UNE'S DESIGNER EGG : RECENT ADVANCES

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SUMMARY

An experiment was undertaken in which laying hens were given 4 diets supplemented (g/kg) with one or a mixture of edible oils and enriched with an antioxidant and vitamin E. Egg production and egg mass were reduced on the diets (1 & 2) highest in fish oil compared to diets 3 & 4. On average the yolk content of vitamin E was increased by 280% as a result of supplementation. Total omega-3 polyunsaturated fatty acids (n-3 PUFA) was increased from 1g to 7g/100g total fatty acids in enriched eggs. Both eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) were highest in eggs from hens fed diets with the highest amount of fish oil. Diets 2 and 3 with vegetable oil gave highest concentrations of alpha linolenic acid (ALA). A 2-day weighed food intake survey of 52 subjects consuming 7 eggs weekly for 22 weeks gave similar intakes for energy and the contribution of protein, fat and carbohydrate to energy intake. Proportions (g/day and %) of saturated, monounsaturated and polyunsaturated fatty acids were also similar between treatments. Although mean plasma cholesterol (n=56) did vary between sampling periods, it was the same at the start and the finish of the study. Triglycerides stayed the same. High density lipoprotein cholesterol (HDL) increased (P<0.05) during the same period. Dietary treatment showed no difference in plasma triglycerides nor HDL; cholesterol was higher (P<0.001) on treatment 1 with eggs from hens given fish oil than on other treatments as was low density lipoprotein cholesterol (LDL). Blood pressure was lower (P<0.01) on treatment 3 when compared to treatments 1 and 2. For the last 2 weeks of the experiment mean egg consumption was increased to 21 eggs. This increase caused a small increase in plasma triglycerides compared to the previous measurement at week 22.

Plasma samples taken from 12 hour fasted subjects showed significant increases in DHA and total n-3 PUFA on treatments 1 and 2 compared to treatment 4. The ratio of n6 n-3 FUFA was significantly reduced on all three treatments compared to the control treatment (4). It is concluded that consumption of only one enriched egg daily can contribute substantially to the recommended daily intake for n-3 PUFA particularly for EPA and DHA. The potential for using egg yolk as a carrier of these fatty acids for use in infant bottle formula is discussed.

INTRODUCTION

There is considerable interest in the omega-3 long chain polyunsaturated fatty acids (n-3 PUFA) in human health (Galli and Simopoulos 1989; Simopoulos et al 1991a; Hargis and Van Elswyk 1993). Sinclair (1991) has reviewed the subject recently and Simopoulos (1988 a,b) a little earlier.

The main dietary source of n-3 PUFA for man is fish. Marine phytoplankton synthesise n-3 PUFA and in this way they are introduced into the food chain. Variation in the oil content of fish differs markedly among species and the n-3 PUFA content of the oil varies between the different species. Generally those fish found in cold water have a higher n-3 PUFA content than those fish in warm water (Evans et al 1986; Fogerty and Sovonos 1987).

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A very few vegetable oils such as linseed, rapeseed (canola) and soybean contain different amounts of alpha-linolenic acid (ALA, 18:3, n-3), while fish contains very little ALA and more eicosapentaenoic acid (EPA, 20:5, n-3) and of docosahexaenoic acid (DHA, 22:6, n-3). In many Western countries fish is not consumed in large quantities for several reasons and a dietary insufficiency of the n-3 PUFA likely occurs (Sinclair 1991).

We have reported recently on the enrichment of hens' eggs (Farrell 1992), poultry meat (Farrell and Gibson 1990, 1991) and duck meat (Farrell 1991) with n-3 PUFA and their effects on humans when they consumed two of the enriched eggs daily for 7 weeks. Production characteristics of the hens were generally enhanced.

In the present experiment we report on the effects of humans consuming only 7 enriched eggs weekly for 20 weeks, and then increasing consumption to 22 eggs during the final 2 weeks. A weighed dietary survey of foods consumed by volunteers over two days was included in the experiment. Hen production data are also given.

MATERIALS AND METHODS

Hens and diets

Seventy-two SIRO-CB (New Hampshire x Australorp) hens were housed in single cages in two identical windowless rooms at constant temperature (22 ± 3oC) and illumination of 16 hours/day. The hens were in production for 16 weeks and were given one of four diets that were formulated to layer specifications (SCA 1987). The diets contained the same ingredients except that diets 1, 2, 3 and 4 contained single or different combinations of edible vegetable and fish oils. Diet 4 contained 40 g sunflower oil. Fatty acid analysis of some of these oils have been reported previously (Farrell 1992). Additional antioxidant was added to each diet. Hens were allowed to adjust to the diets and experimental conditions for 14 days before measurements of feed intake, egg production and egg weight commenced and continued at regular intervals. Eggs were held in a cool room (40C) prior to distribution. About half way through the experiment, vitamin E supplementation of the diet was increased.

Volunteers

Sixty healthy free-living adult volunteers of both sexes were recruited for the study. They were provided with one of the four enriched egg types and asked to consume 7 eggs each week preferably not fryed. In each group there were as far as possible equal numbers of males and females. The experimental protocol was approved by our Committee on Human Research.

Measurements

At the beginning of the experiment, and after 2, 6, 10, 16, 22 and 24 weeks subjects fasted overnight (12 hours). Next morning they were weighed in light clothing, height taken, blood pressure was measured using an automatic digital blood pressure monitor (Omron HEM 703C), a 5 ml blood sample was withdrawn and transferred to a heparanised tube. Subjects were then given an allocation of their enriched eggs. For the first 2 weeks all volunteers consumed the same eggs from a commercial flock of hens. During the last 2 weeks volunteers consumed 22 eggs.

Food intake

Subjects during the last eight weeks of the experiment were asked to describe and weigh all individual foods consumed on an electronic portable balance accurate to 1g for two consecutive week days. The data were transferred to computer diskette and analysed using an appropriate package (SNAP 1992 A.J. Howie) based on Australian food analysis tables.

Analyses

Red blood cells were separated from plasma by centrifugation. Cholesterol, triglycerides and high density lipoproteins (HDL) were determined on plasma using a spectrophotometric analyser (The Cobas Bio, Roche Diagnostics, F Hoffman-la Roche, Basle, Switzerland) and the appropriate diagnostic kit. For fatty acid analysis, egg yolk was separated from the albumin in the whole, preweighed egg and freeze-dried. Yolk lipids were transmethylated (Le page and Roy 1986) and separated by gas chromotography (Hewlett Packard HP 5890 Series II) on a Restek Rtx 2330 30m capillary column (0.32 mm id) using a 0.1-0.2 µl split injection (45:1 split). Plasma free fatty acids, those in red blood cells and in various oils were measured similarly. Fatty acids were identified by relative retention times determined with known standards.

Data were analysed using standard statistical procedures. Where appropriate, differences in initial values were adjusted using covariance analysis. The least significant difference (LSD) test was used to examine differences between treatment means.

RESULTS

Hen performance

Egg production was superior on diet 4 (controls) compared to other diets (Table 1) was significantly ($P<0.05$) reduced on diet 1 compared to diet 4. Egg weight on diets 1-3 was less ($P<0.05$) than on the control diet. Egg mass (g/day) was reduced ($P<0.05$) on diets 1 and 2 compared to diet 4 which contained 4% sunflower oil.

Table 1 Production parameters of hens on the experimental diets measured over 6 months.

	Diet				
	1	2	3	4	LSD (P=0.05)
Egg production (%)	77.9	79.5	81.6	83.6	4.47
Egg weight (g/d)	59.9	57.9	59.0	60.3	1.13
Egg mass (g/d)	48.6	47.5	50.1	52.0	1.98
Feed intake (g/d)	123	119	122	123	4.9
Feed efficiency (g/g)	2.62	2.62	2.47	2.45	0.127

The effect of increasing vitamin E in the feed on yolk content is shown in Table 2.

Table 2 The content of vitamin E in egg yolk (iμ/kg freeze-dryed yolk) before (B) and after (A) increasing vitamin E supplementation.

	iμ/kg	
Diet	B	A
1	56	195
2	66	184
3	74	152
4	76	240
		LSD (P=0.05) 49.3

The mean increase (P<0.01) in vitamin E content of freeze dried egg yolk (iμ/kg) was from 68 to 193 (280%). There was an effect of diet (P = 0.084). The control diet (4) gave a significantly higher response to supplementation than did diets 2 and 3. All diets showed a significant increase (P<0.01) after, compared to before, supplementation.

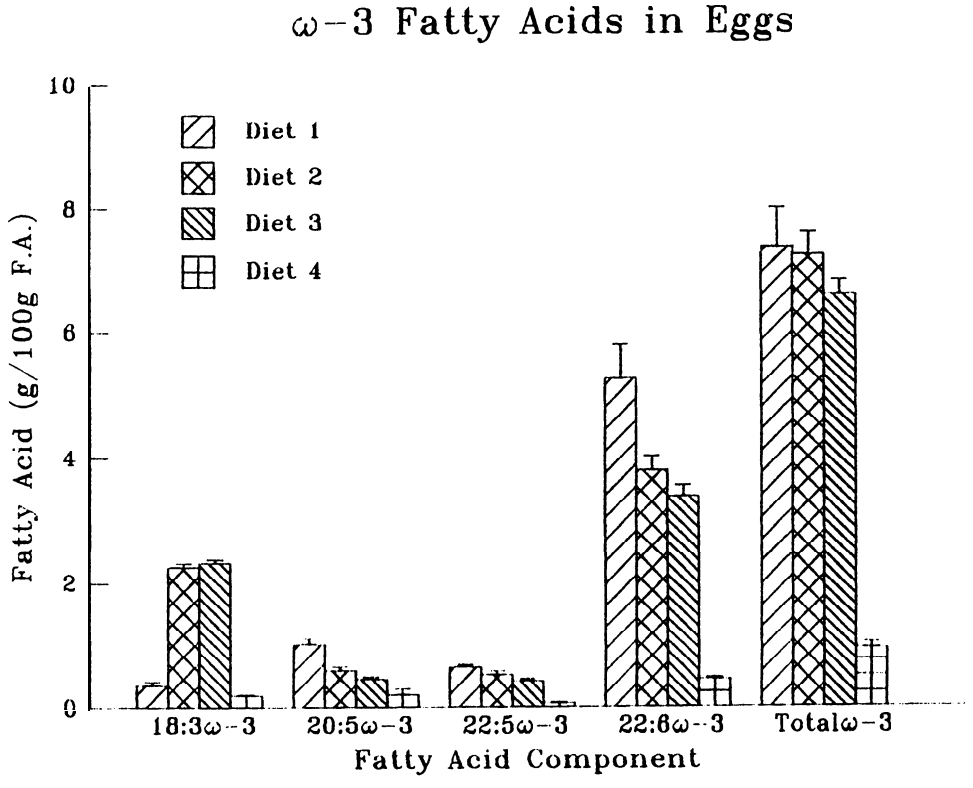

Fig 1. The changes in the free fatty acid content (g/100g) of eggs from hens on four different dietary treatments.

Shown in Fig.1 are the changes in fatty acid content of the enriched and control eggs. The effects of diets 2 and 3 containing some vegetable oil (diet 2) and a mixture of vegetable oils (diet 3) on the ALA content of eggs was significantly greater (P<0.01) than in eggs from hens on diets 1 and 4. EPA was increased significantly on diet 1 but DHA was much higher on all diets compared to the controls. In addition to the three important n-3 PUFA there is docosapentaenoic acid (DPA, 22:5 n-3) which may not have an important direct biochemical role to play in human health. Nevertheless, it is an intermediary in the synthesis of EPA to DHA and in the retroconversion of DHA to EPA.

Total n-3 PUFA was increased in eggs from about 1g to 6-7g/100g total fatty acids on diets 1-3. Other changes that occurred in lipid composition in egg yolk were the decline in palmitic acid (18:0) and the increase in linoleic acid (18:2, n-6) with increasing inclusion of vegetable oil to the diets (1 to 4). Oleic acid (18 : 1) increased in egg yolk from 40.2 to 43.2% on diets 1 to 3 respectively and then declined to 38.6% on diet 4. Total saturated fatty acids declined from 38.8% on diet 1 to 33.2% on diet 4.

Volunteers and dietary survey

Fifty six volunteers successfully completed the experiment. Data were incomplete for a few subjects at the different sampling periods and missing values were calculated by the computer package. Fifty two subjects completed the dietary survey.

The statistics of the volunteers are given in Table 3. The Quetelet index suggests that on average subjects were not obese. Treatment 2 contained a female who weighed 100 kg, this elevated the mean values for this treatment. The mean age of volunteers was 51, 39, 43 and 42 years for treatments 1, 2, 3 and 4 respectively.

Table 3 Statistics of human volunteers participating in the experiment. Mean values and standard deviation in parenthesis

	Height (H,cm)		Weight(kg)		Quetelet index (W,kg/H²)	
Treatment	M	F	M	F	M	F
1	179 (6.3)	164 (6.3)	78 (7.9)	63 (9.6)	24.4 (2.42)	23.6 (4.90)
2	174 (6.7)	163 (6.8)	77 (8.3)	72 (16.0)	225.5 (4.18)	27.0 (7.02)
3	182 (7.2)	165 (7.2)	79 (6.5	61 (12.0)	23.2 (1.61)	22.3 (3.40)
4	179 (5.0)	164 (7.0)	76 (3.4)	67 (14.6)	23.6 (2.25)	24.9 (4.18)

The data for the intake of food components are given in Table 4. Mean fat intake was 95g/day. Of this saturated fat intake was 41%, mono unsaturated fat 34% and polyunsaturated fat 14.4% and similar on all diets.

Table 4 Energy intake (kJ) and its components (% and g/day) of volunteers (14 per treatment) on the four dietary treatments (1-4)

	1		2		3		4		LSD (P=0.05)	
	(%)	(g/d)	(%)	(g/d)	(%)	(g/d)	(%)	(g/d)	(%)	(g/d)
Protein	17.9	92.2	20.6	106.5	16.4	87.0	18.7	97.6	3.51	16.92
Fat	33.6	78.8	35.3	85.9	32.7	80.1	33.9	86.9	5.68	24.57
Carbohydrate	45.2	248.6	42.0	236.8	47.9	259.5	434.4	230.7	7.92	51.33
Alcohol	3.3	9.7	2.6	9.3	3.0	10.1	4.1	15.5	3.41	12.32
Energy	8880		9191		9053		9157		1692.7	

Although variation was quite high, largely due to combining data for the sexes, mean daily energy intake (kJ/day) and the contribution (%) of the major dietary components to energy intake did not differ (P>0.05) between treatments. Except for alcohol consumption, when data were expressed as g/d and for energy (kJ/day), females consumed less of the dietary components shown in Table 4 than males, but not when

expressed as % of total energy intake. However when data were adjusted for differences in bodyweight (W,kg) on a metabolic size basis ($W^{0.75}$) there were no differences ($P>0.05$) between males and females for any of these parameters.

Overall, the contribution to daily dietary energy intake was 18.4% for protein, 32,4% for fat, 46.2% for carbohydrate and 3.0% for alcohol.

The intake of fat type by volunteers on each treatment is shown in Table 5. The contribution (%) of the three different fat types was similar for all treatments. Ratio of saturated to unsaturated fat tended to be higher on treatments 3 and 4.

Although females consumed less of the fat types than males, this was only significant ($P<0.05$) for polyunsaturated fat (10.1 v 13.3 g/day).

Table 5 Intake of the different types of fat expressed as g/day and % of the total for each treatmentand the ratio of % saturated to % unsaturated fat. Standard deviation of the mean is given in parenthesis

Fat type		Treatment			
		1	2	3	4
Saturated	(g/d)	32.5 (4.00)	36.0 (4.32)	34.1 (4.16)	35.6 (4.32)
	(%)	40.2 (1.80)	40.4 (1.95)	41.7 (1.87)	41.6 (1.88)
Mono unsaturated	(g/d)	26.5 (3.36)	29.6 (3.63)	26.6 (3.50)	30.0 (3.63)
	(%)	34.2 (1.42)	34.7 (1.54)	33.5 (1.58)	33.1 (1.48)
Poly unsaturated	(g/d)	12.1 (1.57)	10.9 (1.70	10.9 (1.64)	12.7 (1.70)
	(%)	15.1 (1.19)	13.4 (1.29)	14.0 (1.24)	15.1 (1.25)
Ratio sat:unsat		0.86	0.87	0.90	0.90

Human parameters

Consumption of eggs for the entire 24-week period showed no effect ($P>0.05$) of time (weeks) on plasma triglycerides, no increase ($P>0.05$) in cholesterol, a significant ($P<0.01$) increase in HDL and a decline ($P<0.05$) in low density lipoproteins (LDL) determined by calculation. Blood pressure showed no change. These data adjusted by covariance analysis are given in Table 6 for weeks 2 to 22 when volunteers were consuming 7 eggs weeks. For several parameters there was a significant effect of time.

Table 6 Results of measurements adjusted for the initial differences between individuals (n = 56) for weight, blood pressure, and blood parameters including high density lipo proteins (HDL) and low density lipoproteins (LDL)

Weeks	2	6	10	16	22	Significant effect[1]
Body Weight (kg)	71.8	71.5	71.8	72.0	72.4	*
Triglycerides (m mol/*l*)	1.06	1.04	1.04	1.03	1.10	NS
Cholesterol (m/mol/*l*)	4.7	4.5	4.3	4.8	4.6	*
HDL (m/mol/*l*)	0.81	0.93	0.89	1.03	1.06	**
LDL (m mol/*l*)	3.9	3.5	3.5	3.8	3.5	**
Blood pressure (mm)						
systolic	120	117	116	118	119	**
diastolic	74	72	73	74	73	NS

[1] NS = not significant, * = $P<0.05$, ** = $P<0.01$

There was a small but significant increase in body weight. This can likely be attributed to the nature of the clothing worn. The study commenced in summer and finished in winter. Cholesterol and LDL showed significant variation between sampling times as did systolic blood pressure. HDL increased gradually over time. With the exception of weight and HDL, values at 22 weeks were never significantly higher than those at 2 weeks. The effects of dietary treatment adjusted for initial differences in parameters are given in table 7.

Table 7 The effects of treatment (egg type) on different parameters in individuals (n = 14/treatment) including high density lipoproteins (HDL) and low density lipoproteins (LDL) measured over 22 weeks

Treatment	1	2	3	4	Significant effect[1]
Weight (kg)	71.4	71.6	71.9	72.7	**
Triglycerides (m mol/*l*)	1.10	1.04	1.01	1.06	NS
Cholesterol (m mol/*l*)	4.9	4.5	4.5	4.4	**
HDL (m mol/*l*)	0.96	0.94	0.94	0.93	NS
LDL (m mol/*l*)	3.9	3.5	3.6	3.5	**
Blood pressure (mm)					
Systolic	122	120	114	117	**
Diastolic	75	74	70	73	**

[1] NS = not significant, * $P<0.05$, ** $P<0.01$

Plasma cholesterol and LDL were higher on treatment 1. These eggs were from hens given fish oil. More importantly HDL showed no effect of dietary treatment. Body weight of volunteers on treatment 4 was about 1 kg higher per person than on other treatments.

Blood pressure, both systolic and diastolic, was significantly ($P<0.01$) reduced particularly when compared to treatments 1 and 2. The eggs on this treatment were from hens on the diet with a combination of oils.

During the final 14 days of the experiment mean egg consumption increased to 21 eggs, and was similar on all treatments. In two cases consumption was 12 and 13 eggs.

A paired 't-test' was used to examine effects of increasing egg consumption for 2 weeks (22-24 weeks) on the various parameters. Only triglycderides showed a significant ($P<0.05$) change; they increased by 0.11 m mol/*l*. For bodyweight and blood pressure, mean values for females were lower ($P<0.01$) than males, otherwise there were no other differences during this period.

Fig. 2 gives the changes in the plasma free fatty acids of 12 hour-fasted volunteers consuming the different egg types (7 per week) for 20 weeks and before increasing egg consumption for the next 2 weeks. Although there was an apparent increase in ALA, DPA, DHA and total n-3 PUFA, this was significant only for DPA ($P < 0.085$), DHA ($P = 0.01$) and total n-3 ($P=0.028$). Maximum enrichment of plasma was seen on treatments 1 and 2, compared to treatment 4 (controls) the increase was >65%. The ratio of n-6 to n-3 PUFA declined from 13.6:1 on treatment 4, to 7.3, 7.4 ($P<0.01$) and 9.3 ($P<0.05$) on treatments 1, 2 and 3 respectively (Fig. 3). In egg yolk lipid the reduction was from 25:1 to just over 1:1 for the enriched eggs (Fig. 3).

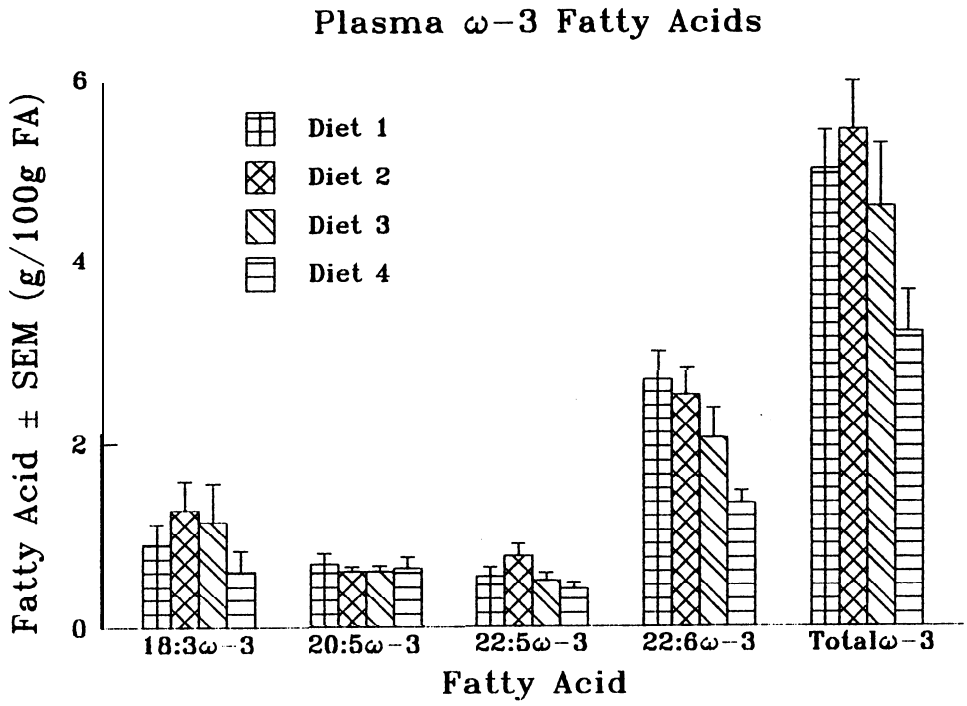

Fig. 2 The increase in free-fatty acids in plasma of humans (n=14/treatment) consuming 7 eggs/weeks of 4 different egg types after 20 weeks.

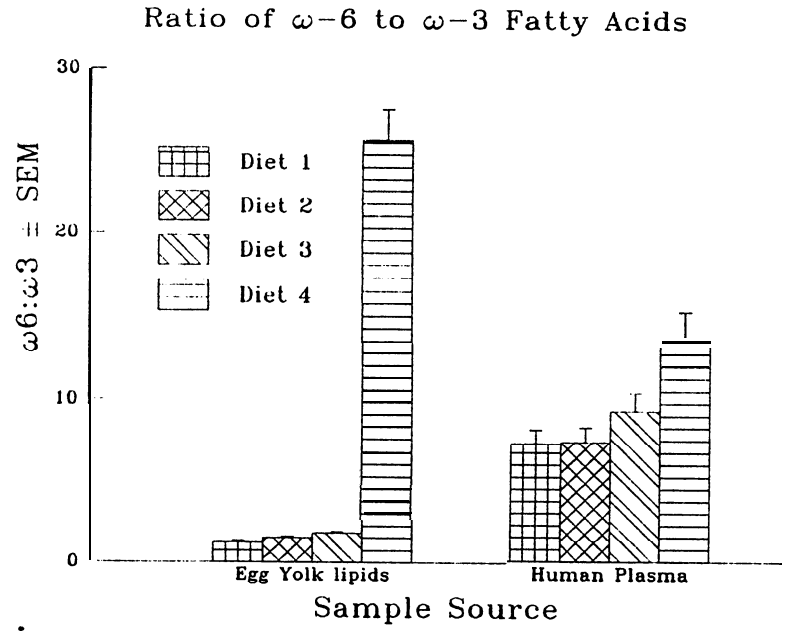

Fig. 3. The change in the ratio of n-6 PUFA in egg yolk lipids and in plasma of humans (n=14 treatment) consuming 7 eggs/week of 4 different egg types after 20 weeks.

Discussion

It is well known that the fatty acid composition of the hens egg can be changed by dietary means (See Noble et al. 1990; Hargis and van Elswyk 1991 for reviews). Normally the eggs from hens fed a conventional, commercial diet contain about 1% n-3 PUFA and a ratio of n-6 : n-3 of about 13:1 (Farrell 1992). Preliminary studies with hens fed various mixtures of vegetable and fish oils showed that fish oil alone resulted in very little enrichment of ALA and small but significant quantities of EPA, but nothing like the relative amounts found in the parent fish oil. Much was deposited as DHA. Similarly

vegetable oils, particularly linseed oil when fed to hens gave large increases in ALA in egg lipid, with virtually no increase in EPA but a substantial increase in DHA (Farrell et al 1991). Knowing the effect of the individual oils on the fatty acids in egg lipid, the mixtures of oils in various concentrations in the hen's diet or singly, will give a predictable fatty acid yolk profile.

There is a reluctance to add significant amounts of fish oil or fish meal to layer diets because of possible off-flavours. Although a taste panel was unable to detect off-flavours in eggs from hens fed 7% linseed, Canola or cod liver oil (Farrell and Gibson 1991), there is still the need to be cautious. In this study, a very few volunteers did complain of a 'fish-taint' on some treatments but the vast majority did not. The inclusion of an antioxidant and the fortification of the diet with vitamin E (Table 2) helped to protect the diet from rancidity and to protect the egg. The elevated content of vitamin E in egg yolk (Table 2) will not only enhance shelf life but will also provide additional vitamin E to the consumer when eating eggs enriched with n-3 PUFA.

Recently Van Elswyk et al (1992) demonstrated that there was no alteration in the fatty acid composition of n-3 PUFA enriched eggs when cooked.

It is likely that hens allowed access to pasture may lay eggs with increased levels of n-3 PUFA. Simopoulos and Salem (1989) found that hens range-fed in Greece had significant increases in ALA, EPA and DHA compared with supermarket eggs. The situation was unusual in that the range-fed hens had access to purslane which is known to be high in ALA (Simopoulos 1988a). Subsequent work by Simopoulos and Salem (1992) showed that these Greek eggs had higher amounts of EPA DPA and DHA than eggs from hens fed fishmeal or flax seed. This is surprising since the latter eggs contained much more ALA than did the Greek eggs and presumably foraging Greek hens were obtaining other dietary sources of EPA and DHA.

The decline in egg production associated with treatments with the higher level of fish oil (Table 1, diets 1 and 2) is not in agreement with our previous observations (Farrell and Gibson 1991). It does agree with the findings of Hargis et al (1991) who observed over an 18 week period a small decline in egg production when hens were fed a diet containing 3% menhaden oil compared to hens on a control diet. This was particularly so towards the end of the experimental period but they showed no difference in egg weight.

The hen has the capacity to rapidly convert ALA to DHA in significant amounts (Farrell 1992) when only linseed oil is added to the diet and measurements are made on hen blood plasma. In rats, conversion of ALA to DHA also takes place rapidly, in the cat not at all but in humans it is thought to occur slowly and in the aged (de Gomez Dumm and Brenner 1975) and Eskimos possibly not at all (Sinclair 1991). The incorporation of dietary n-3 PUFA can be reduced by factors such as the amount and nature of the fat in the diet. Linoleic acid can interfere with the rate of conversion of ALA to EPA and DHA and the efficiency of n-3 PUFA incorporation into tissue (R.A.Gibson 1990, pers. comm., Cleland et al 1992).

In the present study, subjects were asked not to alter their food habits. The dietary survey showed that on average fat intake and the nature of the dietary fat did not differ markedly between treatments (Table 5). The contribution of protein fat and carbohydrate to daily energy intake was also similar (Table 4). Although several subjects consumed on a regular basis some fish, it was surprising to find in free-living subjects that only one enriched egg daily could significantly elevate fasting blood levels of DHA and total n-3 PUFA, as well as reduce the ratio of n-6:n-3 significantly. Previous work, using much higher concentrations of fish oil in the hen's diet than used here and subjects consuming four commercial enriched eggs daily gave elevated values for n-3 PUFA in plasma of fasted subjects (Oh et al 1991) on the former compared to the latter **treatment** and in only

4-week treatment periods. Surprisingly their base line values also showed very high levels of n-3 PUFA, for example DHA was 3.5 - 5.8%. This was particularly surprising as their mean total daily n-3 PUFA intake was 0.6-0.7 g/day during the initial baseline period.

Previously we showed that eating 2 eggs daily for 9 weeks did not elevate significant plasma cholesterol or triglycerides (Farrell and Gibson 1990), it is not surprising therefore that consuming 1 egg daily for 24 weeks did not elevate blood cholesterol or triglycerides (Table 6). The influence of normal egg consumption (1-2/day) on blood cholesterol appears to be negligible (Reiser 1988; O'Dea and Sinclair 1991). Indeed the effect of dietary cholesterol on blood cholesterol is in doubt (Smith 1991). On the other hand the increase in HDL that occurred gradually between week 2 and week 22 (Table 6) is an important finding and known to be beneficial in reducing the incidence of atherosclerosis (Leaf and Weber 1988). In a previous study we found that consuming 2 eggs daily caused a significant reduction in HDL (Farrell and Gibson 1990).

Oh et al (1991) showed that subjects consuming 4 eggs daily from hens on a diet with 10% added fish oil (Max EPA) showed no change in blood cholesterol when consuming the enriched eggs. However blood cholosterol was significantly elevated when subjects consumed 4 commercial eggs daily. The enriched eggs contained 190 mg cholesterol and the commercial eggs 220 mg with a content of n-3 PUFA of >1g per egg. No data were given on egg production parameters.

The reduction in systolic and diastolic blood pressure in volunteers on treatment 3 is also of importance (Table 6). It cannot be explained why this particular combination of oils in the hens' diet and its subsequent effect on volunteers gave such a reduction in blood pressure while treatments 1 and 2 did not. Oh et al 1991) observed a significant decline in systolic and diastolic blood pressure of subjects consuming 4 eggs daily from hens on diets with 10% fish oil (Max EPA). These workers suggested that DHA also has the capacity to reduce blood pressure through retroconversion to EPA.

Although the concentration of EPA in the enriched eggs was small, it has been demonstrated that DHA can also contribute significantly to the inhibition of platelet aggregation (Gaudette and Holub 1991), an important factor in reducing the incidence of cardiovascular disease. Furthermore it is known that there is retroconversion of DHA to EPA (Shacky and Weber 1985; Oh et al 1991) and there was some evidence of this in the significant concentration of DPA in human plasma on treatments 1-3 (Fig. 2). The importance of DHA in neural and retinal tissue of infants and its rapid accumulation in brain cell membranes in the last intra-uterine trimester is well known (Nettleton 1993; Carlson and Salem 1991).

There is an increasing body of evidence that there may be inadequate accumulation of DHA in the premature infant. Visual impairment may result (Neuringer et al 1984; Uavy et al 1990) and learning difficulties may develop if a source of DHA is not provided. There is a linear increase in the tissue accumulation of n-3 PUFA especially DHA of infants to about 2 years of age (Martinez 1991). Human milk usually contains small but significant amounts of DHA while infant bottle formula usually contains negligible quantities (Carlson and Salem 1991). For several reasons, explained by these authors, supplementation of infant formula with fish oil is considered inappropriate. Egg yolk may be an acceptable alternative supplement (Simopulos and Salem 1992).

A major advantage of the hen's egg is its wide acceptability as a human food. In addition the hen acts as a 'biological sieve' to remove or reduce any undesirable contaminants. As demonstrated, there can be control over the concentration and mix of n-3 PUFA in the yolk (Farrell et al 1991).

The recommended dietary intake (RDI) for n-3 PUFA has not been determined, in part because these fatty acids have such a wide role to play in human health, and the separate functions of the different n-3 PUFA. Simopulos et al (1991) suggested that a daily RDI of 0.5 - 1.0 g of total n-3 PUFA was needed to reduce the risk from cardiovascular. Pederson (1991) proposed an RDI of energy intake. Assuming a daily intake of 8 MJ this would be at least 1g of n-3 PUFA.

Bjerve (1991) has given a more precise RDI of 0.86 - 1.2g for ALA in the absence of EPA and DHA, since he suggested that ALA is 2-3 times less effective than EPA in curing and preventing clinical symptoms of n-3 PUFA deficiency. Bjerve (1991) proposed an RDI of 350-400 mg of EPA and DHA with a minimal requirement of 100-200 mg/day. There is little doubt that these estimates are a minimum (Barlow and Pike 1991) particularly for pregnancy and lactation (Nordic Nutrition Recommendations 1989). ALA is now a requirement in infant milk formulas in Canada 1992, but refined unhydrogenated fish oil is not approved for human consumption in the U.S.A. (Nettleton 1993).

A 60 g egg will contain about 6 g lipid (Noble et al 1990). An egg from treatment 2 will provide 430 mg n-3 PUFA, of this the combined DHA, DPA and EPA is 290 mg. On the basis of Bjerve's RDI, the hen's egg will provide almost half of the RDI for n-3 PUFA and all of the requirement for non-ALA n-3 PUFA.

It is estimated that the additional cost of producing a metric ton (mt) of feed with the mix of oils plus additives (treatment 2) would amount to less than $35 per mt of feed. The increase in feed costs to produce a dozen 60 g enriched eggs is about 9c. Fish may be an expensive alternative however fish-oil capsules are relatively expensive and not always acceptable to consumers.

The enriched egg is likely to be a safer alternative for humans; it is not only widely accepted as a cheap and complete nutritional package but it also meets a substantial part of our RDI for n-3 PUFA.

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