

Symposium on ‘How can the *n*-3 content of the diet be improved?’

Current intakes of EPA and DHA in European populations and the potential of animal-derived foods to increase them

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The beneficial effects of long-chain (C chain ≥ 20) *n*-3 PUFA are well documented and, overall, increased intake reduces risk of CVD. Recent evidence also points to a role in reducing age-related decline in cognitive function. The two key fatty acids are EPA (20:5) and DHA (22:6), with current UK recommendation for adults being 450 mg EPA + DHA/d. Whilst some EPA and DHA can be synthesised *in vivo* from α -linolenic acid, recent data indicate this source to be very limited, suggesting that EPA and DHA should be classified as dietary essentials. In many parts of Europe the daily intake of EPA + DHA by adults and especially young adults (18–24 years) is < 100 mg/d, since many never eat oily fish. Poultry meat contributes small but worthwhile amounts of EPA + DHA. Studies to enrich the EPA + DHA content of animal-derived foods mainly use fish oil in the diet of the animal. Recent work has shown that such enrichment has the potential to provide to the UK adult diet a daily intake of EPA + DHA of about 230 mg, with poultry meat providing the largest amount (74 mg). There are, however, concerns that the continued and possibly increased use of fish oils in animals' diets is not sustainable and alternative approaches are being examined, including the genetic modification of certain plants to allow them to synthesise EPA and DHA from shorter-chain precursors.

EPA and DHA intakes: Meat: Milk: Eggs

It was shown in key studies in the 1960s and 1970s that consumption of fish is associated with a reduced risk of CVD in the Greenland Eskimos despite an overall diet rich in fat^(1,2). This work laid the foundation for the concept that the long-chain (C chain length ≥ 20 ; LC) *n*-3 PUFA, in particular EPA (20:5) and DHA (22:6) typically found in marine foods, provide the cardioprotective effects. Subsequently, the beneficial effects have been well documented and include anti-atherogenic, anti-thrombotic and anti-inflammatory effects and, overall, increased intakes lead to reduced risk of CVD (for review, see Scientific Advisory Committee on Nutrition/Committee on Toxicity⁽³⁾). There is also a high requirement for DHA in the last trimester of pregnancy and the first 3 months of life, with the fetus and neonate being dependant on a maternal supply of DHA. There is some evidence that increased maternal LC

n-3 PUFA intake during pregnancy may produce beneficial effects, especially in populations that tend to have a lower background intake of LC *n*-3 PUFA⁽⁴⁾. Current evidence suggests that it is unlikely that the fetus can make sufficient DHA to support its brain development. Thus, maternal DHA will compensate for the limited ability of the fetus to synthesise DHA and therefore it is likely that an adequate intake of LC *n*-3 PUFA could impact fetal development (for example, see Ruxton *et al.*⁽⁵⁾). However, the question of exactly how important dietary DHA is during human brain development remains unresolved⁽⁶⁾.

Evidence is also accumulating that the intake of EPA and DHA may protect against dementia^(7,8) and in particular Alzheimer's disease⁽⁹⁾. Less evidence is available in relation to cognitive function, although recently the fish consumption of 210 male participants (aged 70–89 years in

Abbreviations: ALNA, α -linolenic acid; LC, long-chain.

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1990) of the Zutphen Elderly Study was examined, along with measurements of cognitive function collected in 1990 and 1995⁽¹⁰⁾. A significant ($P < 0.01$) linear trend was seen for the relationship between EPA+DHA intake and cognitive decline, with a mean difference in intake of about 380 mg/d being associated with a 1.1 point difference in cognitive decline. It was thus concluded that moderate intakes of EPA+DHA may delay the decline in cognitive function in elderly men⁽¹⁰⁾.

Theoretically, the dietary essential α -linolenic acid (18:3n-3; ALNA) can be desaturated and elongated to EPA and DHA, but whether the dietary essentiality of ALNA primarily reflects the bioactivity of ALNA itself or of EPA and DHA synthesised from it has been a matter for debate for some time. However, a number of recent studies (for example, see Burdge *et al.*⁽¹¹⁾) and a review⁽¹²⁾ have concluded that the principal biological role of ALNA is indeed as precursor for EPA and DHA but, critically, stable-isotope studies clearly show that the efficiency of conversion of ALNA to EPA is very low, especially in men, and that further transformation to DHA is often minimal. The conversion of ALNA to EPA and DHA is greater in women, possibly as a result of an up-regulatory effect of oestrogen. Overall, it has been concluded that ALNA is probably a quite limited source of EPA and DHA in man⁽¹²⁾, leading to the concept that these PUFA should now be regarded as dietary essentials. This conclusion is supported by a recent systematic review that has concluded that increased consumption of n-3 fatty acids from fish or fish oil supplements, but not from ALNA, reduces the rates of all-cause mortality, cardiac and sudden death and possibly stroke⁽¹³⁾.

The present paper will review current recommendations for intake of EPA and DHA, assess current intakes in various countries and consider how intake may be increased, with emphasis on enriching their concentration in foods of animal origin.

Recommended intakes of EPA and DHA

In the review of dietary factors affecting CVD the Department of Health has recommended that in the UK intake of LC n-3 PUFA should be increased to 200 mg/d from the estimated, then current, intake of about 100 mg/d⁽¹⁴⁾. The subsequent review of the Scientific Advisory Committee On Nutrition/Committee On Toxicity has concluded that the dose required for a demonstrable effect on CVD risk factors such as reductions in plasma TAG concentration, blood pressure, platelet aggregation and the inflammatory response is ≥ 1.5 g/d⁽³⁾. The Scientific Advisory Committee On Nutrition/Committee On Toxicity has also cited other data⁽³⁾, including a study of 240 patients who had suffered a previous myocardial infarction that has demonstrated a reduction in all-cause mortality when these patients were supplemented with 2 g LC n-3 PUFA/d⁽¹⁵⁾. The Scientific Advisory Committee On Nutrition/Committee On Toxicity has concluded, however, that the population recommendation of the Department of Health⁽¹⁴⁾ should be increased from 200 mg/d to 450 mg/d, which is consistent with the consumption of two portions

Table 1. Recommended daily intakes of EPA+DHA for adults in various countries

Country	Recommended intake of EPA+DHA (mg/d)	Reference
UK	200	Department of Health ⁽¹⁴⁾
Various	500*	World Health Organization/Food and Agriculture Organization ⁽⁵⁸⁾
UK	450	Scientific Advisory Committee on Nutrition/Committee on Toxicity ⁽³⁾
Various	500	International Society for the Study of Fatty Acids and Lipids ⁽⁵⁹⁾
USA	270†	Institute of Medicine ⁽¹⁶⁾
Belgium	680†	Belgian Health Council ⁽¹⁷⁾

*Estimated from recommendation to eat one to two portions of fish per week.

†Estimated from original recommendation expressed as % energy intake, assuming an intake of 8.3 MJ/d.

of fish per week, one of which is oil-rich⁽³⁾. No consideration was given to the role of EPA and DHA in arresting cognitive decline because of the paucity of data in this area.

Table 1 summarises the recommended daily intake of EPA+DHA from various sources. Current recommendations range from 270 mg/d in the USA⁽¹⁶⁾ to approximately 700 mg/d in Belgium⁽¹⁷⁾. However, both these intakes are calculated from the original recommendation expressed as % energy intake assuming an intake of 8.3 MJ/d, which may be an underestimate for the USA at least.

Current intakes of long-chain n-3 fatty acids

For the UK some estimates of EPA+DHA intake have been made over the last 15 years but they have produced variable numbers. An intake of 308 mg/d may be calculated from Gregory *et al.*⁽¹⁸⁾, whilst Saunders & Roshanai⁽¹⁹⁾ and Saunders & Reddy⁽²⁰⁾ have reported intakes of 600 mg/d and 500 mg/d respectively. A much lower value of 100 mg/d was used by the Department of Health⁽¹⁴⁾. Some of the variability in estimated mean intake is likely to be the result of the use of different food consumption surveys that suggest different levels of consumption of the key food types. Based on the recommendations of the Scientific Advisory Committee On Nutrition/Committee On Toxicity⁽³⁾ that canned tuna should be excluded from the oil-rich fish food category, it is also likely that some of these studies have substantially overestimated EPA+DHA intake.

A recent study has re-evaluated EPA+DHA intake for UK adults using calculations based on intakes of fish, meat and eggs according to the data of the Scientific Advisory Committee On Nutrition/Committee On Toxicity⁽³⁾, the National Diet and Nutrition Survey⁽²¹⁾ and the British Egg Information Service⁽²²⁾ respectively⁽²³⁾. This re-evaluation followed the principle adopted by the Scientific Advisory Committee On Nutrition/Committee On Toxicity⁽³⁾ of recognising the National Diet and Nutrition Survey data⁽²¹⁾ as being the most appropriate current estimate of consumption by adults except where there is strong

Table 2. Estimated mean intakes of EPA and DHA by adults in the UK (from Givens & Gibbs⁽²³⁾)

Food	Intake (g/week)*	Intake EPA + DHA (mg/d)
Fish		
White fish	104	38.8
Shellfish	27	14.2
Oil-rich fish	50	131
Other fish	36	14.2
Total fish	217	199
Meat		
Beef and veal	249	4.12
Sheep meat	51	2.05
Pork	63	1.34
Bacon and ham	105	1.24
Poultry	374	26.7
Sausages	68	0.26
Other products	216	1.29
Total meat	1126	37.0
Eggs	194	8.8
Total intake		244

*Intake of fish, meat and eggs based on data from Scientific Advisory Committee on Nutrition/Committee on Toxicity⁽³⁾, the National Diet and Nutrition Survey⁽²¹⁾ and British Egg Information Service⁽²²⁾ respectively.

evidence that other or adjusted values should be used. Based on the food intakes as given in Table 2, together with reported values for the concentrations of LC *n*-3 fatty acids in these foods (for details, see Givens & Gibbs⁽²³⁾), Table 2 provides mean estimates of the daily intake of LC *n*-3 fatty acids for UK adults. It is of concern that the mean intake is only about 54% of the target of 450 mg/d⁽³⁾. Of the total mean intake of 244 mg/d, approximately 54% is provided by oil-rich fish, but notably it is poultry meat that is the major contributor (73%) of all the meats. However, it is critical to realise that only about 27% of the adult population consume any oil-rich fish⁽³⁾ and thus for the vast majority of the adult population the daily intake will at best be approximately 100 mg, with almost half this amount provided by animal-derived foods. The contribution made by poultry meat may indeed be higher than this level if the consumption data for poultry meat reported by

the Association of Poultry Processors and Poultry Trade in the EU Countries⁽²⁴⁾ are a truer reflection of reality than those of the National Diet and Nutrition Survey⁽²¹⁾.

A key assumption in this analysis is, however, that the LC *n*-3 fatty acid content of poultry meat purchased by the public is similar to that observed in research studies. It is likely that much of the LC *n*-3 fatty acids found in poultry meat from birds that did not have fish oil in their diets is a result of the diet containing fishmeal, which contains some residual fish oil. In 2004 approximately 48 000 t fish meal (25% total use) was used in the UK for poultry diets⁽²⁵⁾, although this level of use has probably declined somewhat subsequently. There are now considerable amounts of poultry meat imported into the UK both from other EU Member States and from other parts of the world. Whether this imported meat will have similar background concentrations of LC *n*-3 fatty acids is not known, but a study is currently underway in the author's laboratory to analyse a range of poultry meat products at retail to obtain new data.

A number of studies have taken place recently to estimate EPA + DHA intake in various countries. A summary of these studies is given in Table 3. A large variation in mean intake is apparent between studies and countries but many values are considerably below the recommended target intakes. Some of the variation may be a result of the different methods for the collection of food consumption data, with some (for example, see Howe *et al.*⁽²⁶⁾) being based on 24 h recall and some (for example, see Givens & Gibbs⁽²³⁾) being based on 7 d weighed intakes. Two other points should be noted. First, in agreement with the study in UK adults, that with Belgian women shows that the majority of the population consume considerably less than the mean value⁽²⁷⁾. The lack of normality in the distribution of EPA + DHA intake across many populations has recently been highlighted⁽²⁸⁾, indicating the dangers of interpreting mean values derived from non-normally-distributed population data. Second, intakes of EPA + DHA appear to be generally lower in young adults (18–24 years) and children. It has been shown that in the UK at least there is a trend towards increased consumption of oily fish with increasing age, rendering young adults particularly vulnerable to suboptimal intakes of LC *n*-3 PUFA⁽²⁹⁾. The data for Belgian children⁽³⁰⁾ support this view. Low intakes

Table 3. Recent estimated daily intakes of EPA + DHA in various countries

Country	Details	Intake of EPA + DHA (mg/d)	Reference
UK	Adults, 19–64 years, mean	244	Givens & Gibbs ⁽²³⁾
UK	Females, 19–24 years, mean	109	Gibbs <i>et al.</i> ⁽²⁹⁾
Belgium	Females, 18–39 years, mean	209	Sioen <i>et al.</i> ⁽²⁷⁾
Belgium	Females, 18–39 years, median	50	Sioen <i>et al.</i> ⁽²⁷⁾
Belgium	Children, 4–6.5 years, mean	75	Sioen <i>et al.</i> ⁽³⁰⁾
France	Women, 45–63 years	344	Astorg <i>et al.</i> ⁽³¹⁾
Australia	Adults	143	Howe <i>et al.</i> ⁽²⁶⁾
North America	Adults	200	Vermunt & Zock ⁽⁶⁰⁾
Mid-Europe	Adults	250	Vermunt & Zock ⁽⁶⁰⁾
Northern Europe	Adults	590	Vermunt & Zock ⁽⁶⁰⁾
Japan	Adults	950	Vermunt & Zock ⁽⁶⁰⁾

of EPA + DHA in the young are a result of low or zero consumption of oil-rich fish, a habit that young adults may carry forward into middle and later life. It is interesting to note that in young women (19–24 years) the intake of canned tuna has been reported to be considerable⁽²⁹⁾, possibly in the belief that this product is a good source of fish oils.

In most studies the primary source of EPA and DHA is fish and seafood and thus variation in intake is a function of variation in consumption of these foods. Two studies have reported that meat, poultry and eggs contribute substantially to the intake of docosapentaenoic acid (22:5n-3)^(26,31), with docosapentaenoic acid contributing 29% total LC n-3 fatty acids consumed⁽²⁶⁾. These data highlight the need to better understand the physiological effects of dietary docosapentaenoic acid.

Options for increasing intake of EPA and DHA

Clearly, one option to increase intake of EPA and DHA is to encourage increased consumption of oily fish. However, given that young adults (18–24 years) appear to consume only small amounts if any, education in this area needs to start at a very young age and be built into an increased awareness of diet and health in general. Another option is to encourage the increased use of fish oil supplements such as capsules. However, data from the recent UK Low Income Diet and Nutrition Survey⁽³²⁾ indicates that habitual use of fish oil capsules in this population is very much less than that found in the National Diet and Nutrition Survey⁽²¹⁾, suggesting that encouragement to increase use would have less uptake in populations that are perhaps at greatest risk.

A further option is EPA and DHA enrichment of foods that are consumed in relatively large quantities by a large proportion of the population and that are amenable to enrichment. Animal-derived foods are key targets in this context since changes to the animals' diet can be used to bring about enrichment of the food products.

Enriching animal-derived foods with EPA and DHA

There have been many studies aimed at improving the EPA and DHA concentration in animal-derived foods in relation to chronic disease (for reviews, see Givens⁽³³⁾ and Pisulewski *et al.*⁽³⁴⁾), although few studies have attempted to connect the potential for enrichment with current and projected patterns of food consumption. Assuming that consumption of enriched foods would be the same as the current intake of normal foods, estimates have been made of the potential for enrichment of a wide range of animal foods and how these foods could contribute to additional EPA and DHA intake⁽²³⁾. The findings for milk and milk products, meat and eggs are summarised in Table 4, which shows that enrichment of animal-derived foods has the potential to provide a daily intake of EPA + DHA of about 230 mg, with poultry meat providing the largest potential intake (74 mg). Other useful contributions could be provided by eggs and full-fat cheese, although the average

Table 4. Potential mean intakes of EPA and DHA by adults in the UK from enriched animal-derived foods (from Givens & Gibbs⁽²³⁾)

Food	Intake (g/week)*	Concentration† (mg/g) of		Intake of EPA + DHA (mg/d)
		EPA	DHA	
Milk products				
Whole milk	337	0.106	0.141	11.9
Semi-skimmed milk	877	0.045	0.060	13.2
Skimmed milk	215	0.008	0.011	0.57
Cream	12	1.064	1.406	4.27
Other milk	42	0.080	0.105	1.12
Cottage cheese	9	0.104	0.137	0.31
Other cheese	98	0.745	0.984	24.2
Butter	22	2.181	2.882	16.0
Total milk products				71.5
Meat				
Beef and veal	249	0.24	0.053	10.4
Sheep meat	51	0.82	0.97	13.0
Pork	63	0.13	0.167	2.67
Bacon and ham	105	0.072	0.093	2.47
Poultry	374	0.60	0.80	74.8
Sausages‡	68	0.012	0.015	0.26
Other products‡	216	0.036	0.006	1.70
Total meat products				105.4
Eggs	194	0.06	1.90	54.3
Total intake				231

*Intake of milk and milk products and meat from the National Diet and Nutrition Survey⁽²¹⁾ and eggs from British Egg Information Service⁽²²⁾.

†Values for milk based on Chilliard *et al.*⁽⁴¹⁾, beef from Scollan *et al.*⁽⁵¹⁾, sheep meat from Cooper *et al.*⁽⁶¹⁾, poultry meat from Rymer & Givens⁽³⁷⁾ and eggs from Simopoulos⁽⁶²⁾.

‡Unchanged relative to non-enriched.

contributions from liquid milk and other meats are likely to be modest based on current food consumption data.

Enrichment of poultry meat

The EPA and DHA content of poultry meat can in theory be relatively easily modified by dietary means. As early as 1963 it was noted that the fatty acid compositions of broilers' breast, thigh and skin tissues are similar to those of the broilers' diet⁽³⁵⁾, and it was demonstrated that feeding fish oil to turkeys increases the concentrations of EPA and DHA in their depot fat and muscle lipids⁽³⁶⁾. A considerable amount of work has been done to enhance the EPA and DHA content of poultry meat by dietary means in ways that will result in nutritionally-meaningful intakes of these fatty acids by individuals who consume these products (for review, see Rymer & Givens⁽³⁷⁾).

Despite the volume of work, there are few data relating to the relative ability to enrich the meat of modern genotypes of broiler chickens and turkeys⁽³⁷⁾. A study was therefore carried out to determine the effect of different species and genotypes of poultry on their response as measured by increases in the EPA and DHA content of their edible tissues to increased concentrations of fish oil in their diet⁽³⁸⁾. Some key findings for skinless white chicken meat are shown in Table 5. Overall, the results show that in modern broiler genotypes there is no significant difference in the

Table 5. Effect of fish oil in the diet and breed of broiler chicken on the mean EPA and DHA concentration (mg/100g meat) in white chicken meat (from Rymer & Givens⁽³⁸⁾)

Diet* ...	Control		Lofish		Hifish		Statistical significance (<i>P</i>) of:	
	Ross 308	Cobb 500	Ross 308	Cobb 500	Ross 308	Cobb 500	Breed	Diet
EPA	7.5	6.9	17.4	20.0	27.2	30.8	NS	<0.001
DHA	39.6	38.6	54.9	64.3	118	126	NS	<0.001

*Contained fish oil at (g/kg): control, 0; lofish, 20; hifish, 40.

efficiency with which EPA and DHA are incorporated into edible tissue. There is also little evidence to suggest that there is any inherent difference between broilers and turkeys in their ability to incorporate EPA and DHA into their edible tissues, except perhaps for EPA in white meat. There is evidence that white chicken meat is a richer source of DHA than dark meat and also has greater enrichment efficiency than dark meat for DHA. This finding is promising since in the EU the consumption of white poultry meat far outweighs the consumption of dark meat. This result is not perhaps surprising as EPA and DHA preferentially accumulate in the phospholipids, which are much more prevalent in the white meat compared with the dark meat. As shown in Table 5, diets containing 40 g fish oil/kg give rise to white chicken meat containing about 140–160 mg EPA+DHA/100 g, which has the potential to make a real contribution to dietary EPA+DHA intake.

The same study also examined the birds' ability to convert dietary ALNA to EPA and DHA and then deposit these fatty acids in the edible tissues⁽³⁸⁾. The evidence from this experiment suggests that, as in man, the process is extremely limited, which is in agreement with the conclusion of an earlier study⁽³⁹⁾. The latter study suggests that although the birds may be capable of converting ALNA to EPA and DHA to some extent, these acids are not then deposited in skeletal muscle but rather sequestered in the liver or transported to other tissues. It therefore seems that ALNA cannot be used to make worthwhile enrichment of EPA and DHA in poultry edible tissues and that currently reliance continues to be on the use of fish oil.

There are some potential drawbacks to enriching poultry meat with EPA and DHA. These factors are the potentially reduced oxidative stability and hence shelf-life of the products and the possible negative effect that EPA and DHA enrichment may have on the organoleptic qualities of poultry meat. Current work in the author's laboratory indicates that most problems of this type can be overcome by the use of additional vitamin E in the diet of the bird.

Enrichment of milk

As a result of extensive biohydrogenation in the rumen and the inability of ruminant tissue to synthesise PUFA, typical levels of linoleic acid (18:2 n -6) and ALNA in milk fat are extremely low. Even when high amounts of PUFA from plant oils and oilseeds are included in the diet, absolute increases in linoleic acid and ALNA are relatively small. In relation to EPA and DHA, milk from cows fed conventional diets based on forages and cereal-based concentrates has extremely low concentrations (typically <1 g/100 g

Table 6. Effect of including fish oil in the diet of the dairy cow on EPA, DHA, *trans*-18:1 and conjugated linoleic acid (CLA) in milk fat (from Shingfield *et al.*⁽⁴²⁾)

Fatty acids (/100 g total fatty acids)	Control diet	Diet containing herring and mackerel oil (250 g/d)
EPA (mg)	50	110
DHA (mg)	0	100
Total <i>trans</i> -18:1 (g)	4.5	14.4
Total CLA (g)	0.39	1.66

fatty acids⁽⁴⁰⁾. It is possible to increase levels of EPA and DHA in milk fat by including some fish oil in the diet of the cow, although the extent of enrichment in milk fat is very low, with a typical efficiency of transfer of EPA and DHA from the diet into milk of 2.6% and 4.1% respectively⁽⁴¹⁾. These values are much lower than the transfer efficiencies of 18–33% and 16–25% seen for EPA and DHA respectively when fish oil is infused post-ruminally⁽⁴¹⁾. The poor transfer of EPA and DHA into milk when marine lipids are fed arises from extensive (between 74% and 100%) biohydrogenation in the rumen (for example, see Shingfield *et al.*⁽⁴²⁾) and preferential partitioning of these fatty acids into plasma phospholipids and cholesteryl esters, which are poor substrates for mammary lipoprotein lipase⁽⁴³⁾. Table 6 shows the typical effect of including fish oil in the diet of the cow on EPA and DHA concentrations. Whilst milk from the fish oil-containing diet is to some extent enriched with EPA and DHA, a side effect of this process is the substantial increase in the *trans*-fatty acids and conjugated linoleic acid content of the milk fat. Unlike industrially-hydrogenated products the majority of the increased *trans*-fatty acids is *trans*-vaccenic acid (*trans*-11 18:1)⁽⁴²⁾. Whilst most evidence indicates that *trans*-vaccenic acid is not a risk factor for CVD⁽⁴⁴⁾, there are few data from human studies, which have mostly evaluated *trans*-fatty acids from industrial sources. A study is currently underway to directly compare the effects of *trans*-fatty acids from milk and industrial sources on CVD risk factors in healthy human subjects⁽⁴⁵⁾.

There have been various approaches developed to protect fish and other marine lipids from biohydrogenation in the rumen, including encapsulation of oils and the creation of calcium salts of fatty acids or fatty acyl amides. Most of these technologies have been developed to overcome the negative effects on animal performance of feeding high levels of lipid, but can also allow sizeable and strategic changes in milk fatty acid composition.

Another approach is to directly fortify milk with fish oils during processing to provide EPA and DHA, and in theory this process is much more efficient and controllable than inclusion in the diets of dairy cows. Over the last 10 years milk fortified with EPA and DHA has been available commercially in several countries. In the UK, for example, the St Ivel Advance[®] brand (Dairy Crest Group plc, Esher, Surrey, UK) markets a fortified whole milk and a semi-skimmed milk that contain respectively 113 and 63 mg EPA+DHA/250 ml⁽⁴⁶⁾, and in Spain Puleva omega3 (Puleva Food SL, Madrid, Spain) provides approximately 66 mg EPA+DHA/100 ml⁽⁴⁷⁾. The effect of consuming the latter product on CVD risk factors has shown positive effects⁽⁴⁸⁾, although the milk used in this study was also fortified with oleic acid, folic acid and other vitamins and had its SFA content reduced substantially.

Overall, enriched and fortified milk products are likely to provide generally small increases in EPA+DHA intakes at normal levels of consumption, but as noted earlier many populations have substantially suboptimal intakes, and for them the availability of such milk may be very valuable.

Enrichment of eggs

Eggs enriched with EPA and DHA can also be produced by the addition of fish oil to the diet of the hen. A study has been carried out involving diets containing no additional lipid (control), 150 g fish oil/kg or 50, 100 or 150 g linseed/kg⁽⁴⁹⁾. The *n*-3 PUFA were found to be higher in eggs produced from hens fed fish oil or linseed compared with the control. In another study that used linseed to increase the *n*-3 PUFA content of eggs the work was extended to examine the effects of consumption of the eggs produced on plasma and platelet lipids in male subjects⁽⁵⁰⁾. Diets containing 0, 100 and 200 g ground linseed/kg were used and key results are summarised in Table 7. A progressive increase in the ALNA concentration of the eggs was observed relative to linseed inclusion. EPA was not found to be increased but DHA concentration was increased, although no significant difference was found between the two rates of linseed inclusion (DHA content equivalent to 51, 81 and 87 mg per egg respectively for 0, 100 and 200 g linseed/kg diet). It thus seems that laying hens have some capacity to synthesise DHA from ALNA, with little restriction of the process at the EPA level, a mechanism presumably designed to provide the embryo and chick with a source of DHA. Four eggs per d from each treatment were subsequently fed to the subjects for 2 weeks. No significant effects were seen in total cholesterol, HDL-cholesterol or plasma TAG concentrations but increases in total *n*-3 fatty acids and DHA contents of platelet phospholipids were recorded in subjects who ate the eggs from the diets containing linseed. Thus, eggs modified by the inclusion of sources of ALNA in the diet of the hen could provide a useful source of EPA and especially DHA without the use of fish oils. Some studies have shown that enhanced concentrations of EPA and DHA can also occur in ruminant meat as a result of *in vivo* synthesis from dietary ALNA (for example, see Scollan *et al.*⁽⁵¹⁾), although the efficiency of this process is normally very low and it is not likely that this route could be used with confidence.

Table 7. Effect of including linseed in the diet of laying hens on EPA, DHA and other fatty acids in the lipid fraction of eggs (from Ferrier *et al.*⁽⁵⁰⁾)

Fatty acid (/100 g total fatty acids)	Linseed inclusion in diet of hens (g/kg)		
	0	100	200
α -Linolenic acid (g)	0.5 ^a	5.5 ^b	10.7 ^c
EPA (mg)	100	200	200
DHA (mg)	1000 ^a	1700 ^b	1800 ^b
Total <i>n</i> -3 (g)	2.4 ^a	8.2 ^b	13.5 ^c
Total <i>n</i> -6 (g)	21.4 ^a	17.7 ^b	18.2 ^b
<i>n</i> -6: <i>n</i> -3	9.3 ^a	2.2 ^b	1.4 ^c
Total SFA (g)	32.3 ^a	31.7 ^a	28.2 ^b

^{a,b,c}Values with unlike superscript letters were significantly different ($P < 0.05$).

Alternatives to the use of fish oil

Essentially, all approaches to increase intake of EPA and DHA rely directly or indirectly on the use of fish oils. There are concerns, however, that the continued and possibly increased use of fish oils in the food chain is not sustainable and that alternatives are needed. Although some data are available (for review, see Givens *et al.*⁽⁵²⁾), further work on the potential of industrially-produced microalgae as dietary sources of EPA and DHA would seem warranted, although to date this process has proved to be very expensive.

A reason for the poor conversion of ALNA to EPA appears to be low activity of $\Delta 6$ desaturase⁽⁵³⁾, and thus an alternative strategy for increasing EPA supply would be to provide the product of $\Delta 6$ desaturase, stearidonic acid (18:4*n*-3). A number of recent studies (for example, see Miles *et al.*⁽⁵⁴⁾) do indeed indicate that dietary stearidonic acid may be a useful means of increasing the EPA content of human lipids. Although certain oils, such as that in the seed of *Echium plantaginium*, contain some stearidonic acid (approximately 13 g/100 g total fatty acids), this concentration may not be high enough to make diet manipulation easy, and currently the plant is not of major agricultural importance. The potential for discovering plants with much higher concentrations of stearidonic acid in their seed oil would seem large.

It is also noteworthy that efforts are ongoing into the genetic modification of certain plants in order that they will synthesise EPA and DHA in their seeds from shorter-chain precursors⁽⁵⁵⁾. This process involves the introduction of cloned algal genes into the plant. If successful and accepted by the consumer this approach could prove to be a major breakthrough in the long term.

Reducing intake of *n*-6 fatty acids

A number of studies, including the review of Ailhaud *et al.*⁽⁵⁶⁾, have concluded that over the last 40–60 years intake of *n*-6 fatty acids, notably linoleic acid, has increased very substantially in most Western societies, leading to a much increased dietary *n*-6 fatty acids:*n*-3 fatty

acids. This situation has led to concerns that competition between *n*-6 fatty acids and ALNA for the $\Delta 6$ desaturase enzyme has led to reduced efficiency of conversion of ALNA to EPA. This area has recently been reviewed by an expert group with the conclusion that the *n*-6 fatty acids: *n*-3 fatty acids is not a useful concept and distracts attention away from increasing absolute intakes of LC *n*-3 fatty acids⁽⁵⁷⁾. However, some evidence was reported that intake of linoleic acid can influence the proportion of EPA and DHA in membrane lipids, with higher intakes lowering the proportion of EPA and DHA. The long-term effect of the substantially increased intakes of linoleic acid seen over the last half century would seem to warrant further attention.

Acknowledgements

This work was supported by LIPGENE, an EU Sixth Framework Programme Integrated Project (2004–2009) (<http://www.lipgene.tcd.ie>).

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