Fatty acid composition of eggs produced by hens fed diets containing groundnut, soya bean or linseed

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Abstract

The influence of the diet's fatty acid composition on the fatty acid composition, weight, fat and cholesterol contents of chicken eggs was studied in an experiment in which laying hens were fed diets of similar proximate composition but containing either groundnut, soya bean or linseed. The fatty acid composition of the egg fat corresponded with that of the diets: groundnut feeding produced a high oleic acid content of the eggs, soya bean feeding resulted in a high level of linoleic acid and linseed feeding raised the level of α-linolenic acid. Incorporation of groundnut versus linseed into the diet raised the arachidonic acid content of the eggs. Feeding the linseed diet produced higher contents of eicosapentaenoic and docosahexaenoic acid. The three experimental diets did not differently influence the cholesterol and fat contents of the eggs, but linseed versus soya bean significantly reduced egg weight by on average 3.4%. It is indicated that eggs, depending on their composition, can contribute significantly to man's daily requirement of docosahexaenoic acid; high intakes of this fatty acid might reduce coronary heart disease.

Additional keywords: linoleic acid, α-linolenic acid, egg yolk

Introduction

The intake of extra n-3 polyunsaturated fatty acids, i.e., α-linolenic (ALA), eicosapentaenoic (EPA) and docosahexaenoic (DHA) acid, may reduce coronary heart disease and overall mortality in man (Von Schacky, 2000; Harris & Isley, 2001). It is well known that the fatty acid composition of the diet of laying hens affects the fatty acid composition of their eggs. Earlier studies (Wheeler *et al.*, 1959; Chen *et al.*, 1965) mainly focused on the feeding of sunflower seed or soybean oil, whereas more recent studies used diets enriched with linseed oil (Caston & Leeson, 1990; Jiang *et al.*, 1991) or fish oil (Huang *et al.*, 1990; Nash *et al.*, 1995). The content in egg yolk of linoleic acid (LA) – the major n-6 polyunsaturated fatty acid – can be raised. However, because LA is abundant in the western human diet, the enrichment of eggs with this fatty acid has no practical relevance. EPA and DHA are present only in fish products. Eggs enriched with EPA and DHA could be a source of these n-3 polyunsaturated fatty acids for people not consuming fish products. The recommended daily intake of total n-3 polyunsaturated fatty acids has been set at 0.5% of the intake of dietary energy, which is equivalent to 1.1–1.5 g fatty acids per day per adult (Anon., 1990). Ferrier *et al.* (1995) calculated that one egg from a hen fed a diet containing 10% linseed – which is rich in ALA – would supply about 30% of the daily need of total n-3 polyunsaturated fatty acids. Such an egg provides about 264 mg ALA, 10 mg EPA and 82 mg DHA. The latter two fatty acids probably are the most potent n-3 polyunsaturated fatty acids in relation to human health (Von Schacky, 2000).

The contents of EPA and DHA in eggs can be increased by feeding diets containing fish oil to the hens, but this has a negative impact on the palatability of the eggs (Huang *et al.*, 1990). Another possibility to raise the EPA and DHA contents is through the feeding of a diet rich in ALA (Jiang *et al.*, 1991). The ALA is converted into EPA and DHA by the hen's liver and the synthesized fatty acids are subsequently excreted with the egg yolk. The conversion of ALA into EPA and DHA may be inhibited by high intakes of LA (James *et al.*, 2000). The oil in linseed contains about 60% ALA whereas that in soya beans contains about 60% LA. Groundnut oil is rich in oleic acid but contains less ALA than the oil in soya beans.

In this study with laying hens, the feeding of linseed was compared with the feeding of soya bean or groundnut, not only with respect to egg fatty acid composition, but also with respect to egg weight, fat and cholesterol content. By using the three feedstuffs, three different dietary levels of LA and ALA were installed. It was anticipated that the feeding of linseed would produce the highest levels of EPA and DHA in egg yolk whereas the feeding of soya bean would induce the lowest levels.

Materials and methods

Animals and diets

The experiment was carried out at a commercial layer farm (WEKO Egg Products, Ochten, The Netherlands). Lohmann Brown hens, aged 41 weeks, were used. The hens were housed in battery cages with 4 or 5 birds per cage. Four dietary treatments – a reference diet and 3 experimental diets – were allocated at random to 20 cages, so that each treatment was applied to 5 cages.

A commercial layer diet (De Heus BV, Barneveld, The Netherlands) served as the reference diet. The proximate composition as given by the manufacturer was as follows (g per 100 g diet): crude protein, 17.0; crude fat, 9.5; crude fibre, 5.6; ash, 13.7. The analysed composition is given in Table 2. The three experimental diets contained groundnut, soya bean or linseed (Table 1) and were formulated in such a way that their calculated contents of crude protein, crude fat, crude fibre, ash and dry matter were

Table 1. Ingredient composition of the experimental diets.

Ingredient (g per kg diet)	Variable component		
	Groundnut	Soya bean	Linseed
Groundnut[1]	140	–	–
Soya bean[2]	–	115	–
Linseed[3]	–	–	200
Tapioca	114.2	92.6	55.7
Soya bean oil	–	47	–
$Ca(H_2PO_4)_2.H_2O$	9.8	9.8	9.0
D,L-methionine	1.8	1.6	1.3
L-lysine, HCl	0.2	–	–
Constant composition[4]	734	734	734

[1] Groundnut seeds without (red) seed coat; Robi BV, Wijk bij Duurstede, The Netherlands.

[2] Soyax soya bean seeds; Van Eck BV, Wijk bij Duurstede, The Netherlands.

[3] Canadian, brown linseed; Arie Blok BV, Woerden, The Netherlands.

[4] The constant composition consisted of (g): barley, 50; maize, 50; wheat, 200; peas, 100; soya bean concentrate, 114; maize gluten meal, 50; lucerne meal, 70; NaCl, 3; $CaCO_3$, 87; premix, 10. The premix consisted of (mg): retinyl acetate and retinyl palmitate , 20 (10,000 IU); cholecalciferol, 20 (2,000 IU); α-tocopherol, 25; riboflavin, 3.5; niacin amide, 30; D-panthothenic acid, 12; choline Cl, 350; cyanocobalamine, 0.015; menadion, 1.5; folic acid, 0.8; biotin, 0.1; $Na_2SeO_3.5H_2O$, 0.15; $FeSO_4.5H_2O$, 100; MnO_2, 100; $ZnSO_4.H_2O$, 150; ethoxyquin, 100; maize meal, 9086.935.

Table 2. Analysed composition of the reference and experimental diets.

	Reference diet	Experimental diet		
		Groundnut	Soya bean	Linseed
Chemical analysis (g per 100 g diet)				
Dry matter	92.5	91.9	91.7	90.7
Nitrogen	2.52	2.75	2.71	2.66
Crude fibre	7.2	5.1	5.2	6.0
Ash	18.0	13.8	14.4	13.3
Crude fat	10.5	9.2	9.2	11.2
Fatty acids (g fatty acid methyl ester per 100 g esters)				
Palmitic acid	22.3	13.3	13.7	9.0
Stearic acid	10.2	3.8	4.1	3.6
Oleic acid	31.7	28.2	18.5	17.3
Linoleic acid	19.3	29.7	42.2	17.7
α-linolenic acid	1.6	1.3	5.7	36.6

nearly identical. The major ingredient used to balance the diets was tapioca. Table 2 shows that the analysed composition of the experimental diets agrees well with the calculated composition. The formulation of the experimental diets was such that they contained calculated levels of 3.8% (w/w) calcium, 0.42% methionine and 0.9% lysine, and to have a base excess (meq $Na^+ + K^+ - Cl^-$ per 100 g) of 21–24 meq per 100 g. The energy density of the diets containing either groundnut, soya bean or linseed was 10.6, 10.5 and 11.8 MJ metabolizable energy per kg diet, respectively.

The fatty acid composition of the experimental diets (Table 2) reflected the variable components. The diet with groundnut was relatively rich in oleic acid (C18:1 n-9), that with soya bean contained a high percentage of LA and the linseed diet had a high level of ALA. The reference diet differed from the three experimental diets in that it contained a relatively high percentage of palmitic acid (C16:0).

The four diets were in mash form and were provided for *ad libitum* consumption. The diets were enriched with 0.75% (w/w) shell grit. The hens had free access to drinking water through nipples.

Feed intake could not be determined per cage because the hens were able to reach the adjacent feed troughs and thus only the mean intake per dietary group was calculated. Cage adjustments were made so that the reachable adjacent feed troughs contained the same type of diet. Eggs were collected in the period 21–34 days after the hens were first offered the experimental diets. It has been shown earlier that it takes about 21 days for the fatty acid composition of eggs to reach a constant level after diet change (Huang *et al.*, 1990).

Analytical methods

Nitrogen in the diets was determined using the Kjeldahl method, and crude fibre and ash according to the Weende method (Joslyn, 1970). Total fat content of diets and eggs was assessed with the Folch method (Joslyn, 1970) and the fatty acid composition was determined as described by Beynen & Katan (1985). Total egg cholesterol was measured according to Nordby & Nagy (1973).

Statistical analyses

The two-tailed Student's t-test was used to evaluate differences between diet-induced variables. To take into account the enhanced risk of a type I error due to multiple comparisons, the level of statistical significance was pre-set at $P < 0.017$ instead of $P < 0.05$ according to Bonferroni's adaptation (Steel & Torrie, 1980).

Results

Body weight and feed intake

After 34 days of feeding the experimental diets, the body weights (mean ± SD) of 4 randomly chosen birds per dietary group were 1.94 ± 0.18, 1.88 ± 0.17 and 1.74 ± 0.16

kg for the hens fed groundnut, soya bean or linseed, respectively. Mean body weight for the reference group was 1.99 ± 0.14 (n = 4). Mean feed intake for the hens offered the groundnut, soya bean or linseed diets was 151, 139 and 134 g per hen per day, respectively.

Egg production and fat content

During the period 21–34 days after the beginning of feeding the experimental diets, mean rate of lay was 60, 58 and 61% for the diets with groundnut, soya bean and linseed, respectively. This percentage was 56 for the hens fed the reference diet. Administering the linseed diet resulted in a statistically lower egg weight than did the diet with soya bean (Table 3). The hens fed the groundnut-containing diet produced an intermediate egg weight. Egg weight for the hens fed the reference diet did not differ statistically from that for the hens fed the diet with groundnut or soya bean, but was higher than the egg weight for the hens offered the linseed diet.

Total fat and cholesterol contents were analysed in 5 randomly chosen eggs per dietary group. There were no statistical differences in egg fat and egg cholesterol contents between the 4 dietary groups (Table 3).

Table 3. Weight, cholesterol and fat content of eggs produced by laying hens fed the reference or the experimental diets. Results are means ± the standard deviation of 131–147 (egg weight) or 5 (cholesterol and fat content) randomly chosen eggs per dietary group.

	Reference diet	Experimental diet		
		Groundnut	Soya bean	Linseed
Egg weight (g)	66.1 ± 5.24ac[1]	65.2 ± 3.61ab	66.7 ± 3.66c	64.4 ± 4.21b
Cholesterol content (mg per 100 g)	373 ± 18a	399 ± 11a	377 ± 35a	363 ± 26a
Fat content (g per 100 g)	10.3 ± 0.69a	10.8 ± 0.36a	10.3 ± 0.62a	10.5 ± 0.53a

[1] Means in the same line, not followed by the same letter(s) are statistically different ($P < 0.017$).

Fatty acid composition of the eggs

The reference eggs differed in fatty acid composition from the experimental eggs in that their contents of palmitic and oleic acid were statistically higher, whereas the percentage of LA was statistically lower (Table 4). The eggs from the experimental diets differed statistically with regard to their contents of oleic acid, LA, ALA and

Table 4. Fatty acid composition (g fatty acid methyl ester per 100 g esters) of eggs produced by laying hens fed the reference or the experimental diets. Results are means ± the standard deviation of 5 randomly chosen eggs per dietary group.

Fatty acid	Reference diet	Experimental diet		
		Groundnut	Soya bean	Linseed
Palmitic acid	22.8 ± 0.69a[1]	21.2 ± 0.84b	20.5 ± 0.41b	20.8 ± 0.89b
Palmitoleic acid	2.9 ± 0.19a	2.2 ± 0.15b	2.1 ± 0.09b	3.1 ± 0.28a
Stearic acid	7.5 ± 0.42a	8.0 ± 0.55ab	8.8 ± 0.32b	8.0 ± 0.20a
Oleic acid	44.7 ± 1.35a	39.4 ± 0.70b	31.7 ± 0.68c	36.6 ± 1.50d
Linoleic acid	13.6 ± 0.51a	21.8 ± 0.59b	27.7 ± 0.60c	16.1 ± 1.11d
α-linolenic acid	0.5 ± 0.06a	0.5 ± 0.00a	2.1 ± 0.14b	8.2 ± 1.62c
Arachidonic acid	1.8 ± 0.00a	2.1 ± 0.15b	1.9 ± 0.14ab	1.1 ± 0.13c
Eicosapentaenoic acid	0.0 ± 0.00a	0.0 ± 0.00a	0.0 ± 0.00a	0.1 ± 0.08b
Docosahexaenoic acid	1.2 ± 0.06a	0.8 ± 0.06b	1.5 ± 0.08c	2.2 ± 0.24d

[1] Means in the same line, not followed by the same letter(s) are statistically different ($P < 0.017$).

DHA. Eggs from hens fed the groundnut diet had the highest contents of oleic and arachidonic acid [C20:4 (n-6)], but the lowest percentages of ALA and DHA. The diet with soya bean produced eggs with the highest LA and the lowest oleic acid contents. Feeding the linseed diet resulted in eggs with the highest level of ALA, EPA and DHA, and the lowest content of LA.

Discussion

This study confirms that the fatty acid composition of the diet of laying hens influences the fatty acid composition of their eggs. The principal fatty acids in the groundnut, soya bean and linseed, i.e., oleic acid, LA and ALA, respectively, were reflected in the fatty acid composition of the eggs. Moreover, the composition of the diet affected fatty acid metabolism of the hens, which was reflected by the fatty acid composition of the eggs too. The feeding of groundnut raised the arachidonic acid content of the eggs, which cannot be easily explained because the relatively high intake of oleic acid would be expected to inhibit the conversion of LA into arachidonic acid (Brenner, 1989). Enrichment of the diet with soya bean produced lower contents of oleic acid in the eggs than did the feeding of linseed, although the soya bean and linseed diets contained similar levels of oleic acid. Feeding the linseed diet raised the amounts of EPA and DHA in the eggs, which relates to the synthesis of these fatty acids from their precursor, ALA (Brenner, 1989).

Egg production was relatively low for each of the four diets, which may have been caused by the extremely hot weather during the egg-collecting period. The data indicate that egg weight was on average 3.4% lower after linseed than after soya bean feeding. The experimental diets did not differently influence cholesterol and fat contents of the eggs. The few data on body weight and feed intake should be interpreted with caution, but it seems that the hens fed the diet containing groundnut had the highest body weight and the highest feed intake. So the incorporation of n-3 polyunsaturated fatty acids into eggs through linseed feeding may be associated with a small but statistically significant decrease in egg weight without affecting the cholesterol and fat content of the eggs.

At low fat intakes, dietary LA and ALA will be relatively more diluted by *de novo* synthesized fatty acids than at high intakes (Beynen *et al.*, 1980). To take this phenomenon of dilution into account, the levels of dietary LA and ALA may be expressed as energy percentage of metabolizable energy. However, if the dietary polyunsaturated fatty acids are quantified as a percentage of total dietary fatty acids there are strong relationships with the relative percentages of polyunsaturated fatty acids in egg yolk (Sim *et al.*, 1973). In my study, the soya bean diet containing 42.2% LA induced an average egg LA content of 27.7%. The linseed diet containing 36.6% ALA was associated with an egg ALA content of 8.2%. So dietary LA is more efficiently incorporated into egg yolk than ALA.

The soybean and linseed diets contained 5.7 and 36.6% ALA, respectively, and the hens fed these diets produced eggs containing 1.5 and 2.2% DHA. In this study, the eggs from hens fed the linseed diet contained 0.1% EPA, but in the eggs from the two other dietary groups no EPA was detected. It appears that only very high intakes of ALA can lead to detectable amounts of EPA in the egg yolk, but the EPA level remains much lower than that of DHA. The conversion of dietary ALA into DHA, as based on the fatty acid composition of eggs, is not efficient.

In conclusion, this study shows that the contents of LA, ALA, EPA and DHA in eggs can be influenced by the diet of the hens, the most efficient process being the transfer of dietary LA into egg yolk. Eggs produced by hens fed diets rich in linseed or soya bean were found to contain 141 and 98 mg of DHA, respectively, and eggs from hens fed the reference diet contained on average 78 mg DHA. Such eggs can contribute significantly to the daily DHA requirement of humans (Ferrier *et al.*, 1995).

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