. Effects of partial or total substitution of soybean oil with canola oil supplemented or not with SeO in the diet of quails on fatty acid profile (%) and cholesterol content (mg/g) of egg yolk.

<i>Treatments</i>	PALM	PALT	ESTE	OLEI	LILEI	LILEN	ARAC	CHOL
T1-SO	24.14	2.36	10.88	42.24 ^{bc}	12.5 ^{ab}	0.27 ^b	2.05	13.04
T2-SO+SeO	24.23	2.99	10.19	40.00 ^c	13.9 ^a	0.32 ^{ab}	2.16	13.83
T3-CO	23.55	2.65	10.0	45.47 ^a	9.8 ^c	0.30 ^{ab}	1.93	14.33
T4- CO+SeO	24.18	2.91	9.94	44.94 ^{ab}	9.9 ^c	0.29 ^{ab}	1.95	13.57
T5-SO+CO	24.14	2.76	10.29	42.84 ^{abc}	11.9 ^b	0.33 ^{ab}	1.89	11.89
T6-SO+CO+SeO	23.46	2.62	10.32	42.00 ^c	12.8 ^{ab}	0.37 ^a	2.19	14.12
<i>Contrasts</i>	<i>P-value</i>							
C1	NS	NS	NS	0.0001	0.0001	NS	NS	NS
C2	NS	NS	NS	NS	NS	NS	NS	NS
C3	NS	NS	NS	0.004	0.0001	NS	NS	NS
C4	NS	NS	NS	NS	NS	NS	NS	NS

Means within columns with no common superscripts differ significantly by Duncan's test at $P < 0.05$.

C1 = T1 T2 vs. T3 T4 (soybean oil vs. canola oil), C2 = T1 T2 vs. T5 T6 (soybean oil vs. mixture of soybean oil and canola oil), C3 = T3 T4 vs. T5 T6 (canola oil vs. mixture of soybean oil and canola oil), C4 = T2 T4 T6 vs. T1 T3 T5 (Effects of organic selenium supplementation). PALM = palmitic; PALT = palmitoleic; ESTE = stearic; LILEI = linoleic; LILEN = linolenic; ARAC = arachidonic; CHOL = Cholesterol. SO=soybean oil, SO+SeO=soybean oil+organic selenium, CO=canola oil, CO+SeO=Canola oil+SeO, SO+CO=soybean oil (50%) + Canola oil (50%), SO+CO+SeO= Soybean oil (50%) + Canola oil (50%) + SeO.

It is worth highlighting that the total oil inclusion in the feed formulation was only 2.4% (Table 1). Therefore, to further increase the proportion of these fatty acids in egg yolks, a larger percentage of canola oil in the feed formulation would be required.

In terms of linoleic acid, a significant reduction was observed in treatments containing canola oil. In the present study, the reduction of linoleic acid in the yolk reflects the reduction of this fatty acid in diets containing canola oil (Table 1).

Dietary treatment did not affect cholesterol levels in the yolk (Table 2). These results are in agreement with other studies (FAITARONE et al., 2013; CEYLAN et al., 2011; KÜÇÜKERSAN et al., 2010; ROWGHANI et al., 2007).

The amount of cholesterol in the egg yolk is affected, by among other things, the bird species, age, rearing conditions and nutrition (NOWACZEWSKI et al., 2010). Nevertheless, all of the results obtained in the present study for cholesterol content in quail egg yolks were corroborated by those found

in the literature. The cholesterol contents obtained here were lower than those found by Michalska and Stępińska (1996), who observed the cholesterol content of yolk to be between 14.3 and 18.2 mg/g, but were higher than those in Rowghani et al. (2007) and Kaz'mierska et al. (2005) who found values of 12.3 and 7.78 mg/g in yolk, respectively. Therefore, it can be concluded that the methods used in the present experiment were adequate for comparing cholesterol content in egg yolks.

Other studies reported limited success in reducing the cholesterol content of final products (NABER, 1976; HARGIS, 1988; GRIFFIN, 1992; SHAFEY et al., 1999). However, some improvements in reducing cholesterol levels in eggs have been reported previously, suggesting that additional research is needed to attain more conclusive results. Sun et al. (2003) reported that the cholesterol content of egg yolk was significantly reduced by dietary conjugated linoleic acid.

Mazalli et al. (2004) evaluated the effects of PUFA supplementation in bird diets and found

that linseed oil, canola oil, sunflower oil and fish oil were able to decrease cholesterol levels in hen egg yolks. The main goal in decreasing cholesterol levels is to provide healthy foods. Compared with saturated fatty acids, oleic acid can decrease plasma LDL-C concentrations (REAVEN et al., 1994); it also lowers endogenous cholesterol synthesis, compared with PUFA (JONES et al., 1994).

Mediterranean populations who consume foods rich in oleic acid have lower rates of cardiovascular disease, diabetes and obesity (DE LORGERIL; SALEN, 2006). However, Lewis et al. (2000) have not found any reduction in cholesterol levels in

people fed eggs enriched with PUFA.

In Table 2, contrast 4 shows that organic selenium supplementation did not influence the profile and concentration of fatty acids in egg yolks. Similarly, while evaluating another antioxidant, Galobart et al. (2001) verified that vitamin E supplementation did not alter the PUFA content of hen eggs.

In the first evaluation period, quails fed canola oil + SeO showed higher egg weight in comparison with those fed canola oil ($P < 0.05$), but egg weight did not differ from that of other treatments (Table 3). This result shows a possible potentiation effect of SeO on egg weight in the presence of canola oil.

Table 3. Effects of partial or total substitution of soybean oil with canola oil supplemented or not with SeO in the diet of quails on egg weight, Haugh unit and yolk weight.

<i>Treatments</i>	Period 1			Period 2			Period 3		
	Egg weight (g)	Haugh unit	Yolk weight (g)	Egg weight (g)	Haugh unit	Yolk weight (g)	Egg weight (g)	Haugh unit	Yolk weight (g)
T1-SO	14.45 ^{ab}	101.9 ^a	4.44	14.54	99.6	4.50	14.45	99.5 ^a	4.42
T2-SO+SeO	14.39 ^{ab}	101.7 ^a	4.52	14.28	99.8	4.49	14.43	99.4 ^{ab}	4.38
T3-CO	13.98 ^b	99.8 ^{ab}	4.10	14.28	98.1	4.38	14.21	99.6 ^a	4.51
T4-CO+SeO	14.94 ^a	100.9 ^{ab}	4.40	14.40	99.2	4.44	14.18	97.4 ^b	4.59
T5-SO +CO	14.21 ^{ab}	98.2 ^b	4.23	14.26	99.1	4.40	14.23	99.8 ^a	4.47
T6SO+CO+SeO	14.35 ^{ab}	101.6 ^a	4.34	14.49	98.5	4.52	14.37	98.5 ^{ab}	4.38
<i>Contrasts</i>	<i>P-value</i>								
C1	NS	NS	NS	NS	NS	NS	NS	NS	0.04
C2	NS	NS	NS	NS	NS	NS	NS	NS	NS
C3	NS	NS	NS	NS	NS	NS	NS	NS	NS
C4	NS	NS	NS	NS	NS	NS	NS	0.03	NS

Means within columns with no common superscripts differ significantly by Duncan's test at $P < 0.05$.

C1 = T1 T2 vs. T3 T4 (soybean oil vs. canola oil), C2 = T1 T2 vs. T5 T6 (soybean oil vs. mixture of soybean oil and canola oil), C3 = T3 T4 vs. T5 T6 (canola oil vs. mixture of soybean oil and canola oil), C4 = T2 T4 T6 vs. T1 T3 T5 (Effects of organic selenium supplementation). SO=soybean oil, SO+SeO=soybean oil+organic selenium, CO=canola oil, CO+SeO=Canola oil+SeO, SO+CO=soybean oil (50%)+Canola oil (50%), SO+CO+SeO= Soybean oil (50%) + Canola oil (50%) + SeO.

Agah et al. (2010) observed that the weight of eggs decreased with an increase in the level of canola seed in the diet. This decrease in egg weight is attributed to the reduction in linoleic acid in the diet (RASOULPOUR et al., 2011).

Internal egg quality evaluated by the Haugh unit was significantly affected by treatments in the first and third periods. In the first period, birds fed a mixture of soybean oil and canola oil without SeO supplementation showed lower Haugh units than those fed the same treatment combination

supplemented with SeO. During the third period of evaluation, the canola oil + SeO treatment produced the lower Haugh unit.

Through orthogonal contrasts (Table 3), significant differences were found only in the third period of evaluation. In this case, the SeO supplementation produced lower Haugh units compared with no supplementation (T2 T4 T6 vs. T1 T3 T5) and soybean oil showed lower yolk weight in comparison with canola oil (T1 T2 vs. T3 T4).

Therefore, the hypothesis that increasing levels of selenium in hens' diet could improve yolk stability (HESS et al., 2003) was not corroborated by the present experiment. All analyses were performed in fresh eggs (one day) without storage.

Therefore, the antioxidant effect of SeO when transferred from the diet to the egg by stimulating glutathione peroxidase (GSH-Px) and decreasing lipid and protein oxidation in the egg yolk during storage could not be verified.

In the first and second period, no significant differences between treatments on specific gravity, shell weight and shell thickness were found (Table 4). In the third period, canola oil + SeO treatment produced a lower specific gravity, compared with the mixture of soybean + canola oil ($P < 0.05$). Quails fed canola oil showed inferior specific gravity to birds fed soybean oil (T1 T2 vs. T3 T4). On the other hand, quails fed a mixture of soybean oil and canola oil showed better specific gravity as compared with birds fed canola oil (T3 T4 vs. T5 T6).

Table 4. Effects of partial or total substitution of soybean oil with canola oil supplemented or not with SeO in the diet of quails on specific gravity, shell weight and shell thickness.

Treatments	Period 1			Period 2			Period 3		
	SG	SW	ST	SG	SW	ST	SG	SW	ST
T1-SO	1072.9	1.297	25.16	1073.2	1.259	24.75	1074.7 ^a	1.276	25.90
T2-SO+SeO	1072.7	1.229	25.52	1057.3	1.215	24.57	1072.9 ^{ab}	1.276	25.07
T3-CO	1072.7	1.230	25.08	1071.7	1.244	24.67	1072.0 ^{ab}	1.259	25.46
T4-CO+SeO	1073.7	1.246	25.07	1057.7	1.229	24.84	1071.5 ^b	1.261	25.39
T5-SO +CO	1074.4	1.234	25.14	1073.7	1.217	24.57	1074.8 ^a	1.274	25.41
T6SO+CO+SeO	1072.9	1.256	25.93	1074.7	1.260	25.13	1074.1 ^{ab}	1.312	25.81
Contrasts	P-value								
C1	NS	NS	NS	NS	NS	NS	0.04	NS	NS
C2	NS	NS	NS	NS	NS	NS	NS	NS	NS
C3	NS	NS	NS	NS	NS	NS	0.009	NS	NS
C4	NS	NS	NS	NS	NS	NS	NS	NS	NS

Means within columns with no common superscripts differ significantly by Duncan's test at $P < 0.05$.

C1 = T1 T2 vs. T3 T4 (soybean oil vs. canola oil), C2 = T1 T2 vs. T5 T6 (soybean oil vs. mixture of soybean oil and canola oil), C3 = T3 T4 vs. T5 T6 (canola oil vs. mixture of soybean oil and canola oil), C4 = T2 T4 T6 vs. T1 T3 T5 (Effects of organic selenium supplementation). SG=Specific gravity, SW=Shell weight, ST=Shell thickness, SO=soybean oil, SO+SeO=soybean oil+organic selenium, CO=canola oil, CO+SeO=Canola oil+SeO, SO+CO=soybean oil (50%) + Canola oil (50%), SO+CO+SeO= Soybean oil (50%) + Canola oil (50%) + SeO.

In conclusion, the replacement of soybean oil with canola oil in the quail's diet increases oleic acid in the yolk, which is important from a nutritional point of view, without affecting cholesterol and

other egg characteristics. The supplementation of 0.3 ppm SeO in the quail's diet promotes, in some cases, a positive response in egg quality.

References

- AGAH, H. J.; NASRIRI-MOGHADDAM, H.; TAHMASBI, A. M.; LOTFOLLAHIAN, H. Performance and fatty acid compositions of yolk lipid from laying hens fed with locally produced canola seeds (*Brassica napus* L.). *Research Journal of Biological Sciences*, Taiwan, v. 5, n. 2, p. 228-232, 2010.
- BLIGH, E. G.; DYER, W. J. A rapid method of total lipid extraction and purification. *Canadian Journal of Biochemistry and Physiology*, Ottawa, v. 37, n. 8, p. 911-917, 1959.
- BRAGAGNOLO, N.; RODRIGUEZ-AMAYA, D. B. Comparison of the cholesterol content of Brazilian chicken and quail eggs. *Journal of Food Composition and Analysis*, San Diego, v. 16, n. 2, p. 147-53, 2003.
- CANTOR, A. H.; PESCATORE, A. J.; STRAW, M. L.; FORD, M. J.; DUNLAP, M. K. Tissue selenium concentrations of broilers fed diets supplemented with selenized yeast and sodium selenite. *Poultry Science*, Champaign, v. 76, p. 58, 1997. Supplement 1.
- CEYLAN, N.; CIFTÇI, I.; MIZRAK, C.; KAHRAMAN, Z.; EFIL, H. Influence of different dietary oil sources on performance and fatty acid profile of egg yolk in laying hens. *Journal of Animal and Feed Sciences*, Jablonna, v. 20, n. 1, p. 71-83, 2011.
- DE LORGERIL, M.; SALEN, P. The Mediterranean-style diet for the prevention of cardiovascular diseases. *Public Health Nutrition*, Cambridge, v. 9, n. 1A, p. 118-123, 2006.
- EUROPEAN FOOD SAFETY AUTHORITY - EFSA. Scientific opinion on safety and efficacy of Sel-Plex® (organic form of selenium produced by *Saccharomyces cerevisiae* CNCM I-3060) for all species. *EFSA Journal*, Parma, v. 9, n. 4, p. 2110-2110, 2011.
- FAITARONE, A. B. G.; GARCIA, E. A.; ROÇA, R. O.; RICARDO, H. A.; ANDRADE, E. N.; PELÍCIA, K.; VERCESE, F. Cholesterol levels and nutritional composition of commercial layers eggs fed diets with different vegetable oils. *Brazilian Journal of Poultry Science*, Campinas, v. 15, n. 1, p. 31-38, 2013.
- FOULADI, P.; NOBAR, R. S. D.; MAHERI SIS, N.; AHMADZADE, A.; AGHDAM SHAHRIAR, H.; GORBANI, A.; AGAJANZADE, A. Effect of choline chloride supplement and canola oil on the Japanese quail livers and bloods cholesterol and triglyceride contents. *Annals of Biological Research*, Gurgaon, v. 3, n. 1, p. 437-444, 2012.
- GALOBART, J.; BARROETA, A. C.; BAUCCELLS, M. D.; CODONY, R.; TERNES, W. Effect of dietary supplementation with rosemary extract and α -tocopheryl acetate on lipid oxidation in eggs enriched with n-3 fatty acids. *Poultry Science*, Champaign, v. 80, n. 10, p. 1496-1505, 2001.
- GRIFFIN, H. D. Manipulation of egg yolk cholesterol: a physiologist's view. *World's Poultry Science Journal*, Cambridge, v. 48, n. 2, p. 101-112, 1992.
- GÜÇLÜ, B. K.; UYANIK, F.; İŞCAN, K. M. Effects of dietary oil sources on egg quality, fatty acid composition of eggs and blood lipids in laying quail. *South African Journal of Animal Science*, Cape Town, v. 38, n. 2, p. 91-100, 2008.
- GÜL, M.; YÖRÜK, M. A.; AKSU, T.; KAYNAR, Ö. The effect of different levels of canola oil on performance, egg shell quality and fatty acid composition of laying hens. *International Journal of Poultry Science*, Faisalabad, v. 11, n. 12, p. 769-776, 2012.
- HARGIS, P. S. Modifying egg yolk cholesterol in the domestic fowl - a review. *World's Poultry Science Journal*, Cambridge, v. 44, n. 1, p. 17-29, 1988.
- HARTMAN, L.; LAGO, R. C. A. Rapid preparation of fatty acid methyl esters from lipids. *Journal: Laboratory Practice*, London, v. 22, n. 8, p. 175-176, 1973.
- HESS, J. B.; COWNS, K. M.; BILGILI, S. F. Selenium nutrition and poultry meat quality. In: ANNUAL SYMPOSIUM NUTRITIONAL BIOTECHNOLOGY IN THE FEED AND FOOD INDUSTRIES, 19., 2003, Nottingham, UK. *Proceeding...* Nottingham: Nottingham University Press, 2003. p. 107-112.
- JONES, P. J. H.; LICHTENSTEIN, A. H.; SCHAEFER, E. J.; NAMCHUK, G. L. Effect of dietary fat selection on plasma cholesterol synthesis in older, moderately hypercholesterolemic humans. *Arteriosclerosis, Thrombosis, and Vascular Biology*, Dallas, v. 14, n. 4, p. 542-548, 1994.
- KAZ'MIERSKA, M.; JAROSZ, B.; KORZENIOWSKA, M.; TRZISZKA, T.; DOBRZAN'SKI, Z. Comparative analysis of fatty acid profile and cholesterol content of egg yolks of different bird species. *Polish Journal of Food and Nutrition Sciences*, Olsztyn, v. 14, p. 69-73, 2005. Supplement 1.
- KÜÇÜKERSAN, K.; YEŞİLBAĞ, D.; KÜÇÜKERSAN, S. Influence of different dietary oil sources on performance and cholesterol content of egg yolk in laying hens. *Journal Biology Environmental Science*, New York, v. 4, n. 12, p. 117-122, 2010.

- LEESON, S.; SUMMERS, J. D. *Commercial poultry nutrition*. 3th ed. Guelph: Nottingham University Press, 2005. 398 p.
- LEWIS, N. M.; SEBURG, S.; FLANAGAN, N. L. Enriched eggs as a source of n-3 polyunsaturated fatty acids for humans. *Poultry Science*, Champaign, v. 79, n. 7, p. 971-974, 2000.
- MAZALLI, M. R.; SALDANHA, T.; BRAGAGNOLO, N. Determinação de colesterol em ovos: comparação entre um método enzimático e um método por cromatografia líquida de alta eficiência. *Revista Instituto Adolfo Lutz*, São Paulo, v. 62, n. 1, p. 49-54, 2003.
- MAZALLI, M. R.; FARIA, D. E.; SALVADOR, D.; ITO, D. T. A comparison of the feeding value of different sources of fat for laying hens: 2. Lipid, cholesterol, and vitamin E profiles of egg yolk. *Journal of Applied Poultry Research*, Oxford, v. 13, n. 2, p. 280-290, 2004.
- MICHALSKA, E.; STĘPIŃSKA, M. Egg yolk HDL-cholesterol content and its relation to total plasma and yolk cholesterol and some production traits in three lines of Japanese quail. *Animal Science Papers Reports*, Magdalenka, v. 2, n. 14, p. 141-146, 1996.
- MILINSK, M. C.; GOMES, A. E.; MURAKAMI, S. T. M.; MATSUSHITA, M.; SOUZA, N. E. Fatty acid profile of egg yolk lipids from hens fed diets rich in n-3 fatty acids. *Food Chemistry*, Whiteknights, v. 83, n. 2, p. 287-292, 2003.
- NABER, E. C. The cholesterol problem, the egg and lipid metabolism in the laying hen. *Poultry Science*, Champaign, v. 55, n. 1, p. 14-30, 1976.
- NOWACZEWSKI, S.; KONTECKA, H.; WENDA, G. E.; ROSIŃSKI, A.; BEDNARCZYK, M.; KUCHARSKA, A. Eggs quality traits in Japanese quail divergently selected for yolk cholesterol level. *Arch Geflügelk*, Stuttgart, v. 74, n. 2, p. 141-144, 2010.
- PITA, M. C. G.; CARVALHO, P. R.; PIBER NETO, E.; MENDONÇA JUNIOR, C. X. Effect of marine vegetal sources on the hen diets on the PUFAs and PUFAs n-3 in laying hens egg yolk and plasm. *International Journal of Poultry Science*, Faisalabad, v. 9, n. 2, p. 148-151, 2010.
- RASOULPOUR, A.; NOBAKHT, A.; KHODAYI, S.; MANSOUD, N. H. Effects of graded fat/oil on egg production and quality, some biochemical parameters of blood and immunity in laying hens. *Advances in Environmental Biology*, Moorebank-NSW, v. 5, n. 7, p. 1826-1831, 2011.
- REAVEN, P. D.; GRASSE, B. J.; TRIBBLE, D. L. Effect of linoleate-enriched and oleate-enriched diets in combination with α -tocopherol on the susceptibility of LDL and LDL subfractions to oxidative modification in humans. *Arteriosclerosis, Thrombosis, and Vascular Biology*, Dallas, v. 14, n. 4, p. 557-566, 1994.
- ROWGHANI, E.; ARAB, M.; NAZIFI, S.; BAKHTIARI, Z. Effect of canola oil on cholesterol and fatty acid composition of egg-yolk of laying hens. *International Journal of Poultry Science*, Faisalabad, v. 6, n. 2, p. 111-114, 2007.
- SCHRAUZER, G. N. Anticarcinogenic effects of selenium. *Cellular and Molecular Life Sciences*, Basel, v. 57, n. 13-14, p. 1864-1873, 2000.
- SHAFEY, T. M.; DINGLE, J. G.; MCDONALD, M. W. Comparison between wheat, triticale, rye, soybean oil and strain of laying bird on the production and cholesterol and fatty acid content of eggs. *British Poultry Science*, Edinburgh, v. 33, n. 2, p. 339-346, 1999.
- SILVA, J. H. V.; ALBINO, L. F. T.; GODÓI, M. J. S. Effect of annatto extract oil on the egg yolk colour. *Revista Brasileira de Zootecnia*, Viçosa, MG, v. 29, n. 5, p. 1435-1439, 2000.
- SOARES, H. F.; ITO, M. K. O ácido graxo monoinsaturado do abacate no controle das dislipidemias. *Revista de Ciências Médicas*, Campinas, v. 9, n. 2, p. 47-51, 2000.
- SUN, J. H.; KANG, G. H.; JEONG, J. Y.; YANG, H. S.; HA, Y. L.; PARK, G. B.; JOO, S. T. Effect of dietary conjugated linoleic acid on lipid characteristics of egg yolk. *Asian Australasian Journal of Animal Sciences*, Seoul, v. 16, n. 8, p. 1165-1170, 2003.