Modification of egg yolk fatty acids profile by using different oil sources

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Oils and fats are usually added to the diet of poultry to enhance the energy density to produce energy-rich formulations. In order to ensure adequate levels of linoleic acid and to improve palatability and reduce the dustiness of diets, all poultry diets require a minimum of 1% added fat, regardless of other economic or nutritional considerations.

It was shown that there was a different constitution in terms of the structure of fatty acids (FAs). Fatty acids contain carbon, oxygen and hydrogen, and are classified as saturated FAs or SFAs, monounsaturated FAs or MUFAs, or polyunsaturated FAs or PUFAs.

Animal fat contains palmitic acid as a long-chain (LC) SFA, except for fish oil; vegetable oils contain high quantities of long-chain unsaturated FAs. Several studies have shown better utilisation of unsaturated fats with a higher metabolisable energy compared with saturated fats. Rat and broiler studies have shown that unsaturated vegetable oils produce lower faecal energy losses and, consequently, higher ME than animal fats.

In addition, laying hen studies have shown that unsaturated vegetable oils have higher energy levels than saturated animal fats. Oil supplements are added to layer hen rations to increase the absorption of fat-soluble vitamins and to enhance egg yield and weight.

In recent years, consumer demands for more healthy food supported the interest in modifying the FA profile of eggs. Omega-3 FAs are essential for normal growth and development and play important roles in the prevention and treatment of coronary heart disease (CRD), hypertension, inflammatory autoimmune disorders and cancer. However, since the consumption of fish – the richest dietary source of LC n-3 PUFA in diets – is low, intakes of LC n-3 PUFA are low and suboptimal.

Researchers agree that the optimal ratio of omega-6 to omega-3 should not exceed 2:1 to 4:1. The dietary imbalance in FAs (excessive omega-6 and insufficient omega-3) is an underlying cause of many chronic diseases, including cardiovascular disease, cancer, inflammatory diseases, autoimmune diseases and many physiological disturbances.

Fish oil contains unsaturated FAs with long omega-3 chains (LC-n-3 PUFA), eicosapentaenoic acid (EPA20:5n-3) and docosahexaenoic acid (DHA22:6n-3) that improve health-related factors in humans and animals. High intakes of long-chain n-3 PUFAs are associated with a decreased risk of cardiovascular disease. Many studies are directed towards the manipulation of the FA composition of broiler chicks and laying hens in order to increase the n-3 PUFA content and decrease the n-6/n-3 ratio in poultry meat and eggs. It is possible to modify the FA profile of the yolk by changing the lipid sources of the hen diet. Nutritional manipulation of hen diets to include sources of PUFA n-3 promotes deposition of these nutrients in egg yolk.

The objective of this study was to compare the effects of different oil sources on performance and the FA composition of egg yolk in laying hens.

Animals and experimental design

For the study, 72 23-week-old laying hens (Tetra-SL) were divided randomly into six dietary treatments (four replicates and three birds per replication). Each three-bird group was housed in one cage (40 x 40 x 50cm). Environmental temperature was set at 22°C.

A regime of 14 hours of constant lighting (15 lux) and continuous ventilation were provided. All the birds were kept under uniform management conditions throughout the experimental period. Experimental diets included:

- Control (no oil).
- 3% fish oil.
- 3% olive oil.
- 3% grapeseed oil.
- 3% canola oil.
- 3% soya bean oil.

Diets were formulated according to the recommendations of the National Research Council. Feed and water were provided *ad libitum* throughout the experiment. Diets were modulated as isoenergetic and isonitrogenous. At the end of the trial (ninth week), seven eggs were randomly selected from each group to determine the FA profile of the egg yolk.

Performance record

Egg production, egg weight, feed intake, egg mass and feed conversion ratio of each pen were recorded weekly.

Fatty acid content

The FA composition of the dietary oil and yolk samples was determined by gas chromatography according to the method described by Metcalfe *et al.* The FA content was determined using a gas chromatograph (Model 4600, Unicam, Cambridge, England) equipped with a BPX70 fused silica capillary column and a flame ionisation detector (Unicam, Cambridge, England). The column head pressure of the carrier gas (helium) was 20psi and the sample volume injected was 0,2µL. Pentadecanoic acid (Sigma-Aldrich, St. Louis, Unite States) was used as internal standard. The FAs were identified by matching their retention times with those of their corresponding standards.

Statistical analyses

Data was analysed in a completely randomised design using SAS software (Version 8.0, SAS Institute, Cary, United States). Significant differences among treatments were determined according to the general linear model (GLM) procedure. Means were compared by using Duncan's multiple-range test and significance was determined when the p-value was less than 0,05.

The following model was used: $X_{ij} = \mu + \tau_j + \varepsilon_{ij}$ where X_{ij} is the observation of jth treatment and ith pen; μ is the overall means of the sampled observation; τ_j is the effect of treatment; and ε_{ij} is the experimental error component.

Table 1: Fatty acid composition (%) of oils included in the diets of laying hens.

Fatty acids[1]	Fish oil	Olive oil	Grapeseed oil	Canola oil	Soya bean oil
C14	5,61	0,10	0	0,08	0,34
C16:0	27,43	14,18	7,28	6,21	15,57
C16:1	7,97	0,89	0,07	0,17	0
C18:0	4,44	4,41	3,99	2,67	3,97
C18:1	32,84	68,88	20,62	57,86	23,50
C18:2	1,59	10,59	67,80	27,45	55,53
C18:3	0,23	0,84	0,24	5,56	1,10
C20:4	0,09	0,09	0	0	0
EPA	5,59	0	0	0	0
DHA	14,2	0	0	0	0
SFA	37,48	18,70	11,27	8,97	19,88
MUFA	40,81	69,77	20,69	58,02	23,5
PUFA	21,71	11,53	68,04	33,01	56,62
n-3	20,02	0,84	0,24	5,56	1,1
n-6	1,69	10,69	67,8	27,45	55,53
n-6/n-3	0,08	12,69	283,11	4,94	50,67

1 C14:0 = Myristic acid; C16:0 = Palmitic acid; C16:1 = Palmitoleic acid; C18:0 = Stearic acid; C18:1 = Oleic acid; C18:2 = Linoleic acid; C18:3 = Linolenic acid; C20:4 = Arachidonic acid; EPA = Eicosapentaenoic acid; DHA = Docosahexaenoic acid; SFA = Saturated fatty acid; MUFA = Monounsaturated fatty acid; PUFA = Polyunsaturated fatty acid; n6/n3 = Ratio of n-6 to n-3 PUFA.

Composition of different oils

The FA composition of the fish, olive, grapeseed, canola and soya bean oil is shown in *Table 1*. The fish oil was a rich source of LC PUFA, n-3 and contained EPA (5,59 %) and DHA (14,20%). The fish oil had the highest amount of C14:0, C16:0, C18:0, EPA, DHA, SFA and total omega-3 FAs and the lowest amount of C18:2. Olive and canola oil were rich in C18:1 and had 69 and 58% of this FA, respectively. The grapeseed and soya bean oil were rich in C18:2 and had 68 and 55,5% of this FA, respectively.

Performance

The effects of different sources of oil on laying hen performance are summarised in *Table 2*. At the end of the experiment, the hens' egg production, feed intake, feed conversion ratio, egg weight and egg mass changes were not significantly affected by treatments (p > 0,05).

Table 2: Performance of laying hens in response to experimental diets.

Experimental diets	Egg production (%)	Feed intake (g/day)	Feed conversion ratio	Egg mass (g/day)	Egg weight (g)
Control	98,06	119,61	2,06	58,07	59,21
Fish oil	98,51	117,41	2,03	57,83	58,7
Olive oil	98,51	118,31	2,06	57,36	58,22
Grapeseed oil	96,87	117,13	2,1	55,83	57,66
Canola oil	99,26	121,19	2,1	57,63	58,06
Soya bean oil	98,36	117,99	2,02	58,4	59,36
P value	0,295	0,955	0,813	0,568	0,736
SEM	0,249	0,970	0,016	0,345	0,736

Table 3: Fatty acid composition (%) of the egg yolk of laying hens fed different sources of oil.

Fatty acids[1]	Control	Fish oil	Olive oil	Grape seed oil	Canola oil	Soya bean oil	Significance	SEM
C14	0,45^{c}	0,51^{b}	0,58^{a}	0,45^{bc}	0,39^{d}	0,43^{cd}	*	0,016
C16:0	29,25	29,47	29,32	30,20	27,05	26,74	NS	0,372
C16:1	4,47^{a}	4,19^{a}	3,42^{b}	3,13^{bc}	3,16^{bc}	2,90^{c}	*	0,131
C18:0	7,86^{bc}	6,95^{d}	7,63^{c}	8,16^{b}	7,58^{c}	8,95^{a}	*	0,134
C18:1	45,66^{a}	45,86^{a}	47,00^{a}	37,06^{c}	46,99^{a}	41,27^{b}	*	0,837
C18:2	10,21^{d}	8,76^{e}	9,79^{d}	18,72^{a}	12,29^{c}	17,32^{b}	*	0,898
C18:3	0,17^{e}	0,28^{d}	0,37^{c}	0,10^{f}	0,81^{a}	0,55^{b}	*	0,052
C20:4	1,74^{ab}	0,59^{c}	1,52^{b}	1,97^{a}	1,74^{ab}	1,83^{a}	*	0,099
EPA	0,00^{b}	0,18^{a}	0,00^{b}	0,00^{b}	0,00^{b}	0,00^{b}	*	0,014
DHA	0,19^{c}	3,21^{a}	0,37^{b}	0,20^{c}	0,00^{d}	0,00^{d}	*	0,243
SFA	37,56	36,93	37,53	38,82	35,01	36,13	NS	0,342
MUFA	50,13^{a}	50,04^{a}	50,42^{a}	40,19^{c}	50,15^{a}	44,17^{b}	*	0,905
PUFA	12,31^{d}	13,02^{d}	12,05^{d}	20,99^{a}	14,83^{c}	19,70^{b}	*	0,841
n-3	0,36^{e}	3,66^{a}	0,74^{c}	0,30^{e}	0,81^{b}	0,55^{d}	*	0,246
n-6	11,95^{d}	9,35^{e}	11,31^{d}	20,69^{a}	14,03^{c}	19,15^{b}	*	0,965
n-6/n-3	33,52^{b}	2,55^{d}	15,33^{c}	68,61^{a}	17,45^{c}	34,94^{b}	*	4,398

*a-f Different superscripts indicate significant differences (p < 0,05); NS = Not significant; SEM = Standard error of the mean; * = p < 0,01.*
1 EPA = Eicosapentaenoic acid; DHA = Docosahexaenoic acid; SFA = Saturated fatty acid; MUFA = Monounsaturated fatty acid; PUFA = Polyunsaturated fatty acid; n6/n3 = Ratio of n-6 to n-3 PUFA.

Egg yolk fatty acid composition

The effects of different feed sources on the composition of egg yolk FAs is shown in *Table 3*. The FA profile of the egg yolk was significantly affected by the treatments (except C:16 and SFA). As shown in *Table 3*, the values of EPA, DHA and total omega-3 FAs were significantly higher ($p < 0{,}01$) in the egg yolk of laying hens fed fish oil compared to the eggs of other treatments.

Fish oil reduced C18:0, C18:2 (14% less than control group), C20:4 (66% less than control group), total omega-6 FAs and the n-6/n-3 ratio in egg yolk compared to other groups. Egg yolk C18:2 and total omega-6 FAs from birds fed grapeseed oil were significantly higher than those of the other five oils.

Furthermore, canola oil supplementation enhanced the linolenic acid content of egg yolk. In this study, the highest C18:1 concentration was found in the eggs of hens fed olive and canola oil, which were rich in C18:1. Similarly, the C18:2 concentration of the egg yolk was high in the groups fed soya bean and grapeseed oil, which are rich in C18:2. The highest C18:3 concentration of the egg yolk was found in the group fed canola oil, which is also rich in C18:3.

Discussion

Guclu *et al*. reported that quails fed a diet supplemented with sunflower and olive oil produced significantly heavier eggs. In another study, Kucukersan *et al.* reported that supplementing four different kinds of oil sources (sunflower, fish, soya bean and hazelnut oil) at 3% concentration had a significant effect on egg production and egg weight.

All these results are in contrast with the present study. In addition, it is indicated that supplementing fish oil and tallow at the level of 1,5% to the corn/soya bean meal diet may affect egg production performance and egg weight without any adverse effects on body weight.

In the present study, SFA content was not significantly different between the treatments, which are in agreement with other reports. Hens have the ability to synthesise SFA and if the values of their rations decrease, hens can compensate for the lack of these FAs.

Some studies have reported that the DHA and EPA concentrations of the egg yolk of birds fed diets containing fish oil is a reflection of the DHA and EPA concentration of fish oil. Reportedly, inclusion of fish oil in the diet could increase the proportion of n-3 PUFA relative to n-6 PUFA in the tissues of poultry.

There is competition among the enzymes involved in the elongation and desaturation of omega-3 and omega-6 FAs. Delta-6 desaturase is the critical enzyme in these reactions, for which the greatest affinity appears to be conferred by the greatest number of double bonds in the C18 substrate. Hence, using diets rich in omega-3 FAs (e.g. fish oil) reduces the omega-6 FAs content of egg yolk.

In conclusion, the results of the present study demonstrated that different oil sources had varying effects on the FA composition of egg yolk. This is reflected by the FA composition of the oils added to the diet. Based on the results, adding 3% fish oil to the diet of laying hens could increase the DHA and EPA content of egg yolk, with their consequent health benefits.

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