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The Effect of Different Levels of Sesame Oil on Productive Performance, Egg Yolk and Blood Serum Lipid Profile in Laying Hens

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Abstract

Addition of sesame oil into layer diets has been proved to enrich the proportion of polyunsaturated fatty acids in animal's products. In this study, the effects of different levels of sesame oil in the diets on the performance, egg yolk and blood serum lipid profile of Isa Brown laying hens were investigated. A total of 96 layers were assigned into 4 groups to receive either 1 of 4 different diets contained 0.0%, 1.5%, 3.0% and 4.5% sesame oil, respectively. Sample of 12 eggs obtained from each groups were assessed for egg quality. The egg yolk fatty acid profile was determined with gas chromatography. Results revealed that the higher levels of sesame oil in the diet decreased egg production, egg weight, and egg yolk color except feed conversion ratio. In addition, supplementation of sesame oil increased the flow index of the eggs and the Haugh unite. The egg yolk lipid profile was not significantly different in the sesame oil fed groups, whereas, compare to control, it decreased the level of cholesterol. The blood serum lipid profile decreased in the sesame oil groups compare to control group. Meanwhile, monoacildigliserol also decreased in the sesame addition groups. In parallel with increasing levels of sesame oil, monounsaturated fatty acid (oleic acid) in the egg yolks significantly increased compared to the control (37.00%, 42.89%, 42.20% and 43.48%, respectively). It can be implied that sesame oil supplementation into the laying hens diet is necessary to produce monounsaturated fatty acid (MUFA) enriched eggs.

Keywords

Sesame Oil, Egg Yolk Lipid Profile, Performance, Laying Hens

1. Introduction

Addition of vegetable oil into layers diet has been world widely used in order to increase absorption of fat so-

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luble vitamin (A, D, E, K), minerals and to enhance egg production. On the other hand, this addition is also for the seeking of production of functional nutrient enriched egg in poultry industry, in which, it is desired to reduce cholesterol concentrations in egg for those consumers who need to lower their dietary cholesterol intake. It has been known that the supplementation of vegetable oil provided essential fatty acid precursor, which cannot be synthesized by animal or human. It has been proved that the supplemental oils significantly altered egg yolk lipid profiles or lipid composition ratios [1] [2]. These essential fatty acids are commonly found in some animal oils such as fish oil and vegetable oils such as sesame oil, canola oil, soybean oil etc. Sesame seed was described to have originated from Africa and it is thought to be the oldest oil seed known to man. Sesame oil is very resistant to rancidity due to the presence of natural anti-oxidants such as sesamolin, sesamin, and sesamol. It is therefore useful in increasing the shelter life of margarine and other vegetable oil products. Sesame oil is plant-derived oil rich in oleic acid (53.8%) which is a monounsaturated fatty acid [3]. However, it also contains a significant amount of linoleic (22.1%) and alpha-linoleic fatty acids. Sesame oil has been considered as a perfect balance of n-6 to n-3 polyunsaturated fatty acids (PUFAs), in which 54.68% of this ratio has been found matched human requirements. The egg yolk lipid profiles have been shown to vary depending on number of factors including genetic selection of the laying hen [4]. However, the results of genetic selection for lower cholesterol concentration in eggs also associated with lower egg production [5]. In order to modify egg yolk cholesterol and fatty acids contents, the nutritional strategies or dietary manipulations have been carried out, in which supplementary of different dietary oils supplementation in layer diets such as flaxseed, perilla oil, fish oil, vegetable oil have given some promising effects. Addition of sesame oil was also considered in several studies because of its rich in omega-3 fatty acids [6]-[8]. As the result, this supplement increased the amount of omega-3 in the form of alpha-linoleic fatty acid in egg, animal organs and tissue [9]. In addition, it also resulted in better proportion of n-3 PUFAs [10]. The present of omega-3 in the animal diet was also believed to improve the taste of animal's products (meat and egg) [11]. However, there is still little known how different levels of sesame oil would alter egg yolk lipid profile and the composition of monounsaturated fatty acids (MUFAs) in the egg yolk. Therefore, the aim of this study was to investigated the effect of different levels of sesame oil (1.5%, 3.0% and 4.5%) in layer diet on laying performance such as egg production, egg weight, feed intake, feed conversion ratio, egg quality, egg yolk lipid profile and composition of MUFAs.

2. Materials and Methods

2.1. Experimental Materials and Diets

The experiment was conducted on 96 laying hens of Isa Brown at 40 weeks of age when experimental birds reached the peak of egg production. All birds were selected to obtain uniformity of 93% then randomly allocated in six replicates groups caging (50 cm × 46 cm × 46 cm) consist of 4 birds per cage to receive either one of four diets containing 0.0%, 1.5%, 3.0% and 4.5% sesame oil, respectively. All birds were cared in comply with the MARD Vietnam standard (2010) [12]. The basal diet (**Table 1**) was formulated to meet the NRC 1994 recommendations [13]. Basal diet containing 1.5% soybean oil was considered as a control group. In the experimental groups, different levels (0.0%, 1.5%, 3.0% and 4.5% of sesame oil) were supplemented into the basal diet. The fatty acid composition of sesame oil used in the current study is presented in **Table 2**. The diets were stored in cold conditions (minus 4˚C to 0˚C). Thus, addition of antioxidant to prevent oil degradation was not required. The metabolizable energy level (ME) of the feeds was calculated by the following formula which is described in Vietnamese National Standards (TCVN 8762/2011) [14]:

$$\mathrm{ME}\left(\mathrm{MJ/kg}\right) = 0.1551\times\%\ \text{crude protein} + 0.3431\times\%\ \text{crude fat} + 0.1669\times\%\ \text{starch} + 0.1301\times\%\ \text{sugar}.$$

During the 3 months of experiment, the hens were fed ad libitum once daily at 07:30 with free access to water. The hens were housed in cages that luminated 17 hours per day. The ethical approval of using animal in experiment was not applicable in this study since there are not any of such regulations in effect in Vietnam.

2.2. Egg Quality Analysis and Collection of Samples

The composition of the feed samples were analyzed for dry matter (DM), Crude protein (CP), Crude fat (CF), Neutral Detergent Fiber (NDF) and ash contents [15]. Feed intake and egg production were recorded daily; egg weight was measured weekly. Before the determination of egg weight, a sample of 12 eggs from each experimental group was stored for 24 hours at room temperature. The feed conversion ratio was expressed as the kilo-

Table 1. Chemical compositions and compound of experimental diets (%).

Items	Control	1.5% sesame oil	3.0% sesame oil	4.5% sesame oil
Yellow corn	52.00	52.00	52.00	46.00
Soybean meal	21.59	21.59	22.50	22.50
Barley	2.00	2.00	-	2.00
Wheat bran	10.22	10.22	9.92	12.42
Calcium carbonate	7.95	7.95	7.95	7.95
Sesame oil	-	1.50	3.00	4.50
Soybean oil	1.50	-	-	-
Full fat soybean	2.50	2.50	2.50	2.50
DCP	1.32	1.32	1.32	1.32
Salt	0.40	0.40	0.29	0.29
Vit.+Min.	0.20	0.20	0.20	0.20
D-L Methionine	0.12	0.12	0.12	0.12
Antioxidant	0.20	0.20	0.20	0.20
ME, kcal/kg**	2762	2760	2868	2876
Crude Protein (%)	16.54	16.63	16.32	16.75
Ether Extract (%)	11.53	11.18	11.41	12.35
Crude ash (%)	10.93	9.84	11.42	10.69
Dry matter (%)	87.16	87.36	87.56	86.48

*Each kilogram of feed: 12,000 IU Vitamin A, 2500.00 IU Vitamin D3, 30.000 mg Vitamin E, 34.000 mg Vitamin K, 3.000 mg Vitamin B1, 6.000 mg Vitamin B2, 30.000 mg Nicotinamide, 10.000 mg Cal.-D-Palm, 5.000 mg Vitamin B6, 15 mg Vitamin B12, 1.000 mg Folic Acid, 50 mg D-Biotin, 300.000 mg Cholin, 50.000 mg Vitamin C, 80.000 mg Manganese (Mn), 60.000 mg Iron (Fe), 60.000 mg Zinc (Zn), 5.000 mg Copper (Cu), 2.000 mg Iodine (I), 500 mg Cobalt (Co), 150 mg Selenium (Se), 1000 mg Antioxidan. **Calculated analysis. DCP: Dicalcium phosphate.

gram of feed consumed per kilogram of egg produced. Another 12 egg samples were randomly collected from each experimental group every month in order to assess egg quality parameters. Egg quality parameters include shape index, shell strength, shell thickness, albumen index, yolk index, yolk color (Yolk Colour Fan, the CIE standard colorimetric system, F. Hoffman-La Roche Ltd., Basel, Switzerland) and Haugh unit. These parameters were calculated using following formulas as summarized by Ergun *et al.* (1987) [16]. Egg quality parameters were assessed using the following formulas:

$$\text{Shape index}(100) = \left[\text{egg width (cm)} / \text{egg length (cm)}\right] \times 100.$$

Shell strength (kg/cm × cm) determined by using a machine with a spiral pressure system; Shell thickness (mm) was determined in 3 different parts (upper and lower ends and middle) using a micrometer.

Albumen index (%) = [(albumen height(mm)/average of albumen length (mm) and albumen width (mm)] × 100; Yolk index (%) = [(yolk height (mm)/yolk diameter (mm)] × 100; Yolk color was determined using commercially available yolk colour fan according to the CIE standard colorimetric system; Haugh unit = 100 × log (H+7.57 − 1.7 × W0.37), where H = albumen height (mm) and W = egg weight (g).

Lipid oxidation was assessed on the basis of the MDA (Malondialdehit) formed during refrigerated storage. MDA was the compound used as an index of lipid peroxidation [17].

2.3. Fatty Acids and Blood Analysis

Blood samples were collected by venipuncture from the sub-wing vein and placed in non-additives blood collection tubes to produce serum from a sub-sample of 5 randomly selected laying hens from each treatment groups

Table 2. Fatty acids composition (%) of sesame oil included in the diets of laying hen.

Numeric name	Common name	Fatty acids content (%)
C12:0	Lauric acid	0.016
C14:0	Myristic acid	0.64
C15:0	None	0.04
C16:0	Palmitic acid	18.72
C17:0	Margaric acid	0.14
C18:0	Stearic acid	2.69
C20:0	Arachidic acid	0.24
C21:0	None	0.01
C22:0	Behenic acid	0.22
C23:0	None	0.02
C24:0	Lignoceric acid	0.11
C14:1	Myristoleic acid	-
C15:1	None	-
C16:1	Palmitoleic acid	0.49
C17:1	None	0.09
C18:1 n9	Oleic acid	20.34
C20:1 n9	Gadoleic acid	0.26
C22:1 n9	Erucic acid	-
C24:1 n9	Nervonic acid	0.06
C18:3 n3	Alpha linolenic acid	0.72
C20:3 n3	None	0.01
C20:5 n3	Eicosapentenoic acid (EPA)	0.20
C22:6 n3	Docosahexaenoic acid (DHA)	0.06
C18:2 n6	Linoleic acid	55.03
C18:3 n6	Gamma linolenic acid	0.05
C20:2 n6	11, 14 – Eicosadienoic acid	0.06
C22:2 n6	13, 16 – Docosadienoic acid	-
Total of saturated fatty acids		22.84
Total of mono unsaturated fatty acids		21.03
Total of polyunsaturated fatty acids		56.13
Total of omega-3 fatty acids		0.99
Total of omega-6 fatty acids		54.14
Total of omega-6/total omega-3 fatty acids ratio		54.68

at the end of experiment period. Serum was separated by centrifugation at 3000 rpm for 10 minutes and analyzed for blood lipid a profile includes: Hydrocarbons, Triachyleglyserol, free fatty acids, cholesterol, mono-

diacilgliserol and pola lipid.

Fatty acids were analyzed by gas chromatography at the Institute of Biotechnology and Food Technology, Hanoi Vietnam. Fat (0.15 to 0.20 g) extracted by the ether method from each sample (total of two), was saponified with 5 ml NaOH with methanol in a water bath for 10 minutes. Previously, at this mixture 5 mL BF3-methanol was added and the extraction process was refluxed for 2 minutes. After adding 5 mL heptane to the mixture, it was boiled again for 1 minute. The content of this mixture was transferred into 25 mL volumetric flasks and the volume was adjusted with saturated NaCl to 25 ml. 1 mL of the heptane phase from upper layer of the volumetric flasks was used to determine the fatty acids composition. Fatty acids were analyzed with gas chromatography (Agilent 6890N, Hewlett Packard, Palo Alto, CA) with a capillary column (supel covax 10, 60 m × 0.25 mm ID). The chromatographic conditions were: detector temperature 280˚C; injector temperature 200˚C; initial column temperature 100˚C for 8 min, programmed to increase at a rate of 5˚C per five minutes up to 200˚C and then at 4˚C per minute up to the final temperature of 250˚C. The helium carrier gas flow was set at 1.2 mL/min, hydrogen at 30 mL/min and air at 300 mL/min. Injection of the 1-µL samples was performed with a split ratio of 20:1. Identification of individual fatty acids was based on comparisons of retention times of unknown peaks to authentic fatty acid methyl ester standards.

2.4. Statistical Analysis

Differences between groups were analyzed with one-way analysis of variance (ANOVA) by using the statistical package SPSS for Windows (1999), version 10.0. Significant means were subjected to a multiple comparison test (Duncan) at alpha level 0.01 and 0.05.

3. Results and Discussion

3.1. Laying Performance of Experiment Birds

Additional of oil to balance the energy level in the diet has been widely practiced in commercial animal feeds industry. In this study, the expectation of sesame oil is to alter the egg nutrient composition toward “healthier lipid profile” and should maintain animal performance. Laying performance of laying hens depends on several factors includes breed, live weight, age and energy level of the ration. In our study, laying performance of experiment birds such as egg production, egg weight; feed intake were decreased with the supplementation of different dietary sesame oil levels ($p < 0.05$) egg but not for the feed conversion ratio (**Table 3**). The lower feed intake of birds which seen in birds fed diet contained 3.0% and 4.5% sesame oil might be due to the higher concentration of energy level in these diet compared to diet contained 1.5% sesame oil and in the control ($p < 0.05$), although feed conversion ratio was not affected by supplementing sesame oil. Our study finding was supported by the findings from Cherian, 2008 [18] when study on supplementation of n-3 fatty acid-rich oil into diet in Cobb laying hens in which the results also indicated that the different levels of oil supplemented in the diet also negatively affected layers performance. When increase the levels of oil supplemented in the diet this also resulted in increased level of energy level of the ration thus egg production, egg weight, and feed intake decreased [6]. This can also be explained that the shortage of linoleic acid in the diet might be the limiting factor contributed to the decrease in egg weight [19] [20]. Supplementation of different oils levels into diet had decreased feed intake [9], egg weight [21], feed conversion was not affected by supplementation [22] [23]. The data obtained from this study were consistent with some research findings that reported a decrease in egg weight [18] [19] [23] [24] when sesame oil was supplemented into diet for laying hens.

3.2. The Effect of Sesame Oil on Egg Quality

The egg quality was expressed in several criteria such as shape index, breaking strength, shell thickness, shell weight, yolk color, yellow index, flow index, Haugh unit, egg yolk lipid profile and egg yolk fatty acid composition. These parameters depend greatly on diet composition rather than just energy levels. It can be seen from our study that increased levels of sesame oil in the diet did not affect the shape index, breaking strength, shell thickness; shell weight and yolk color index. However, supplementation of sesame oil increased the flow index of the eggs and the Haugh unite (**Table 4**). These findings of our study were in contrast with some other findings in which it was reported that these parameters were improved with the supplementation of different dietary oil levels [25].

Egg yolk colour in the experimental groups decreased by approximately 20% compared to the control ($p < 0.05$) and this was similar among the groups. This difference is thought to be related to the amount of xanthophylls in the ration [26].

The egg yolk and blood serum lipid profile values are presented in **Table 5** and **Table 6**. The egg yolk lipid profile was not different among treatment groups in terms of hydrocarbons, although hydrocarbons in serum lipid profile increased in sesame oil fed groups. No statistical differences were observed among groups for free fatty acids, although free fatty acids were identified as being lower in the 4.5% sesame oil group compared to the control group. Identified blood serum and egg yolk cholesterol in egg yolks from the sesame oil groups was observed to be higher than the control group. In addition to the different oil levels, the type of supplement oils also affects cholesterol levels in the egg yolk [8]-[24]. The amount of egg yolk cholesterol Increased because of the decrease in lipogenesis with fatty acids, although the cholesterol level increased in the liver. Therefore, it is increased in the egg yolk [1].

Table 3. Egg production, egg weight, feed intake and feed conversion ratio of trial groups.

Groups	EP (%)	EW (g)	FI (g)	FCR (kg)
Control	83.68^{a}	65.21^{a}	126.76^{a}	1.53
1.5% sesame oil	80.22^{a}	64.09^{a}	127.06^{a}	1.56
3.0% sesame oil	78.18^{ab}	62.22^{b}	121.05^{b}	1.48
4.5% sesame oil	74.77^{b}	61.58^{b}	120.09^{b}	1.50
SEM	2.76	0.59	3.00	0.06

a, b, c: Means with different superscripts in the same column differs significantly $p < 0.05$. EP: Egg Production, EW: Egg Weight, FC: Feed Intake, FCR: Feed Conversion Ratio.

Table 4. The effects of sesame oil on egg quality.

Groups	Control	1.5% sesame oil	3.0% sesame oil	4.5% sesame oil	SEM
Quality criteria					
SI (%)	74.53	73.54	74.65	74.33	0.84
BS (kg/cm^{2})	2.19	1.58	2.01	1.85	0.17
ST(mm)	0.39	0.38	0.38	0.38	0.01
SW(g)	7.88	7.90	7.62	7.66	0.17
YC	8.01^{a}	6.64^{ab}	6.65^{ab}	6.58^{ab}	0.14
YI(%)	38.66	39.70	41.20	40.99	0.42
FI(%)	7.98^{b}	9.09^{a}	8.56^{b}	9.10^{a}	0.40
HU	79.00^{b}	82.69^{ab}	81.08a^{b}	83.32^{a}	1.48

a, b, c: Means with different superscripts each column differs significantly $p < 0.05$. SI: Shape index; BS: Breaking Strength; ST: Shell Thickness; SW: Shell Weight; YC: Yolk Colour; YI: Yellow Index; FI:Flow Index; HU: Haugh Unit.

Table 5. The egg yolk lipid profile (%).

Groups	HC	TAG	FFA	Col	M-DAG	PL
Control	9.19	65.27^{a}	3.86	15.20^{b}	5.14	1.01
1.5% sesame oil	9.55	63.44^{b}	3.99	16.66^{a}	5.45	1.06
3.0% sesame oil	9.42	64.48^{ab}	3.33	17.11^{a}	5.01	0.82
4.5% sesame oil	9.09	63.94^{b}	3.35	17.70^{a}	5.24	0.81
SEM	0.32	0.42	0.25	0.38	0.28	0.09

a, b, c: Means with different superscripts in the same column differs significantly $p < 0.05$. HC: Hydrocarbons; TAG: Triachyleglyserol; FFA: Free Fatty Acids; Col: Cholesterol; M-DAG: Mono-Diaçilgliserol; PL: Polar Lipids.

Differences level of feed oils on egg yolk cholesterol levels are thought to be related to the genetic structure of chickens with factors connected to the diet. The supplement of sesame oil had significantly altered fatty acids composition of egg yolk in this study. Especially, supplement of sesame oil had significantly increase level of Linoleic acid ($p < 0.05$) but not that of Oleic acid, whereas, it decreased the alpha-linoleic acid in the egg yolk. The lowest amount of oleic acid was determined in the control group (37.0%), while the highest amount was in the group with 4.5% sesame oil (43.48%). Conversely, the highest amount of linoleic and alpha-linoleic acid (20% 22% and 0.90%, respectively) were found in the control group, the lowest amount of linoleic and alpha-linoleic acid were also found in the group with 4.5% sesame oil. No statistical differences were observed among groups for Eicosapentaenoic acid (EPA) and Docosapentaenoic acid (DHA), as well as total Saturate Fatty Acids (SFA). Polyunsaturated Fatty Acids (PUFA) in the groups with sesame oil was decreased ($p < 0.05$) compared to the control. It can be observed that the levels of n-3 PUFAs in diets did not affect the egg yolk fatty acid profile (PUFAs, MUFAs and SFAs) [7]-[18]. It is readily that egg yolk lipid profile depends on the type and levels of oil supplemented. It was reported that egg yolk MUFAs level was higher when chickens fed with tallow oil and olive oil. The similar trend also observed for egg yolk PUFA level when soybean oil and flax seed oil were supplemented [21]. An increase in the PUFA of egg yolk was believed to be associated to the high level of linoleic acid in soybean and flaxseed oil [22]. In our study, it was determined that the amounts docosapentanoic acid (DPA, C22:5n-3) and docosahexanoic acid (DHA, C22:6n-3) were highest in the group with 4.50% sesame oil (1.50%) and lowest in the control (0.90%) (**Table 7**) [25]. It can be explained by the different of oil profile

Table 6. The blood serum lipid profile (%).

Groups	HC	TAG	FFA	Col	M-DAG	PL
Control	17.86^{c}	45.67^{a}	4.29^{bc}	20.11^{ab}	2.20^{a}	9.89^{b}
1.5%	22.09^{b}	39.32^{b}	3.80^{c}	20.58^{a}	1.76^{b}	12.00^{a}
3.0%	24.11^{b}	37.90^{b}	5.57^{ab}	19.30^{bc}	1.70^{b}	11.96^{a}
4.5%	27.29^{a}	34.66^{c}	6.00^{a}	18.66^{c}	1.72^{b}	11.42^{ab}
SEM	0.93	0.86	0.44	0.33	0.15	0.50

a, b, c: Means with different superscripts each columns differ significantly $p < 0.05$. HC: Hydrocarbons; TAG: Triachyleglyserol; FFA: Free Fatty Acids; Col: Cholesterol; M-DAG: Mono-Diacilgliserol; PL: polar lipid.

Table 7. The effects of sesame oil on egg yolk fatty acid composition (%).

Fatty acids	Control	1.5% sesame oil	3.0% sesame oil	4.5% sesame oil	SEM
C14:0 (miristic acid)	0.26^{b}	0.32^{a}	0.28^{ab}	0.26^{b}	0.299
C16:1 T7 (palmitoleic acid)	2.18^{ab}	2.33^{a}	1.88^{b}	1.25^{c}	0.103
C18:1 T9 (oleic acid)	37.00^{b}	42.89^{a}	42.20^{a}	43.48^{a}	0.528
C18:2 T6 (linoleic acid)	20.22^{a}	13.79^{b}	14.11^{b}	14.22^{b}	0.788
C18:3 T3 (alpha-linolenic acid)	0.90^{a}	0.52^{c}	0.66^{b}	0.69^{b}	0.055
C20:5 T3 (EPA)	0.05	0.04	0.03	0.03	0.006
C22:5 T3 (DPA)	0.10	0.11	0.12	0.11	0.014
C22:6 T3 (DHA)	0.90^{c}	0.83^{c}	1.14^{b}	1.50^{a}	0.050
GSFA	34.55	33.88	34.59	33.46	1.033
GMUFA	40.0^{b}	47.58^{a}	46.21^{a}	46.6^{a}	0.541
GPUFA	24.51^{a}	17.72^{b}	18.32^{b}	19.95^{b}	0.948
GT6	22.56^{a}	16.22^{b}	16.37^{b}	16.62^{b}	0.859
GT3	1.95^{b}	1.50^{c}	1.95^{b}	2.33^{a}	0.097

a, b, c: Means with different superscripts in the same column differs significantly $p < 0.05$.

in the diet [26]. In this study, a high amount of MUFA and a low amount of PUFA were observed in the experimental group. This explained by the differences in the fatty acid profile of sesame oil [21] [27]-[30].

4. Conclusion

Our results indicated that increasing levels of sesame oil in layer diet had negative effect on egg production, egg weight, feed intake and yolk color. Nevertheless, it is important in producing monounsaturated fatty acid (MUFAs) enrich eggs with lower cholesterol levels. Thus 4.5% inclusion of sesame oil could be used in commercial production for healthier food demand by consumers.

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