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Effect of dietary fat source on fatty acid profile and lipid oxidation of eggs

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Abstract

This study investigated the effects of supplementary dietary lipid sources on the fatty acid profile and lipid oxidation of eggs. Five isoenergetic (12.6 MJ AME/kg DM) and isonitrogenous (170 g CP/kg DM) diets were formulated, using a control diet (50 : 50 blend of fish- and linseed oil), fish oil, sunflower oil, high oleic acid (HO) sunflower oil and tallow at a 30 g/kg inclusion level. Two hundred individually caged HyLine Silver-Brown hens (20 weeks of age) were randomly allocated to the five dietary treatments (n = 40 hens/treatment). Birds received the experimental diets from 20 weeks of age. At 30 weeks of age, 12 eggs per treatment were randomly selected for analyses of egg yolk fatty acid methyl esters (FAME), thiobarbituric acid reactive substances (TBARS) and peroxide values (PV). Dietary lipid sources affected FAME, TBARS and PV of egg yolk significantly. The fish oil treatment resulted in the highest TBARS (0.27 mg malonaldehyde/kg yolk) and PV (3.96 milli-equivalent peroxide/kg fat) whereas the HO sunflower oil resulted in the lowest TBARS (0.13 mg malonaldehyde/kg yolk) and PV (2.77 milli-equivalent peroxide/kg fat). Fish oil also resulted in the lowest n-6 to n-3 ratio (1.16 to 1), while sunflower oil resulted in the highest ratio (24.6 to 1). Results indicate that the fatty acid profile of eggs could be altered by means of dietary intervention. However, an improvement of omega-3 type fatty acids of eggs will result in a higher susceptibility to lipid oxidation and possibly a shorter shelf-life of stored eggs.

Keywords: Fish oil, high oleic sunflower oil, linseed oil, percentage yolk fat, peroxide value, tallow
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Introduction

Eggs are one of the most complete foods, yet the industry is being put under severe pressure, largely due to controversial reports regarding its cholesterol content (Basmacioglu *et al*., 2003). However, Simopoulos (2000) illustrated that cardiovascular diseases are more related to the fatty acid composition of the specific food source, rather than the cholesterol content itself. Koutsos (2007) reported that the fatty acid profile of eggs can be enriched between five- to thirty folds, without any negative effect on egg quality and/or production performances, depending on the type of fatty acids fed to the hens. Furthermore, Jiang *et al*. (1991) concluded that unsaturated dietary lipid sources could reduce the cholesterogenic effect of eggs by altering the fatty acid composition and incorporating more omega-3 and omega-6 type fatty acids into egg yolk. Furthermore, Cachaldora *et al*. (2008) reported that the dietary inclusion of linseed oil and fish oil would result in a linear increase in the α-linolenic-, eicosapentaenoic-, docosapentaenoic- and docosahexaenoic acid concentrations of egg yolk, depending on dietary lipid inclusion levels.

However, increases in the polyunsaturated fatty acid (PUFA) content of eggs by means of omega-3 fatty acid enrichment would also result in an increased susceptibility to lipid oxidation. This could affect egg quality negatively, mainly due to a decrease in organoleptic properties of eggs, decreasing consumer acceptability toward "enriched" products (Hargis & Van Elswyk, 1991). Additionally, Dunn-Hurrocks *et al*. (2011) reported that using dietary fish- and flaxseed oil in layer diets to manipulate the fatty acid profile of eggs resulted in higher (P <0.01) thiobarbituric acid reactive substances (TBARS) with a consequent

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negative impact on the egg quality and its shelf-life. It seems that factors such as lipid source and inclusion levels (Scheideler & Froning, 1996), as well as bird age and genotype (Schneider *et al*., 1990), could influence the metabolic efficiency of dietary fatty acids into egg yolk fatty acids. The divergent information in terms of lipid sources and inclusion levels, as well as their effects on the oxidation stability of end-products produced (eggs), clearly indicates the need for more research.

The aim of this study was to investigate the effects of supplementary dietary lipid sources on the fatty acid profile and lipid oxidation of egg yolk.

Materials and Methods

Two hundred (n = 200) HyLine Silver-Brown hens (20 weeks of age) were randomly divided into five dietary treatments, each consisting of 40 birds/treatment. Five isoenergetic (12.6 MJ AME/kg DM) and isonitrogenous (170 g CP/kg DM) diets were formulated, consisting of deodorised fish oil (n-3 source), sunflower oil (n-6 source), high oleic acid (HO) sunflower oil (n-9 source) and tallow (saturated fatty acid source), as well as a control diet consisting of a blend (50 : 50) of fish- and linseed oil, at a 30 g/kg dietary inclusion level. Diets were formulated to ensure that the calcium (39.4 g Ca/kg DM) and available phosphorus (3.2 g AvP/kg DM) content, as well as the ratio of Ca : AvP (12.3 : 1), were constant across treatments. Birds were housed individually in metabolic cages (1600 cm^2/bird), while feed and water were provided *ad libitum*. At 30 weeks of age, 12 eggs per treatment (n = 60 in total) were randomly selected, weighed and broken-out for the separation of egg yolk from the albumen. An aliquot of the yolk was taken for analyses of fatty acid methyl esters (FAME), while the remainder of the yolk was sampled for the determination of egg lipid oxidation. Total lipid content was extracted from yolk using the method firstly described by Folch *et al*. (1957). Fatty acid methyl esters were prepared for gas chromatography by methylation of the extracted fat, using methanol–BF_3 (Diaz *et al*., 2005) and were quantified using a Varian GX 3400 flame ionization GC, with a fused silica capillary column, Chrompack CPSIL 88 (100 m length, 0.25 mm ID, 0.2 μm film thickness). Individual fatty acids identified, were expressed as a percentage of the total fatty acids present in the sample (% FAME). To determine egg lipid oxidation, yolk samples were analysed for peroxide values (AOAC, 2000) and thiobarbituric acid reactive substances (Raharjo *et al*., 1992). Data were statistically analyzed using a fully randomized one-way ANOVA design. The PROC ANOVA procedures of the SAS program (SAS, 2001) was used to test for significant (P <0.05) differences between treatment means, while Tukey's honest significant difference (HSD) test was used to separate treatment means.

Results and Discussions

The effect of dietary lipid source on the fat (%) and fatty acid methyl ester concentration (FAME %) of egg yolk is presented in Table 1. Dietary lipid source had no effect (P = 0.24) on the total fat content of egg yolk. These results are in agreement with Jiang *et al*. (1991) and Cachaldora *et al*. (2008) who reported that dietary lipid source and fatty acid saturation did not affect the total lipid content of egg yolk. As expected, egg yolk FAME as well as fatty acid ratios were affected (P <0.0001) by dietary lipid sources (Table 1). These results concur with Cachaldora *et al*. (2008) who found similar effects of dietary lipid source on the FAME of egg yolk. Although the effect of dietary lipid sources on the concentration of total saturated fatty acids (SFA) of egg yolks were relatively constant across treatments (varied from 34.6% to 40.3%), differences (P <0.0001) in total polyunsaturated fatty acids (PUFA) of egg yolks were more noticeable between the treatments (15.7% to 25.5%). Also, both the control (1.51 : 1) and fish oil (1.16 : 1) treatments had lower (P <0.0001) n-6 to n-3 (n-6 : n-3) ratios than the other treatments. By calculating the total n-3 content of eggs (mg/g egg), a definite distinction (P <0.0001) could be made between the omega-3 enriched treatments (control and fish oil) and all other experimental treatments.

The effect of dietary lipid sources on the lipid oxidation characteristics of egg yolk, as expressed by thiobarbituric acid reactive substances (TBARS) and peroxide values (PV), are presented in Table 2. Fish oil (n-3 source) resulted in the highest TBARS (P <0.0001) and peroxide values (P <0.002) compared to all other lipid sources used. Additionally, the HO sunflower oil treatment resulted in the lowest (P <0.002) PV value (2.77 milli-equivalent peroxide/kg fat). Supportive to the results of the present study, Dunn-Hurrocks *et al*. (2011) also reported higher (P <0.01) TBARS in egg yolks from hens fed fish oil compared to

Table 1 The mean fat content, fatty acid content and fatty acid methyl esters (FAME) content and fatty acid ratios of egg yolk as affected by dietary lipid sources

	Control	Fish oil	Sunflower oil	HO[1] sunflower oil	Tallow	Significance (*P*)
Egg yolk fat content (%)	31.28	30.51	30.35	31.45	30.98	NS[2]
FAME (% of total fatty acids)						
C16:0	26.66^{b}	30.00^{a}	26.67^{b}	25.59^{b}	26.52^{b}	<0.0001
C18:0	10.16^{ab}	9.51^{bc}	11.29^{a}	8.61^{c}	10.04^{ab}	<0.0001
C18:1c9	34.64^{c}	31.91^{c}	33.06^{c}	44.80^{a}	41.82^{b}	<0.0001
C18:2c9,12 (n-6)	12.71^{b}	11.03^{d}	20.41^{a}	12.25^{bc}	11.58^{cd}	<0.0001
C18:3c9,12,15 (n-3)	2.42^{a}	0.22^{b}	0.14^{b}	0.12^{b}	0.16^{b}	<0.0001
C20:2c11,14 (n-6)	0.10^{c}	0.08^{c}	0.26^{a}	0.13^{b}	0.10^{bc}	<0.0001
C20:3c11,14,17 (n-3)	0.16^{bc}	0.14^{c}	0.24^{a}	0.16^{bc}	0.20^{ab}	<0.0001
C20:4c5,8,11,14 (n-6)	1.23^{c}	1.00^{c}	3.71^{a}	2.77^{b}	2.75^{b}	<0.0001
C20:5c5,8,11,14,17 (n-3)	0.54^{b}	0.95^{a}	ND[3]	ND[3]	ND[3]	<0.0001
C22:5c7,10,13,16,19 (n-3)	0.56^{b}	0.90^{a}	ND[3]	ND[3]	0.08^{c}	<0.0001
C22:6c4,7,10,13,16,19 (n-3)	5.76^{b}	8.33^{a}	0.66^{c}	0.65^{c}	0.88^{c}	<0.0001
SFA	37.40^{b}	40.33^{a}	38.35^{ab}	34.64^{c}	37.43^{b}	<0.0001
MUFA	39.13^{b}	37.03^{b}	36.15^{b}	49.23^{a}	46.83^{a}	<0.0001
PUFA	23.47^{b}	22.64^{b}	25.50^{a}	16.14^{c}	15.74^{c}	<0.0001
Fatty acid ratios						
PUFA / SFA	0.63^{a}	0.56^{b}	0.67^{a}	0.47^{c}	0.42^{d}	<0.0001
n-6 / n-3	1.51^{d}	1.16^{d}	24.60^{a}	17.22^{b}	11.90^{c}	<0.0001
mg n-3 / g egg	7.26^{a}	7.78^{a}	0.73^{b}	0.74^{b}	0.95^{b}	<0.0001

^{a,b,c,d} Row means with different superscripts differ significantly (P <0.0001); [1] High oleic acid sunflower oil; [2]NS – not significant (P >0.05); [3] ND – not detected.
SFA: total saturated fatty acids; MUFA: total monounsaturated fatty acids; PUFA: total polyunsaturated fatty acids.

Table 2 The mean (± SD) thiobarbituric acid reactive substances and peroxide value of egg yolk as affected by dietary lipid source

	Control	Fish oil	Sunflower oil	HO[1] sunflower oil	Tallow	Significance (*P*)
Thiobarbituric acid reactive substances (mg malonaldehyde/kg yolk)	0.19^{b} ±0.05	0.27^{a} ± 0.09	0.16^{b} ± 0.05	0.13^{b} ± 0.04	0.13^{b} ± 0.03	<0.0001
Peroxide value (milli-equivalent peroxide/kg fat)	3.07^{b} ± 0.64	3.96^{a} ± 0.67	3.53^{ab} ±1.01	2.77^{b} ± 0.45	3.23^{ab} ±0.73	<0.002

^{a,b} Row means with different superscripts differ significantly (P <0.05).
[1] High oleic acid sunflower oil.

sunflower oil. In the present study, the control diet consisting of a blend between fish- (15 g/kg) and linseed oil (15 g/kg) seems to resist lipid oxidation better (P <0.002) than the pure fish oil (30 g/kg) treatment. Supportive to the present findings, Hayat *et al*. (2010) reported that the inclusion of flaxseed in layer diets resulted in a higher oxidative stability compared to fish oil and related this improvement in oxidative

stability to the higher levels of shorter chain omega-3 type PUFA such as α-linolenic acid within flaxseed oil, compared to the longer chain omega-3 type PUFA such as eicosapentaenoic-, docosapentaenoic- and docosahexaenoic acid in fish oil.

Conclusions

This study has shown that it is possible (P <0.0001) to alter the fatty acid profile of egg yolk without any negative effects (P = 0.24) on the total fat content of eggs itself. Furthermore, it seems that combination of plant (linseed oil) and fish oil would withstand lipid oxidation in egg yolk better than the dietary inclusion of pure fish oil in diets to produce omega-3 enriched eggs. Nutritionist should therefore be cautious during diet formulation for "enriched eggs" in ensuring that that use of specific lipid sources would not result in a shorter shelf-life of eggs due to an increase in lipid peroxidation.

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