ESSENTIAL AND *TRANS*-FATTY ACIDS IN NUTRITION

T. A. B. SANDERS

Department of Food and Nutritional Sciences, King's College, University of London, Campden Hill Road, London W8 7AH

CONTENTS

INTRODUCTION

Polyunsaturated fatty acids can be divided into two categories, essential (EFA) and non-essential. For the purpose of the present review the EFA are defined as linoleic ($18:2\omega6$) and linolenic ($18:3\omega3$) acids and their respective derivatives. The non-essential fatty acids include *trans*-isomers resulting from the hydrogenation of fats. Kummerow (1986) has

https://doi.org/10.1079/NRR19880007 Published online by Cambridge University Press

suggested that *trans*-isomers are undesirable constituents of human diets. The EFA are an example of a class of nutrients which in small amounts are indispensible and in larger amounts can modify disease processes. The present review discusses new developments concerning the role of polyunsaturated fatty acids in nutrition and attempts to explain their physiological functions.

ESSENTIAL FATTY ACIDS

DIETARY SOURCE OF EFA

Linoleic acid (18:2ω6) is the predominant polyunsaturated fatty acid in most culinary vegetable oils. γ-Linolenic acid (18:3ω6) is a minor constituent except in linseed, soya-bean and rapeseed oil; it is the major fatty acid in chloroplasts and so is also supplied by green leafy material. α-Linolenic acid (18:3ω3) is found in a few obscure vegetable oils such as evening primrose (*Oenothera biennis*), borage (*Borago officinalis*) and blackcurrant (*Ribes nigrum*) seed oils (8, 20 and 15% of total fatty acids respectively). The fatty acid composition of fat from simple-stomached animals reflects the composition of their dietary fat. Thus pigs fed on maize oil will deposit fat high in 18:2ω6. The fat from ruminants always has a low content owing to biohydrogenation of dietary polyunsaturated fatty acids in the rumen. The phospholipids and cholesterol esters from animals tissues contain derivatives of 18:2ω6 and 18:3ω3, notably arachidonic acid (20:4ω6), eicosapentaenoic acid (20:5ω3) and docosahexaenoic acid (22:6ω3). Eggs and offal especially, liver and brain, contain significant amounts of C_{20-22} polyunsaturated fatty acids but the highest concentrations are found in fish and fish oils. Fish oils also contain C_{16-18} polyunsaturated fatty acids with up to four double bonds.

Average adult intakes of 18:2ω6 in the United Kingdom are approximately 10 g/d (about 4% of the energy intake), 18:3ω3 intakes are in the region of 0·5–1·5 g/d (Roshanai & Sanders, 1984; Thomson *et al.* 1988). Mediterranean countries such as Italy and Greece, where olive oil is the main culinary oil, also have similar intakes (Keys, 1970). Intakes are considerably greater in the United States, where the intake of 18:2ω6 has risen markedly since the late 1960s (Hunter & Applewhite, 1986). Most human diets only contain small amounts of 20:4ω6, about 0·1 g daily obtained from the consumption of offal, meat and eggs. O'Dea & Sinclair (1982) found considerable amounts of 20:4ω6 in the tropical seafood diet consumed by Australian aboriginees. The intake of 20:5ω3 and 22:6ω3 depends on the consumption of offal and fish. The diets of Greenland Eskimos provide 5 and 6 g 20:5ω3 and 22:6ω3 respectively/d (Bang & Dyerberg, 1980), whereas Japanese fishermen consume approximately half these amounts (Hirai *et al.* 1980). A number of studies have investigated the effects of 20:5ω3 and 22:6ω3 using a fish-oil concentrate called MaxEPA (Seven Seas Health Care), which contains 1·8 g 20:5ω3 and 1·2 g 22:6ω3 /10 g; the oil is low in vitamin A and contains additional vitamin E and antioxidants.

EFA DEFICIENCY

Essential fatty acid deficiency (EFAD) in the rat is characterized by a scaly dermatitis, poor growth, an increased voluntary food intake and reproductive failure (Table 1). These symptoms result from defects in the structure of lipids or from an inability to form sufficient quantities of active eicosanoids (prostaglandins, thromboxanes, prostacyclins, leukotrienes).

Fatty acids with *cis* double bonds at C-6 C-9 from the terminal methyl group possess EFA activity (Fig. 1). Synthetic odd-chain fatty acids with ω5 or ω7 unsaturation also show some activity. Columbinic acid 18:3ω6-*cis*,9-*cis*,13-*trans* is able to restore growth and cure the dermal symptoms but as it cannot be converted into prostaglandins it does not correct

Table 1. *Symptoms of essential fatty acid deficiency in the rat*

	Symptom	Cured by:
Somatic functions	Poor growth Impaired energy utilization Structural abnormalies in membranes	Linoleic or linolenic
Skin	Scaly dermatitis Increased transepidermal water loss	Linoleic
Kidney	Abnormal papilla, apical necrosis, haematuria	Linoleic
Reproduction	Testicular degeneration Impaired parturition	Linoleic
Platelet	Impaired aggregation	Linoleic
Immunity	Increased susceptibility to infection, decreased inflammatory response	Linoleic

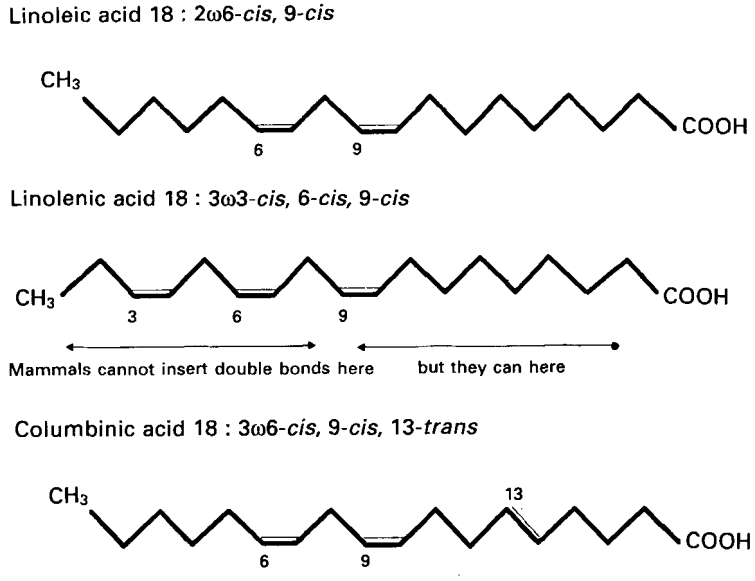

Fig. 1. Structure of polyunsaturated fatty acids with essential fatty acid activity.

those symptoms that are dependent upon prostaglandin production such as reproduction, haemostasis and inflammation (Houtsmuller, 1981).

Linoleic acid (18:2ω6) cures all the symptoms but α-linolenic acid (18:3ω3) only restores the growth. Arachidonic (20:4ω6) and γ-linolenic (18:3ω6) acid are 3 and 1·5 times more potent respectively than 18:2ω6 in curing EFAD in the rat (Rivers & Frankel, 1981).

A requirement for 18:2ω6 has been established in all species studied. Between 0·5 and 1% of the energy intake provided as 18:2ω6 will prevent classical EFAD, and 18:3ω3 provided at 0·5% of the energy intake will restore growth in EFA-deficient rats (Pudelkewicz *et al.* 1968). EFA requirements are greater in male than in female rats. Linoleic acid may be conserved for its essential function more efficiently in females due to abnormal effects on fatty acid-binding proteins (Hagve & Christophersen, 1987).

Classical EFAD does not occur naturally in man consuming self-selected diets but can occur under abnormal circumstances such as in infants fed on skimmed milk, kwashiorkor,

https://doi.org/10.1079/NRR19880007 Published online by Cambridge University Press

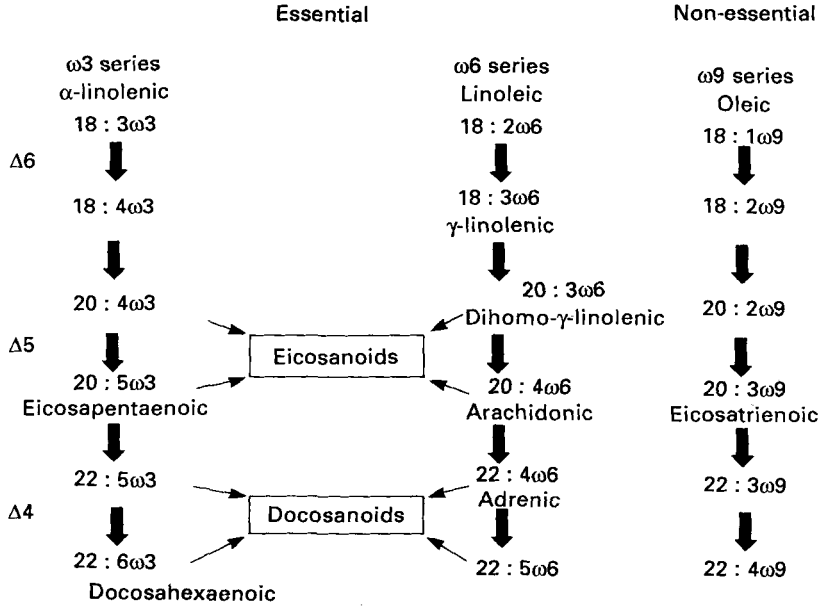

Fig. 2. Metabolism of polyunsaturated fatty acids.

chronic fat malabsorption, total parenteral nutrition and in patients receiving nasogastric feeds (Yamanaka *et al.* 1980). Total parenteral nutrition can result in rapid EFAD because high glucose concentrations inhibit lipolysis and so isolate the patient from the store of 18:2ω6 in adipose tissue. Intravenous lipid emulsion or cutaneous application of safflower oil can be used to prevent EFA.

METABOLISM OF EFA

Neither linoleic (18:2ω6) nor linolenic (18:3ω3) acid can be synthesized in mammalian tissues but both can undergo further desaturation and chain elongation to give two series of derivatives, the ω6 and ω3 series (Fig. 2). Competitive inhibition occurs between the ω3 and ω6 series of polyunsaturated fatty acids and the balance of derivatives is determined by the ratio, 18:2ω6:183ω3 in the diet.

Species differences exist in the capacity to convert the parent EFA to their longer-chain derivatives. The cat is unable to synthesize 20:4ω6 in sufficient quantities and has a dietary requirement for 20:4ω6. It lacks Δ6 desaturase and can only form 20:4ω6 from 18:2ω6 via an alternative pathway involving Δ8 desaturation of 20:2ω6 (MacDonald *et al.* 1984). It has been argued that the activity of Δ6 desaturase may be low in man under certain conditions and that this 'metabolic block' may be overridden by consuming 18:3ω6 (Manku *et al.* 1982). Studies of vegans, whose diets are devoid of derived EFA, suggest that the conversion of 18:2ω6 to 20:4ω6 occurs readily in man but that of 18:3ω3 into 22:6ω3 may be limited by a slow rate of Δ4 desaturation (Sanders *et al.* 1978). The proportion of 22:6ω3 in plasma phospholipids was not increased by additional 18:3ω3 in the diet (Sanders & Younger, 1981). Moreover, dietary 20:5ω3 fails to increase the proportion of 22:6ω3 in plasma and erythrocyte phospholipids (Von Schacky & Weber, 1985).

The extent to which the parent EFA are converted to more unsaturated derivatives also depends on the affinity of the parent fatty acid for other metabolic pathways. Chain-length and degree of unsaturation influence the rate of oxidation of polyunsaturated fats and

affinity for acyl transferases. The rate of oxidation of fatty acids decreases with increasing chain length. Linoleic is oxidized at a faster rate than γ-linolenic or 20:4ω6. However, 18:3ω3 is oxidized at a faster rate than 18:2ω6, this may be because 18:3ω3 is not readily incorporated into phospholipids and 20:4ω6 and 22:6ω3 are avidly incorporated into phospholipids and are poor substrates for β-oxidation. It has become clear that the consumption of preformed derivatives of EFA have different effects from the consumption of the parent EFA.

In the absence of EFA from the diet, oleic acid which can be synthesized de novo is converted to eicosatrienoic acid (20:3ω9) (Fig. 2). The presence of 20:3ω9 is regarded as the hallmark of EFA deficiency. The ratio, 20:3ω9/20:4ω6 (triene:tetraene ratio) has been used as a biochemical indicator of EFA deficiency. Holman *et al.* (1979) initially suggested that a ratio of greater than 0·4 was indicative of deficiency, but now suggests that a ratio of 0·2 be taken as the upper limit of normality. The ratio, 20:3ω9/20:4ω6 in plasma phospholipids is probably the most sensitive measure of 18:2ω6 status (Press *et al.* 1974). There are instances when the measurement ratio is inappropriate; for example, when the formation of 20:3ω9 is limited owing to inactivity of the necessary desaturase or when significant amounts of dietary ω3 polyunsaturated fatty acids are present in the diet.

EICOSANOIDS

The terms eicosanoid and docosanoid are used to describe products resulting from the enzymic oxidation of C_{20} and C_{22} polyunsaturated fatty acids respectively. Esterified fatty acids must first be liberated before they can act as substrates for eicosanoid synthesis. Liberation of substrate from phospholipids occurs when tissues are stimulated or damaged. It appears that initially substrate is derived from the phosphatidyl inositol fraction. Hydroperoxide formation may then amplify eicosanoid biosynthesis and stimulate the release of further substrate from other phospholipid pools. Circulating free fatty acids may also be substrates for eicosanoid synthesis. This might explain why the consumption of 20:4ω6 leads to a rapid increase in basal prostaglandin production (Seybeth *et al.* 1975).

Arachidonic acid is the major eicosanoid precursor in most tissues (Fig. 3) but dihomogamma-linolenic (20:3ω6) and adrenic (22:4ω6) acids also give rise to prostaglandins in some tissues. Generally hydroxy fatty acids derived from 20:4ω6 are chemotactic towards leucocytes. Leukotriene B4 is leucotactic, leukotrienes C_4, D_4, and E_4 (the sulphidopeptide leukotrienes) are synonymous with the slow-releasing substance of anaphylaxis (SRS-A). Eicosanoids with opposing actions produced in different locations are involved in tissue homoeostasis. As a rule eicosanoids are produced, degraded and have their action locally. Several hormones stimulate the production of eicosanoids. For example, angiotensin II stimulates the production of PGE_2 and TxH_2 by renal glomeruli (Lefkowith & Schreiner, 1987). Eicosanoid metabolites of 20:4ω6 have been shown to be involved in the regulation of intraocular pressure, bone calcification, cell-mediated immunity, insulin release, renal function, reproduction, haemostasis and inflammation (Moncada & Vane, 1979; Poyser, 1981; Lewis, 1983).

Much of our understanding about prostaglandins has been derived from the study of anti-inflammatory drugs, such as aspirin and indomethacin, whose mechanism of action is the inhibition of cyclo-oxygenase. Certain polyunsaturated fatty acids can inhibit the production of eicosanoids from 20:4ω6. For example 20:5ω3 besides acting as a substrate also acts as a competitive inhibitor of cyclo-oxygenase and 5-lipoxygenase. Some naturally occurring polyunsaturated fatty acids are more potent cyclo-oxygenase inhibitors than indomethacin, in particular 18:3ω9-*cis*,11-*trans*,14-*cis* (Nugteren & Christ-Hazelhof, 1987). This might explain the toxicity of some plants to animals.

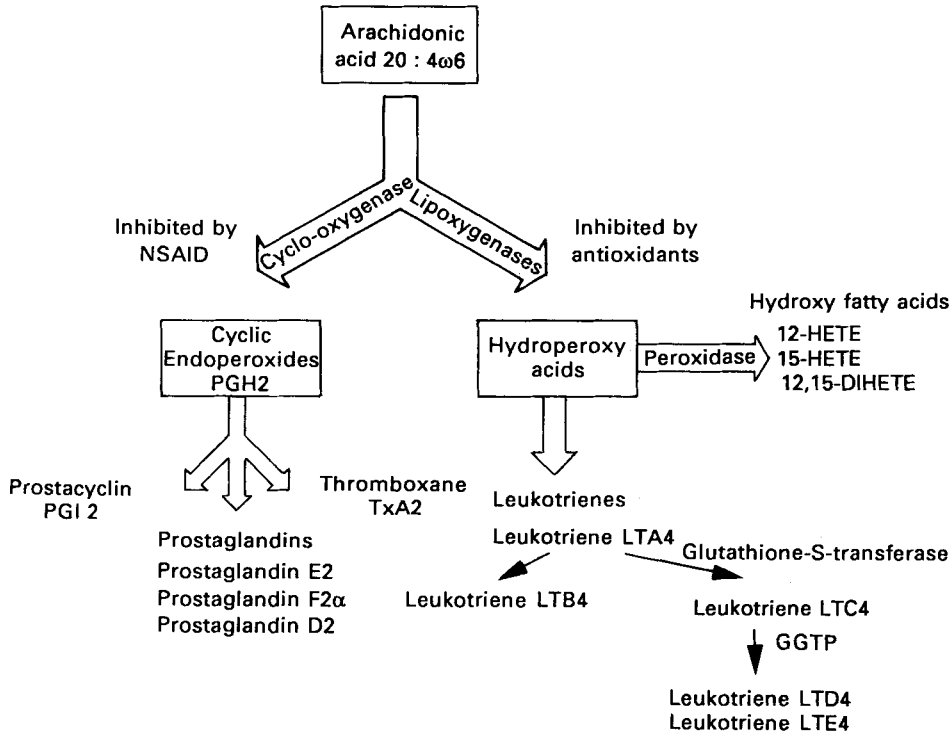

Fig. 3. Conversion of arachidonic acid into eicosanoids. HETE, hydroxyeicosatetraenoic acid; DIHETE, dihydroxyeicosatetraenoic acid; NSAID, non-steroidal anti-inflammatory drugs; GGTP, γ-glutamyl transpeptidase (*EC* 2 . 3 . 2 . 2).

EFA AND REPRODUCTIVE FUNCTION

Rats fed on EFA-deficient diets show testicular degeneration which can be prevented by dietary 18:2ω6 but not by ω3 fatty acids (Leat *et al.* 1983). Semen contains relatively high concentrations of prostaglandins PGE_1 and PGE_2 (Poyser, 1981). Prostaglandin PGE_2 prevented infertility in rats fed on an EFA-deficient diet (Hafiez, 1974). As PGE_3 and PGF_3 are inactive this probably explains why ω3 EFA as the only source of dietary EFA cannot support normal testicular development in the male.

EFA-deficient animals are unable to give birth to their young owing to an inability to produce sufficient prostaglandin F_2, which causes uterine contractions. Reproductive function in the rat cannot be restored with 18:3ω3. This is because prostaglandins PGE_3 and PGF_3 are relatively inactive. 18:3ω3 on the other hand is able to sustain normal fetal growth throughout pregnancy (Leat & Northrop, 1981). There is no evidence to suggest that the consumption of fish oil leads to fetal abnormalities. Decreased prostaglandin E_2 production by the uterus and delayed parturition have been reported in animals fed on fish oil as the only source of fat (Leaver *et al.* 1986). Premature birth is associated with a high risk of perinatal death. Increased rates of prostaglandin PGE_2 production are associated with premature birth. The onset of labour can be delayed by compounds that inhibit the formation of PGE_2. Prostaglandin PGE_2 can be used to induce labour. Prolonged gestation and increased birth weight have been observed among the population of the Faro Isles and it has been argued that this may be due to the consumption of 20:5ω3 and 22:6ω3 during pregnancy (Olsen *et al.* 1987).

https://doi.org/10.1079/NRR19880007 Published online by Cambridge University Press

Table 2. *Physiological properties of eicosanoids derived from arachidonic (20:4ω6) and eicosapentaenoic (20:5ω3) acid*

	20:4ω6	20:5ω3
Platelet	TxB_2, PGH_2 Proaggregatory vasocontrictor	TxA_3, PGH_3 Weakly proaggregatory Weak vasocontrictor
Endothelium	PGI_2 Anti-aggregatory vasodilator	PGI_3 Anti-aggregatory vasoldilator
Uterus	PGE_2, PGF_2 Powerful smooth muscle contractor	PGE_3, PGF_3 Weak smooth muscle contractor
Neutrophil	LTB_4 Strongly chemotactic	LTB_5 Weakly chemotactic
Lung	LTC_4, LTD_4 Bronchoconstrictor	LTC_5, LTD_5, LTE_5 Bronchoconstrictor

TxA_2, TxB_2, TxA_3, TxB_3, thromboxanes; PGH_2, PGH_3, cyclic endoperoxide; PGI_2, PGI_3, prostacyclins; PGE_2, PGE_3, PGF_2, PGF_3, prostaglandins; LTB_4, LTB_5, LTC_4, LTC_5, LTD_4, LTD_5, LTE_4, LTE_5, leukotrienes.

EFA AND HAEMOSTASIS

The role of prostaglandins in haemostasis is relatively well understood (Moncada & Vane, 1979). Thromboxane TxA_2 is the major prostaglandin produced by activated platelets. This compound stimulates platelet aggregation and causes vasoconstriction. Prostacyclin PGT_2 produced by the vascular endothelium antagonizes the effects of thromboxane TxA_2 and therefore localizes platelet aggregation to the site of injury.

Linoleic acid deficiency leads to a decrease in the proportion of 20:4ω6 in platelets. This is accompanied by a decreased capacity to synthesis thromboxane and the inhibition of second-phase platelet aggregation. Dietary 18:2ω6 restores thromboxane production and normalizes second-phase platelet aggregation. Further increasing the intake of 18:2ω6 above that required to correct EFA deficiency leads to further increases in thromboxane A_2 production and thrombotic tendency; although at very high intakes of 18:2ω6 (more than 15% of energy intake) thrombotic tendency is reduced. Thus, in the range of human intakes at 2–10% of energy intake, an increase in dietary 18:2ω6 leads to an increase in thromboxane A_2 production and thrombotic tendency (Sanders, 1983).

It has been argued that the ω3 fatty acids play an important role in modulating the production of active eicosanoids from the ω6 series (Leaf & Weber, 1988). Significant reductions in thromboxane A_2 production can be obtained with moderate intakes of fish oils containing 20:5ω3 and 22:6ω3. The partial replacement of arachidonic with 20:5ω3 in membrane lipids decreases the production of active eicosanoids in two ways: (1) by leading to the generation of less-active eicosanoids derived from 20:5ω3 (Table 2); (2) by inhibiting the formation of eicosanoids from 20:4ω6 (22:6ω3 contributes to this effect by acting as an inhibitor of cyclo-oxygenase). It has also been claimed that dietary 22:6ω3 is retroconverted to 20:5ω3 and transformed into prostacyclin PGI_3 (Fischer *et al.* 1987).

Template bleeding time is prolonged and leukotriene B_4 and thromboxane A_2 production are decreased by intakes of 20:5ω3 and 22:6ω3 in excess of 3–5 g/d (Sanders, 1987). The prolongation of template bleeding time is not related to a decrease in platelet counts or changes in the levels of clotting factors. Some but not all controlled trials have reported a slight reduction in blood pressure with fish-oil supplements (Norris *et al.* 1986). Unlike

treatment with non-steroid anti-inflammatory drugs, the protective prostacyclin PGI_2 is not decreased in man following fish-oil supplements (Knapp *et al.* 1985). Prostacyclin PGI_3 has also been reported in fish-eating populations.

EFA AND CARDIAC FUNCTION

EFA deficiency results in electrocardiographic abnormalities notably a notching in the QRS complex. This symptom indicates impaired electrical conduction and can be corrected by either dietary 18:2ω6 or 18:3ω3 (Caster & Ahn, 1963). Cardiocytes only form small amounts of prostaglandins and do not exhibit lipoxygenase activity (Hohl & Rosen, 1987). Consequently it appears that the impaired conduction defect is due to a deficit of structural lipid rather than an inability to produce eicosanoids. It is possible to manipulate the proportions of 18:2ω6, 20:4ω6 and 22:6ω3 in cardiac phospholipids by diet. Jenkins & Ewan (1967) reported cardiomyopathy in rats fed on cod-liver oil, which was attributed to lipid peroxides in the oil. The myopathy was mitigated by additional selenium or vitamin E. Hulan *et al.* (1977) suggested that the high level of 22:6ω3 in the hearts of rats fed on low-erucic-acid rapeseed oil was related to their high risk of myocardial fibrosis. Kramer *et al.* (1985) showed that increasing the intake of saturated fat reduced the incidence of lesions in rats without influencing the proportion of 22:6ω3 in cardiac lipids. The myocardial necrosis observed in animals fed on low-erucic-acid rapeseed oil may well be related to damage caused by lipid peroxides.

Prostacyclin PGI_2 is the main prostaglandin produced by the isolated heart and this is produced by the vascular tissue. Ten Hoor *et al.* (1980) showed that increasing the dietary intake of 18:2ω6 led to an increase in cardiac prostacyclin production and coronary flow and that these effects could be abolished by aspirin or indomethacin. Animals fed on fish oils containing 20:5ω3 and 22:6ω3 or linseed oil, containing 18:3ω3, showed a decreased capacity to produce prostacyclin (Ten Hoor *et al.* 1980). At that time it was argued that prostaglandins were cardioprotective. The wisdom of this view may be questioned by the following observations: the rates of prostacyclin and thromboxane production are greater in patients with atherosclerosis (Knapp *et al.* 1985); mortality from coronary heart disease (Antiplatelet Trialists' Collaboration, 1988) can be decreased by the prophylactic use of aspirin.

During an ischaemic episode, arachidonic acid is released from membrane phospholipids and is rapidly converted to eicosanoids by the vascular endothelium, platelets and leucocytes. The release of these compounds into the coronary venous effluent during ischaemia plays a profound role in determining the outcome of the ischaemic attack. The infusion of 20:4ω6 into the coronary circulation perfused with buffer leads to a biphasic response: the initial response is vasoconstriction accompanied by a decreased coronary flow followed by vasodilation and an increase in coronary flow. The initial response is probably caused by the formation of PGH_2 which is an intermediate in the synthesis of PGI_2. Infusion of either 18:2ω6 or 22:6ω3 decreases the vasoconstrictor response induced by 20:4ω6 (Talesnik, 1986).

Lepran *et al.* (1981) showed that a diet rich in 18:2ω6 reduced mortality from coronary artery ligation in rats but that this effect was reversed by indomethacin, which implies that prostaglandins were affording protection. However, the experimental drug BW755, which blocks both cyclo-oxygenase and lipoxygenase pathways, is cardioprotective in experimental coronary occlusion. Leukotriene B_4 is believed to be responsible for the massive accumulation of leucocytes in an infarcted area and its formation can be inhibited by BW755 (Mullane *et al.* 1984). Consequently, the abolition of the protective effect of 18:2ω6 with indomethacin might be due to the diversion of 20:4ω6 down the lipoxygenase

pathway. Infarct volume is reduced following experimental myocardial infarction in dogs fed on menhaden oil (Culp *et al.* 1980). Recent studies of experimental myocardial infarction in rats show that fish oil decreases cardiac damage to greater extent than maize oil (Hock *et al.* 1987). This effect could not be obtained in animals fed on an EFA-deficient diet, which suggests that the fish oil was exerting a protective effect.

Charnock and colleagues (Abeywardena *et al.* 1987*a*, *b*) have shown that susceptibility to cardiac arrhythmia is influenced by fat intake. Animals fed on sheep fat show a higher incidence of cardiac arrhythmia than those fed on sunflower oil (high in linoleic) or on tuna oil (high in 22:6ω3) (Abeywardena *et al.* 1987*a*). The animals fed on the sheep-fat diet were not EFA deficient and produced more prostacyclin and thromboxane than did the sunflower-oil-fed animals (Abeywardena *et al.* 1987*b*). The tuna oil, as expected, produced the lowest levels. If the main role of EFA in cardiocytes is structural then it might be advantageous for this role to be fulfilled by 22:6ω3 rather than 20:4ω6, as the latter exacerbates myocardial ischaemia. The influence of mackerel oil on cardiac lipids and function has recently been studied in pigs (Hartog *et al.* 1987) and no adverse effects on cardiac function were observed. Further studies are required to determine whether fish oil protects against reperfusion injury.

EFA AND MEMBRANE LIPIDS

Many cells in culture do not require EFA in order to grow (Spector *et al.* 1982). However, polyunsaturated fatty acids are required for the structural integrity of tissues. Houtsmuller (1973) found that 18:2ω6, 20:4ω6 or 22:6ω3 would correct the mitochondrial membrane abnormalities of EFA deficiency. It seems that derivatives of both ω6 and ω3 series can perform some of the membrane functions of EFA. Yet there may be functions that are specific to one series or another.

Unlike triglyceride structure which is clearly dependent on dietary intake, modification of membrane phospholipid-fatty acid composition is regulated. There are marked differences in the composition of phospholipid species within the same animal and there is specificity by different phospholipids for certain polyunsaturated fatty acids. However, the balance between the ω6 and ω3 fatty acids in membranes is determined mainly by the balance in the diet.

Changes in the balance between ω6 and ω3 fatty acids may effect the physical properties of the membrane as well as the activity of membrane-bound enzymes (Bernsohn & Spitz, 1974; Morson & Clandinin, 1986). The incorporation of 20:5ω3 into erythrocyte membranes has been shown to decrease the deformability of the membrane (Terano *et al.* 1983).

THE ROLE OF LINOLEIC ACID IN SKIN

EFA deficiency is characterized by a scaly skin and an increased rate of transepidermal water loss. The increased energy intake and expenditure in EFA deficiency can partially be explained by high transepidermal water loss and can be prevented by keeping the animals in a high humidity environment. Linoleic acid has a unique role in the skin. This is supported by the observation that 18:2ω6 prevents scaly skin and increased transepidermal water loss in EFA-deficient cats (MacDonald *et al.* (1983). Different mechanisms may be involved in causing these two symptoms.

The scaly dermatitis is a consequence of epidermal proliferation and is reflected by an increased rate of DNA synthesis (Sherertz, 1986). It can be corrected by dietary or topical linoleic or 20:4ω6 but not by ω3 EFA or *trans*-isomers of 18:2ω6. It is believed that the scaly dermatitis is caused by an inability to form a 13-hydroxy linoleic acid (Elliot *et al.*

1985*a*; Nugteren & Kivits, 1987) which is believed to be involved in the regulation of the differentiation of skin cells.

Sphingolipids rich in 18:2ω6 appear necessary for forming and maintaining the extracellular lamellae in the stratum compactum which are considered to be the barrier against water loss. Early reports suggesting that these lamellae were absent in EFA deficiency have recently been contradicted (Wertz *et al.* 1987). Acylglucosyl ceramides (AGC) produced in the stratum granulosum contain 18:2ω6 as their major fatty acid esterified onto the ω-hydroxyl group of C_{30-34} acyl side chain. The AGC are converted to acyl ceramides and to acyl acids as they migrate from the stratum granulosum to stratum compactum. The acyl acids and polyoxyacyl ceramides make up the lipid material in the lamellae. In EFA deficiency 18:2ω6 is replaced by 18:1ω9 in these lipid fractions. The increased transepidermal water loss can be corrected by dietary or topically applied linoleic, columbinic and 20:4ω6 but not by 18:3ω3 (Houtsmuller & Van der Beek, 1981; Hansen & Jensen, 1985). The evidence suggests the permeability barrier is not dependent on the formation of a lipoxygenase product (Elliot *et al.* 1985*b*). However, derivatives of the lipoxygenase pathway may be responsible for the normal differentiation of cells that lead to the synthesis of these lipids (Nugteren & Kivits, 1987). Until recently it was uncertain why 20:4ω6 when applied topically to EFA-deficient rats normalizes the transdermal water loss. It was suggested that dietary 20:4ω6 would displace 18:2ω6 from membrane lipids and make it available for AGC synthesis (Hansen & Jensen, 1985). Hansen *et al.* (1986) have shown that retroconversion of 20:4ω6 to 18:2ω6 occurs in the EFA-deficient rat.

USE OF EFA IN THE TREATMENT OF SKIN DISORDERS

Several cutaneous disorders characterized by epidermal proliferation, scaliness and inflammation may be influenced by the dietary intake of polyunsaturated fats. Some patients with atopic eczema have been observed to improve after treatment with 18:2ω6 (Sherertz, 1986). More recently evening primrose oil, which also contains γ-18:3ω3, has been claimed to be of benefit in atopic eczema (Wright & Burton, 1982). It has been suggested 18:3ω6 would be chain elongated in skin and converted to PGE_1, which has anti-inflammatory effects (Chapkin *et al.* 1987), because epidermal tissue cannot convert 20:3ω6 to 20:4ω6. Psoriasis shares with EFA deficiency abnormalities in the stratum corneum and barrier function. However, the levels of 20:4ω6 and its metabolites are elevated in psoriatic skin. Topical application of 18:2ω6 does not improve the clinical lesions nor reduce transepidermal water loss (Sherertz, 1986) but the administration of fish oil (MaxEPA, in excess of 10 g/d) leads to a mild improvement in the disorder (Bittiner *et al.* 1988). Fish-oil supplements may also be of value in preventing the hyperlipidaemia associated with retinoid therapy in skin disease (Marsden, 1987).

INFLUENCES OF EFA ON THE IMMUNE SYSTEM AND THE INFLAMMATORY RESPONSE

Eicosanoids play an important role in the regulation of the immune system. Cell-mediated immunity (CMI) is impaired in EFA deficiency and the severity of several autoimmune disorders is ameliorated by low intakes of 18:2ω6 or high intakes of 20:5ω3 and 22:6ω3 (Robinson *et al.* 1985). The relationship between 18:2ω6 intake and CMI may follow a bell-shaped curve with both low and high intakes being immunosuppressive (Mertin & Stackpoole, 1981). The immunosuppressive effect of high intakes of 18:2ω6 is abrogated by splenectomy and inhibited by indomethacin or antisera to prostaglandin E_1 (Mertin & Stackpoole, 1981). The injection of 18:2ω6 into animals increases the production of suppressor cells in the bone marrow (Young *et al.* 1987).

Eicosanoids are mediators of inflammation, in particular they cause leucocyte migration

and oedema. The inhibition of their formation is the mode of action of most non-steroidal anti-inflammatory drugs. The inflammatory response to carrageenan injection is reduced in EFAD and is corrected by 18:2ω6 but not by columbinic acid (Houtsmuller, 1981). The inflammatory response to carrageenan is also decreased by dietary 20:5ω3. It is of interest that although the heroic attempt of Brown to induce EFA deficiency failed, it cured his migraine (Brown *et al.* 1938). This may have been due to decreased production of eicosanoids. Fish-oil supplements, which also decrease the production of eicosanoids, have been claimed to be of value in the treatment of migraine (McCarren *et al.* 1985). Kremer *et al.* (1987) claim that fish-oil supplements are of therapeutic value in the management of rheumatoid arthritis. However, like aspirin fish-oil supplements can exacerbate asthma, probably by increasing the production of SRS-A (Ritter & Taylor, 1988).

One major side effect resulting from the use of non-steroidal anti-inflammatory drugs such as aspirin is gastric ulceration. Prostaglandins derived from 20:4ω6 have a powerful cytoprotective effect on the gastric mucosa. It has been argued that the fall in peptic ulcer in the United States and other Western countries might be related to an increase in the intake in 18:2ω6 over the past decade (Hollander & Tarnawski, 1986).

ESSENTIALITY OF ω3 FATTY ACIDS

A requirement for fatty acids of the ω3 series has been clearly established for certain insects and for certain fish (Tinoco, 1982). The position of ω3 fatty acids in mammalian nutrition was until recently in limbo. There were several reasons for this: the lack of a demonstrable deficiency syndrome; the apparent normal growth and reproduction of rats fed on diets lacking 18:3ω3. However, it may not be necessary to demonstrate the existence of a deficiency disease syndrome to establish the essentiality of a nutrient in the diet.

In all mammalian species 22:6ω3 is concentrated in the retina and in the brain. The highest concentrations are found in the rod-outer segment which contain 40–50 mol/100 mol as 22:6ω3. Brain and synaptic endings are similarly enriched in 22:6ω3. Recently, elongated homologues of 22:6ω3 notably 32:6ω3 have been found in retinal phosphatidyl cholines (Aveldano, 1987).

The rod-outer segment is responsible for transducing photons to electrical signals. Rhodopsin is an integral membrane protein and accounts for 90% of the protein in vertebrate rods. Its molecule spans the disc membrane bilayer and is half imbedded within it. Activation of rhodopsin leads to a conformational change and the high fluidity and flexibility of the disc membrane appears necessary for this dynamic behaviour. The hypothesis that the role of 22:6ω3 in the membrane was to increase fluidity has not been supported by studies of model membranes (Dratz & Deese, 1986). Indeed, these workers found that phosphatidyl cholines containing 22:6ω3 had an unexpectedly high melting point (22:6ω3 is believed to exist in a helical conformation in membrane). It has been argued that this may produce greater flexibility and compressibility and therefore lower resistance to conformational change in rhodopsin. It has also been suggested that the high 22:6ω3 content may increase the permeability of the disc to ions involved in neural signalling.

Wheeler *et al.* (1975) reported a reduction in electroretinogram amplitude in rats deprived of 18:3ω3. Leat *et al.* (1986) were unable to confirm this finding in rats or guinea-pigs. Impaired visual acuity and an abnormal retinogram have been reported in monkeys deprived of 18:3ω3. These changes were accompanied by marked reductions in the proportion of 22:6ω3 in retinal and cerebral cortex phospholipids and its replacement with 22:5ω6. The biochemical changes were shown to be reversible by supplementing the diet with fish oil (MaxEPA). However, the electroretinogram recordings did not recover, suggesting that the functional abnormalities were irreversible (Neuringer & Connor, 1987). Behavioural abnormalities and impaired learning ability have been reported in rats

(Lamptey & Walker, 1976; Yamamoto *et al.* 1987) and monkeys (Fiennes *et al.* 1973) deprived of 18:3ω3.

Although the arguments for a specific role for the ω3 fatty acids in man are persuasive, the reported cases (Holman *et al.* 1982; Bjerve *et al.* 1987*a*, *b*) of deficiency are not convincing.

Bjerve *et al.* (1987*b*) reported on two patients who had been fed by gastric tube. The patients developed thin atrophic skin with dermatitis on the shoulders which responded to supplements of cod-liver and soya-bean oil. Bjerve (1987*a*) also reported another patient who was treated with 0·1 ml ethyl linolenate orally. It is claimed that the patient's skin condition improved after 5 d. However, these patients showed low levels of 18:2ω6 in their plasma phospholipids, suggesting that their dietary intake of 18:2ω6 was extremely low. Their energy intakes were extremely low, so the conclusion drawn by the authors that these patients were not suffering from 18:2ω6 deficiency because they were obtaining more than 0·5% of their energy intake from 18:2ω6 does not seem justified. The study would have been more convincing had the subjects first been treated with dietary linoleate, in view of the known functions of 18:2ω6 in the skin.

If 22:6ω3 is indispensible then the essentiality of 18:3ω3 hinges on the extent to which it can be converted to 22:6ω3. Man can convert 18:3 to 20:5ω3 but the capacity to convert 18:3 to 22:6ω3 does appear limited (Sanders *et al.* 1978; Sanders & Younger, 1981). This might imply that preformed dietary 22:6ω3 is more important that 22:6ω3 derived from 18:3ω3.

The development of brain and retina occurs in utero and in infancy. It is relevant therefore to consider the metabolism of ω3 fatty acids in infants. Under natural conditions human milk supplies the infant with ω3 fatty acids. It has been argued that these fatty acids provided by breast-milk play an important role in meeting the essential fatty acid requirements of the human infant (Crawford & Sinclair, 1972). The fatty acid composition of human milk is variable and depends on the maternal dietary intake. On average, 18:3ω3 provides 0·4% of the energy intake. Human milk contains significant amounts of docosahexaenoic (22:6ω3). In vegans despite higher levels of 18:3ω3 in their breast-milk the proportion of 22:6ω3 was lower (0·2 v. 0·6%) than in omnivores (Sanders *et al.* 1978). The proportion of 22:6ω3 in breast-milk is increased following the consumption of fish oil (Harris *et al.* 1984). The levels of 22:6ω3 in erythrocyte lipids of breast-fed infants born to vegan mothers are considerably lower than those found in infants breast-fed by omnivores (Sanders *et al.* 1978). Lower proportions of 22:6ω3 in the erythrocyte phospholipids of infants fed on cow's milk formula have also been reported (Sanders & Naismith, 1979).

Putnam *et al.* (1982) compared the influence on erythrocyte lipids of infant milk formulas containing similar amounts of linoleic and 18:3ω3 as breast-milk. The breast-fed infants had levels of 22:6ω3 twice as high as those given the infant formula. The authors attributed this difference to the contribution made by 22:6ω3 in the breast-milk. The postnatal fall in 22:6ω3 in plasma phospholipids can only be prevented by dietary 22:6ω3 and not by dietary 18:3ω3; an amount equivalent to that in breast-milk was effective when dispersed in the feed (Liu *et al.* 1987).

EFA AND CANCER

Several workers have suggested that increasing the intake of polyunsaturated fatty acids may increase risk of cancer by increasing exposure to lipid peroxides. Fatty acid peroxides are only weakly mutagenic (MacGregor *et al.* 1985), but carbonyl compounds resulting from the oxidation of fat such as malonaldehyde are both mutagenic and carcinogenic (Shamberger *et al.* 1974). However, Se deficiency which leads to the accumulation of fatty

acid peroxides inhibits tumour growth (Reddy & Tanaka, 1986) and additional vitamin E in the diet has no effect.

Tumour growth may be promoted by dietary 18:2ω6. Ip (1987) has shown that the yield of chemically induced mammary tumours increases in a dose-dependent manner with up to 4% of the energy intake as 18:2ω6. Further increases in 18:2ω6 intake did not further promote tumour growth. Treatment of rats with indomethacin decreased tumour growth in animals fed on 4 or 12% of energy intake as 18:2ω6 compared with 0·5% of energy intake as 18:2ω6. These findings would imply that the prostaglandins promote tumour growth. High intakes of 20:5ω3 and 22:6ω3 inhibit tumour growth in several animal models (Karmali, 1987).

EFA AND CARDIOVASCULAR DISEASE

Sinclair (1956) first proposed that atherosclerosis was related to an inadequate intake of EFA or an inappropriate value for essential:saturated fatty acids in the diet. Epidemiological studies show a relationship between the proportion of 18:2ω6 in adipose tissue and risk of coronary heart disease (CHD) (Wood *et al.* 1987). Similar observations have been made between low levels of 20:5ω3 in platelets and CHD. The predictive power of these measurements still held when corrected for other known risk factors. CHD is rare among populations that have high intakes of 20:5ω3 and 22:5ω3 (Dyerberg & Bjerregaard, 1987; Hirai *et al.* 1980). Kromhout *et al.* (1985) in a prospective study showed that fish consumption was associated with a decreased risk of CHD. However, the amount of 20:5ω3 and 22:6ω3 provided by the fish was very small.

EFA AND PLASMA LIPOPROTEIN CONCENTRATION

Fatty liver is a common finding in EFAD. However, in part this can be attributed to the high sucrose content of the diet, which leads to increased hepatic triglyceride synthesis. It has been argued that EFA are required for the normal transport of lipids. Plasma lecithin acyl transferase (*EC* 2.3.1.43), an enzyme involved in the formation of cholesterol esters shows high specificity for 18:2ω6 (Myant, 1981). Patients with EFAD show abnormal liver function which is corrected by intravenous fat emulsions but not by topically-applied safflower oil (Miller *et al.* 1987).

Many studies have shown that the substitution of 18:2ω6-rich oils for saturated fats in the diet leads to a reduction in plasma cholesterol concentrations. In part this effect can be attributed to a reduction in saturated fatty acid intakes because similar reduction in low-density-lipoprotein (LDL)-cholesterol can be obtained with oils rich in oleic acid (Mattson & Grundy, 1985). Cortese *et al.* (1983) argue that high intakes of 18:2ω6 decrease the synthetic rate of LDL. Others have suggested that 18:2ω6 leads to a slight increase in the fractional catabolic rate of LDL (Illingworth *et al.* 1981).

Fish oils rich in eicosapentaenoic acid (20:5ω3) and docosahexaenoic acid (22:6ω3) lower plasma triglycerides and cholesterol but mainly in the very-low-density-lipoprotein (VLDL) fraction (Goodnight *et al.* 1982). Linseed oil, which is rich in linolenic acid (18:3ω3), does not have the same effect at comparable dosages (Sanders & Roshanai, 1983). The consumption of fish-oil supplements (20 g/d) slightly increases high-density-lipoprotein-cholesterol concentrations and markedly lowers plasma triglyceride concentrations in both normal and hypertriglyceridaemic subjects by reducing the synthesis of both VLDL-triglyceride and -apoprotein B (Sanders, 1987).

Very-high intakes of fish oil (90–120 g/d) do lower the concentration of both LDL-cholesterol and LDL-apoprotein B by decreasing the rate of LDL synthesis (Illingworth *et al.* 1984). Such high intakes also prevent the rise in plasma cholesterol obtained with

dietary cholesterol (Nestel, 1986). However, the reduction in LDL pool size is smaller than would be predicted from the decrease in synthesis. Moreover, despite the reduced LDL pool size there was no increase in the fractional catabolic rate of LDL. This might imply down regulation of the LDL receptors by fish oils. Indeed Wong & Nestel (1987) have reported decreased binding of LDL to Hep G2 cells treated with 20:5ω3. With lower intake of fish oils (15 g/d) or oily fish providing 3–5 g ω3 fatty acids there is a tendency for LDL-cholesterol and LDL-apoprotein B concentration to rise (Sullivan *et al.* 1986). In patients with type V hyperlipoproteinaemia (Phillipson *et al.* 1985), LDL-cholesterol levels rise even at high intakes (a phenomenon seen with most forms of triglyceride-lowering therapy). A likely explanation is that a moderate intake of fish oil decreases hepatic triglyceride synthesis so that smaller than normal VLDL particles are secreted (Sullivan *et al.* 1986). These small particles are known to be more readily converted to LDL than the larger triglyceride-rich ones. Moderate intakes of fish-oil concentrates are not useful for the treatment of hypercholesterolaemia. However, they offer a safe and effective means of treatment of hypertriglyceridaemia resulting from excessive VLDL synthesis, especially in patients with type III and type V hypoliproteinaemias.

EFA AND EXPERIMENTAL ATHEROSCLEROSIS

The severity of experimental atherosclerosis is influenced by the type rather than the quantity of fat in the diet (Mahley, 1982). When vegetable oils such as sunflower-seed oil are substituted for butterfat or lard, this is accompanied by less atherosclerosis. However, results in animals have been notoriously variable.

Polyunsaturated fatty acids from fish oils inhibit atherogenesis in pigs, dogs and monkeys even in the presence of hypercholesterolaemia. It seems that fish oil offers protection against atherosclerosis by a mechanism that overrides the influence of plasma lipoproteins. Protection may be afforded by changes in eicosanoid metabolism that favourably influence macrophage function (Leaf & Weber, 1988). Studies to evaluate whether fish oil prevents restenosis following coronary angioplasty in man are in progress.

TRANS-FATTY ACIDS

DIETARY SOURCES OF *TRANS*-FATTY ACIDS

A wide variety of polyunsaturated fatty acids are found in plants, containing conjugated, acetylinic and *trans*-double bond structures as well as cyclopropenoid and substituted fatty acids, but these are not normal constituents of human diets. *Trans*-isomeric unsaturated fatty acids are formed during the industrial and biological hydrogenation of unsaturated fats and are found in ruminant fats and margarines and shortenings. C_{18} *trans*-monounsaturated fatty acids are the major *trans*-isomers found in partially hydrogenated vegetable oil and ruminant fats, *trans*-polyunsaturated fatty acids are only found in trace amounts. Hardened marine oils contain a mixture of geometrical and positional isomers of C_{20-22} monounsaturated fatty acids and di- and tri-unsaturated fatty acids (British Nutrition Foundation, 1987).

The average intake of *trans*-fatty acids in the United Kingdom is estimated to be 7 g/d, ranging from 5 to 27 g/d, mainly in the form of monounsaturated fatty acids (British Nutrition Foundation, 1987). The mean intake of *trans*-fatty acids in the United States is similar except that unlike the United Kingdom, they are derived entirely from vegetable oil (Hunter & Applewhite, 1986).

METABOLISM OF *TRANS*-FATTY ACIDS

Generally *trans*-fatty acids are incorporated to a limited extent into phospholipids and cholesterol esters and remain mainly in the triglyceride fraction. Their metabolism is similar to that of saturated and *cis*-monounsaturated fatty acids. *Trans*-monounsaturated fatty acids have a similar conformation to saturated fatty acids owing to the geometry of the *trans*-double bond. *Trans*-fatty acids are utilized for energy production similar to their corresponding *cis*-isomers. Although *trans*-isomers appear to be metabolized at a slower rate by mitochondrial β-oxidation than their corresponding *cis*-isomers, their overall rates of oxidation are similar. *Trans*-isomers are also oxidized by the peroxisomal oxidation system. In common with C_{20-22} *cis*-monounsaturated fatty acids, the corresponding *trans*-isomers are initially poorly oxidized until the peroxisomal oxidation system becomes active (Christophersen *et al.* 1982). *Trans*-fatty acids are usually poor substrates for desaturation, but at very-high intakes act as inhibitors. *Trans,trans*-linoleic acid can partially inhibit the conversion of linoleic to 20:4ω6 (Beare-Rogers, 1983). This might account for the relatively-higher value for linoleic:20:4ω6 in tissues from animals fed on *trans*-isomers with adequate amounts of 18:2ω6. Alternatively, it may reflect an increased rate of peroxisomal oxidation of 20:4ω6 induced by diet (Dommes *et al.* 1981).

Holman (1985) and his colleagues have shown that *trans*-isomers of 20:4ω6 can be formed and have argued that these might be converted into prostaglandins. Many of the isomers lack the structural requirements for prostaglandin synthesis. It has yet to be shown whether pharmacologically active metabolites are produced under normal physiological conditions.

The feeding of *trans,trans*-linoleic acid to rats brings about a modest reduction in the proportion of 20:4ω6 in platelets. The reduction in the proportion of 20:4ω6 is modest and considerably less than can be brought about by feeding eicosapentaenoic acid (20:5ω3). *Trans-trans*-linoleic acid slightly reduced serum thromboxane B_2 concentrations but serum concentrations of 6-keto-prostaglandin $F_{1-\alpha}$, which is the stable metabolite of prostacyclin I_2, were unaffected (Bruckner *et al.* 1983). The fall in serum thromboxane production is consistent with a reduction in arachidonic concentrations in platelets. However, these changes were not physiologically significant because bleeding times were not altered. Recent studies indicate that it is necessary to decrease the proportion of 20:4ω6 in the phosphatidyl inositol fraction before changes in cell function mediated by prostaglandins can occur. The proportion of 20:4ω6 in phosphatidyl inositol appears to be conserved when the proportion of 20:4ω6 falls in other lipid fractions (Hornstra, 1982). Zevenbergen (1987) reported that thromboxane A_2 and prostacyclin PGI_2 formation by platelets or aorta was not altered in rats fed on partially hydrogenated soya-bean oil when the animals were supplied with an amount of 18:2ω6 sufficient to prevent classical EFA deficiency.

TOXICOLOGICAL STUDIES OF *TRANS*-ISOMERS

Few studies have examined the influence of purified *trans*-fatty acids and knowledge about their effects has been derived from observations with diets containing hydrogenated fats. The interpretation of such studies necessitates allowing for other variables such as the intake of saturated fatty acids and EFA. Generally *trans*-fatty acids do not possess EFA activity and like *cis*-monounsaturated fatty acids can exacerbate EFAD in the rat (Aaes-Jorgensen, 1958), but no effects are noted in animals fed on more than 1% of the energy as 18:2ω6. The safety of industrially hydrogenated vegetable oils has been comprehensively reviewed by Federation of American Societies for Experimental Biology (1985) and Zevenbergen (1987) and that of industrially-hydrogenated fish oils by Barlow & Duthie (1984). Long-term feeding studies have never shown adverse effects of *trans*-fatty acids on

longevity, reproductive performance, growth or revealed any consistent abnormalities on histopathological examination of many organs. Nor have any mutagenic and teratogenic effects been noted.

Partially hardened marine oils may lead to an acute myocardial lipidosis in rats and in primates, the severity depending on the concentration of C_{20-22} monounsaturated fatty acids (both *cis* and *trans*) in the diet (Svaar, 1982). Lipidosis is usually only seen when C_{20-22} monoenes account for more than 5% of the energy intake. Long-term feeding studies indicate that adaptation to high intakes of C_{20-22} monounsaturated fatty acids does occur in rodents but that myocardial lipidosis persists in primates (Schiefer *et al.* 1982). However, the amounts provided by British diets are considerably less than 5% of the energy intake.

TRANS-FATTY ACIDS AND CANCER

Associations between the incidence of cancer of the colon, breast and prostate with the use of industrial hydrogenated vegetable fats in various areas inside the United States have been reported (Enig *et al.* 1978). However, there is good evidence that *trans*-fatty acids are not carcinogens and studies using different tumour models in mice and rats have shown no effect of *trans*-fatty acids on tumour promotion (Federation of American Societies for Experimental Biology, 1985; Hunter *et al.* 1985).

TRANS-FATTY ACIDS AND CARDIOVASCULAR DISEASE

Kummerow (1986) and Booyen *et al.* (1988) have argued that *trans*-fatty acids may be involved in the causation of CHD. Thomas *et al.* (1981) claimed that the level of *trans*-fatty acids in adipose tissue was associated with the incidence of CHD in various regions of the United Kingdom. The work of Thomas *et al.* (1981) can be criticised on methodological grounds and because no allowance was made for other risk factors or for the differences in prosperity between the varying regions. Moreover, case-controlled studies have failed to show that patients with CHD have higher levels of *trans*-fatty acids in their adipose tissue than those who are free from the disease (Thomas *et al.* 1987).

A number of studies have evaluated the influence of *trans*-fatty acids on plasma cholesterol concentrations in man (Anderson *et al.* 1961; Mattson *et al.* 1975; Vergroesen & Gottenbos, 1975; Laine *et al.* 1982). The majority of these studies have used industrially hardened vegetable oils, only one study has employed partially hydrogenated marine oils (de Iongh *et al.* 1965). *Trans*-polyunsaturated fatty acids do not lower plasma cholesterol concentration as do all *cis*-polyunsaturated fatty acids. Elaidic acid 18:1 Δ9-*trans* has an equivalent effect on plasma cholesterol to oleic acid 18:1 Δ9-*cis*. *Trans*-fatty acids provided as partially hydrogenated vegetable oils consumed in amounts ranging from 3 to 14% energy have little influence on plasma cholesterol concentrations in man. The apparent lack of influence of *trans*-fatty acids on plasma cholesterol concentrations may well be related to their chain length. For it is well known that only C_{12-16} saturated fatty acids have a hypercholesterolaemic effect.

Early studies suggested that industrially hydrogenated fats were more atherogenic than beef tallow. However, the experimental diets used were deficient in 18:2ω6. Later studies, which used diets that supplied an adequate intake of 18:2ω6, showed there was no increase in the incidence of atherosclerosis (Gottenbos, 1983). Kritchevsky (1982) concludes that *trans*-fatty acids, resulting from the industrial hydrogenation of vegetable oils when fed in amounts up to 14% of the energy intake are not atherogenic. Partially hydrogenated fish oils do not appear to be atherogenic in primates (Mueller *et al.* 1982; Schiefer, 1982).

CONCLUSIONS

Although small amounts of EFA in the diet are necessary, their importance in human diets is probably related to the effects exerted by higher intakes and by the relative balance between the ω6 and ω3 series. It is not possible to delineate an optimum intake or an ideal balance of EFA without first defining the criteria by which they are to be measured. These criteria may well differ for different groups. For example, a high capacity to produce prostaglandins may be undesirable in patients with CHD, but would be desirable in a woman in labour. Further work is needed to catalogue the biological effects of dietary polyunsaturates and their interactions with other dietary constituents.

The plethora of studies carried out to demonstrate harmful effects from the consumption of *trans*-fatty acid have failed to provide any convincing evidence that the current consumption of *trans*-fatty acids in biologically or industrially hydrogenated fats poses a risk to health. In view of the lack of evidence supporting an association between the consumption of *trans*-isomers and atherosclerosis, it might seem perplexing why the Department of Health and Social Security (1984) recommended including *trans*-fatty acids with saturated fatty acids. However, the latter report (Department of Health and Social Security, 1984) preceded the major Federation of American Societies for Experimental Biology (1985) review on the subject and it was believed at that time that *trans*-unsaturated fatty acids were metabolized in a similar manner to saturated fatty acids. However, unlike saturated fatty acids, *trans*-monounsaturated fatty acids are not hypercholesterolaemic in man or prothrombotic in animals (Hornstra, 1982). Considerably less is known about the effects of partially hydrogenated fish oil than of hydrogenated vegetable oils and more research in this area is required.

REFERENCES

Aaes-Jorgensen, E. (1958). Essential fatty acid deficiency. III. Effects of conjugated isomers of dienoic and trienoic fatty acids in rats. *Journal of Nutrition* **66**, 465–483.

Abeywardena, M. Y., McLennan, P. L. & Charnock, J. S. (1987*a*). Diet and cardiac arrhythmia: involvement of eicosanoids. *Proceedings of the AOCS Short Course of Polyunsaturated Fatty Acids and Eicosanoids*, pp. 62–64 [W. E. M. Lands, editor]. Champaign, Illinois: American Oil Chemists' Society.

Abeywardena, M. Y., McLennan, P. L. & Charnock, J. S. (1987*b*). (Long-term saturated fat supplementation in the rat causes an increase in PGI2/TXB2 ratio of platelet and vessel wall compared to *n*-3 and *n*-6 fatty acids. *Atherosclerosis* **66**, 181–189.

Anderson, J. T., Grande, F. & Keys, A. (1961). Hydrogenated fats in the diet and lipids in the serum of man. *Journal of Nutrition* **75**, 388–394.

Antiplatelet Trialists' Collaboration (1988). Secondary prevention of vascular disease by prolonged antiplatelet treatment. *British Medical Journal* **296**, 320–331.

Aveldano, M. (1987). A novel group of very long chain polyenoic fatty acids in dipolyunsaturated phosphatidylcholine from vertebrate retina. *Journal of Biological Chemistry* **262**, 1172–1179.

Bang, H. O. & Dyerberg, J. (1980). Lipid metabolism and ischemic heart disease in Greenland Eskimos. *Advances in Nutrition Research* **3**, 1–40.

Barlow, S. M. & Duthie, I. F. (1984). An evaluation of the safety of partially hydrogenated marine oils (PHMO) in the human diet. *Nutrition Abstracts and Reviews A* **54**, 17–30.

Beare-Rogers, J. L. (1983). *Trans*- and positional isomers of common fatty acids. *Advances in Nutrition Research* **5**, 171–200.

Bernsohn, J. & Spitz, F. J. (1974). Linoleic and linolenic dependency of some membrane-bound enzymes after lipid deprivation in rats. *Biochemical and Biophysical Research Communications* **57**, 293–298.

Bittiner, S. B., Tucker, W. F. G., Cartwright, I. & Bleehen, S. S. (1988). A double-blind, randomised, placebo-controlled trial of fish oil in psoriasis *Lancet* **i**, 378–380.

Bjerve, K. S., Fischer, S. & Alme, K. (1987*a*). Alpha-linolenic acid deficiency in man. Effect of ethyl linolenate on plasma and erythrocyte fatty acid composition and biosynthesis of prostanoids. *American Journal of Clinical Nutrition* **46**, 570–576.

Bjerve, K. S., Mostad, I. L. & Thoresen, L. (1987*b*). Alpha-linolenic acid deficiency in patients on long-term gastric-tube feeding: estimation of linolenic acid and long-chain unsaturated *n*-3 fatty acid requirement in man. *American Journal of Clinical Nutrition* **45**, 66–77.

Booyen, J., Louwrens, C. C. & Katzeff, I. E. (1988). The role of unnatural *trans* and *cis* unsaturated fatty acids in the epidemiology of coronary artery disease. *Medical Hypotheses* **85**C, 21–23.

British Nutrition Foundation (1987). *Trans* fatty acids. *Report of the British Nutrition Foundation's Task Force.* London: British Nutrition Foundation.

Brown, W. R., Hansen, A. E., Burr, G. O. & McQuarrie, I. (1938). Effects of prolonged use of extremely low-fat diet on an adult human subject. *Journal of Nutrition* **16**, 511–524.

Bruckner, G., Trimbo, S., Goswami, S. & Kinsella, J. E. (1983). Dietary trilinoelaidate: effects on hematological parameters, serum eicosanoids and tissue fatty acid composition in rats. *Journal of Nutrition* **113**, 704–713.

Caster, W. O. & Ahn, P. (1963). Electrocardiographic notching in rats deficient in EFA. *Science* **139**, 1213.

Chapkin, R. S., Ziboh, V. A. & McCullogh, J. L. (1987). Dietary influences of evening primrose and fish oil on the skin of essential fatty acid-deficient guinea pigs. *Journal of Nutrition* **117**, 1360–1370.

Christophersen, B. O., Norseth, J., Thomassen, M. S., Cristiansen, E. N., Norum, K. R., Osmundsen, H. & Bremer, J. (1982). Metabolism and metabolic effects of $C_{22:1}$ fatty acids with special reference to cardiac lipidosis. In *Nutritional Evaluation of Long-chain Fatty Acids in Fish Oil*, pp. 89–139 [S. M. Barlow and M. E. Stansby, editors]. New York: Academic Press.

Cortese, C., Levy, Y., Janus, E. D., Turner, P. R., Rao, S. N., Miller, N. E. & Lewis, B. (1983). Modes of action of lipid-lowering diets in man: studies of apolipoprotein B kinetics in relation to fat consumption and dietary fatty acid composition. *European Journal of Clinical In* *n* **13**, 79–85.

Crawford, M. A. & Sinclair, A. J. (1972). Nutritional influ the evolution of mammalian brain. In *Lipids, Malnutrition and the Developing Brain*, pp. 267–292 obing and J. Knight, editors]. Amsterdam: Elsevier.

Culp, B. R., Lands, W. E. M., Lucches, B. R., Pitt, R. & Romson, J. (1980). The effect of dietary supplementation of fish oil on experimental myocardial infarction. *Prostaglandins* **20**, 1021–1031.

de Iongh, H., Beerthuis, R. K., den Hartog, C., Dalderup, L.M. & van der Spek, P. A. F. (1965). The influence of some dietary fats on serum lipids in man. *Bibliotheca Nutritio et Dieta* **7**, Suppl., 137–152.

Department of Health and Social Security (1984). *Diet and Cardiovascular Disease. Committee on Medical Aspects of Food Policy. Report of the Panel on Diet in Relation to Cardiovascular Disease.* London: HMSO.

Dommes, V., Baumgart, C. & Kunau, W. H. (1981). Degradation of unsaturated fatty acids in peroxisomes. *Journal of Biological Chemistry* **256**, 8259–8262.

Dratz, E. A. & Deese, A. J. (1986). The role of docosahexaenoic acid (22:6ω3) in biological membranes: examples from photoreceptors and model membrane bilayers. In *Health Effects of Polyunsaturated Fatty Acids in Seafoods*, pp. 319–351 [A. P. Simopoulos, R. R. Kifer and R. E. Martin, editors]. New York: Academic Press.

Dyerberg, J. & Bjerregaard, P. (1987). Mortality from ischaemic heart disease and cerebrovascular disease in Greenland. In *Proceedings of the AOCS Short Course on Polyunsaturated Fatty Acids and Eicosanoids*, pp. 2–8 [W. E. M. Lands, editor]. Champaign Illinois: American Oil Chemists' Society.

Elliot, W. J., Morrison, A. R., Sprecher, H. W. & Needleman, P. (1985*a*). The metabolic transformations of columbinic acid and the effect of topical application of the major metabolites on rat skin. *Journal of Biological Chemistry* **260**, 987–92.

Elliot, W. J., Sprecher, H. & Needleman, P. (1985*b*). Physiologic effects of columbinic acid and its metabolites on rat skin. *Biochimica et Biophysica Acta* **835**, 158–160.

Enig, M. G., Munn, R. J. & Keeney, M. (1978). Dietary fat and cancer trends: a critique. *Federation Proceedings* **37**, 2215–2220.

Federation of American Societies for Experimental Biology (1985). *Health Aspects of Dietary* Trans *Fatty Acids.* Bethesda, Maryland: Federation of American Societies for Experimental Biology.

Fiennes, R. N., Sinclair, A. J. & Crawford, M. A. (1973). Essential fatty acid studies in primates: linolenic acid requirements of capuchins. *Journal of Medical Primatology* **2**, 1253–1254.

Fischer, F., Vischer, A., Preac-Mursic, V. & Weber, P. C. (1987). Dietary docosahexaenoic acid is retroconverted in man to eicosapentaenoic acid, which can quickly be transformed into prostaglandin I3. *Prostaglandins* **34**, 367–375.

Goodnight, S. H., Harris, W. S., Connor, W. E. & Illingworth, D. R. (1982). Polyunsaturated fatty acids, hyperlipidemia, and thrombosis, *Arteriosclerosis* **2**, 87–113.

Gottenbos, J. J. (1983). Biological effects of *trans* fatty acids. In *Dietary Fats and Health, American Oil Chemists' Society Monograph* no. 10. pp. 375–390. [E. G. Perkins & W. J. Visek, editors]. Champaign, Illinois: American Oil Chemists' Society.

Hafiez, A. A. (1974). Prostaglandin E2 prevents impairment of fertility in rats fed a diet deficient in essential fatty acids. *Journal of Reproduction and Fertility* **38**, 273–286.

Hagve, T. & Christophersen, B. O. (1987). Sex-related differences in desaturation and chain elongation of essential fatty acids studied in isolated rat hepatocytes. *Biochimica et Biophysica Acta* **920**, 149–154.

Hansen, H. S. & Jensen, B. (1985). Essential function of linoleic acid esterified in acylglucosylceramide and acylceramide in maintaining the epidermal water permeability barrier. Evidence from feeding studies with oleate, linoleate, arachidonate, columbinate and α-linolenate. *Biochimica et Biophysica Acta* **834**, 357–363.

Hansen, H. S., Jensen, B. & von Wettstein-Knowles, P. (1986). Apparent in vivo retroconversion of dietary arachidonic to linoleic acid in essential fatty acid-deficient rat. *Biochimica et Biophysica Acta* **878**, 284–287.

Harris, W. S., Connor, W. E. & Lindsey, S. (1984). Will dietary omega-3 fatty acids change the composition of human milk? *American Journal of Clinical Nutrition* **40**, 780–785.
Hartog, J. M., Verdouw, P. D., Klompe, M. & Lamers, J. M. J. (1987). Dietary mackerel oil in pigs: effect on plasma lipids, cardiac sarcolemmal phospholipids and cardiovascular parameters. *Journal of Nutrition* **117**, 1371–1378.
Hirai, A., Hamazaki, T., Terano, T., Nishikawa, T., Tamura, Y., Kumagai, A. & Sajiki, J. (1980). Eicosapentaenoic acid and platelet function in Japanese. *Lancet* **ii**, 1132–1133.
Hock, C. E., Holaham, M. A. & Reibel, D. K. (1987). Effect of dietary fish oil on myocardial phospholipids and myocardial ischemic damage. *American Journal of Physiology* **252**, H554–H560.
Hohl, C. M. & Rosen, P. (1987). The role of arachidonic acid in rat heart cell metabolism. *Biochimica et Biophysica Acta* **921**, 356–363.
Hollander, D. & Tarnawski, A. (1986). Dietary essential fatty acids and the decline in peptic ulcer disease – a hypothesis. *Gut* **27**, 239–242.
Holman, R. T. (1985). Influence of hydrogenated fats on the metabolism of polyunsaturated fatty acids. In *The Role of Fats in Human Nutrition*, pp. 48–61 [F. B. Padley and J. Podmore, editors]. Chichester: Ellis Horwood.
Holman, R. T., Johnson, S. B. & Hatch, T. F. (1982). A case of human linolenic acid deficiency involving neurological abnormalities. *American Journal of Clinical Nutrition* **35**, 617–623.
Holman, R. T., Smythe, L. & Johnson, S. (1979). Effect of sex and age on fatty acid composition of human serum lipids. *American Journal of Clinical Nutrition* **32**, 2390–2399.
Hornstra, G. (1982). Dietary fats, prostanoids and arterial thrombosis. *Developments in Hematology and Immunology*, vol. 4. London: Martinus Nijhoff.
Houtsmuller, U. M. T. (1973). Differentiation in the biological activity of polyunsaturated fatty acids. In *Dietary Lipids and Postnatal Development*, pp. 145–155 [C. Galli, G. Jacini and A. Pecile, editors]. New York: Raven Press.
Houtsmuller, U. M. T. (1981). Columbinic acid, a new type of essential fatty acid. *Progress in Lipid Research* **20**, 889–896.
Houtsmuller, U. M. T. & Van der Beek, A. (1981). Effects of topical application of fatty acids. *Progress in Lipid Research* **20**, 219–224.
Hulan, H. W., Kramer, J. K. G. & Corner, A. H. (1977). The effect of dietary fatty acid balance on myocardial lesions in male rats. *Lipids* **12**, 951–956.
Hunter, J. E. & Applewhite, T. H. (1986). Isomeric fatty acids in the US diet: levels and health perspectives. *American Journal of Clinical Nutrition* **44**, 707–717.
Hunter, J. E., Ip, C. & Hollenbach, E. J. (1985). Isomeric fatty acids and tumorogenesis: a commentary on recent work. *Nutrition and Cancer* **7**, 199–209.
Illingworth, D. R., Harris, W. S. & Connor, W. E. (1984). Inhibition of low density lipoprotein synthesis by dietary omega-3 fatty acids in humans. *Arteriosclerosis* **4**, 270–275.
Illingworth, D. R., Sundberg, E. E., Becker, N., Connor, W. E. & Alaupovic, P. (1981). Influence of saturated, monounsaturated and ω6-PUFA on LDL metabolism in man. *Arteriosclerosis* **1**, 380.
Ip, C. (1987). Fat and essential fatty acid in mammary carcinogenesis. *American Journal of Clinical Nutrition* **45**, 218–224.
Jenkins, K. J. & Ewan, L. M. (1967). Promotion of myopathy by polyunsaturated fatty acids in cod liver oil. *Canadian Journal of Biochemistry* **45**, 1873–1880.
Karmali, R. A. (1987). Omega-3 fatty acids and cancer: a review. In *Proceedings of the AOCS Short Course on Polyunsaturated Fatty Acids and Eicosanoids*, pp. 222–232 [W. E. M. Lands, Editor]. Champaign, Illinois: American Oil Chemists' Society.
Keys, A. (1970). Coronary heart disease in seven countries. *Circulation* **41**, 1–211.
Knapp, H. R., Reilly, A. G., Allesandrini, P. & Fitzgerald, G. A. (1985). In vivo indexes of platelet and vascular function during fish oil administration in patients with atherosclerosis. *New England Journal of Medicine* **314**, 937–942.
Kramer, J. K. G., Farnworth, E. R. & Thompson, B. K. (1985). Cardiac lipid changes in rats fed oils enriched in saturates and their apparent relationship to focal heart lesions. *Lipids* **20**, 635–644.
Kremer, J. M., Jubix, W., Michalek, A., Rynese, R. I., Bartholmew, L. E., Bigaouette, J., Timchlak, M., Beeler, D. & Leninger, L. (1987). Fish-oil fatty acid supplementation in active rheumatoid arthritis. A double-blind, controlled, crossover study. *Annals of Internal Medicine* **106**, 497–503.
Kritchevsky, D. (1982). *Trans* fatty acid effects in experimental atherosclerosis. *Federation Proceedings* **41**, 2813–2817.
Kromhout, D., Bosschieter, E. B. & Coulander, C. L. (1985). The inverse relation between fish consumption and 20-year mortality from coronary disease. *New England Journal of Medicine* **312**, 1205–1209.
Kummerow, F. A. (1986). Dietary effects of *trans* fatty acids. *Journal of Environmental Pathology Toxicology and Oncology* **6**, 123–149.
Laine, D. C., Snodgrass, C. M., Dawson, E. A., Ener, M. A., Kuba, K. & Frantz, I. D. (1982). Lightly hydrogenated soy oil versus other vegetable oils as a lipid-lowering dietary constituent. *American Journal of Clinical Nutrition* **35**, 683–690.

Lamptey, M. S. & Walker, B. L. (1976). A possible essential role for dietary linolenic acid in the development of the young rat. *Journal of Nutrition* **106**, 86–93.

Leaf, A. & Weber, P. C. (1988). Cardiovascular effects of *n*-3 fatty acids. *New England Journal of Medicine* **318**, 549–557.

Leat, W. M. F., Curtis, R., Millchamp, N. & Cox, R. W. (1986). Retinal function in rats and guinea-pigs reared on diets low in essential fatty acids and supplemented with linoleic or linolenic acids. *Annals of Nutrition and Metabolism* **30**, 166–174.

Leat, W. M. F. & Northrop, C. A. (1981). Effect of dietary linoleic and linolenic acids on gestation and parturition in the rat. *Quarterly Journal of Experimental Physiology* **66**, 99–103.

Leat, W. M. F., Northrop, C. A., Harrison, F. A. & Cox, R. W. (1983). Effect of dietary linoleic and linolenic acids on testicular development in the rat. *Quarterly Journal of Experimental Physiology* **68**, 221–231.

Leaver, H. A., Lytton, F. D., Dyson, F. D., Watson, M. L. & Mellor, D. J. (1986). The effect of dietary omega-3 and omega-6 polyunsaturated fatty acids on gestation, parturition and prostaglandin E2 in intrauterine tissues and the kidney. *Progress in Lipid Research* **25**, 143–146.

Lefkowith, J. B. & Schreiner, G. (1987). Essential fatty acid deficiency depletes rat glomeruli of resident macrophages and inhibits angiotensin II-induced eicosanoid synthesis. *Journal of Clinical Investigation* **80**, 947–956.

Lepran, I., Nemecz, G., Koltai, M. & Szekers, S. (1981). Effect of linoleic acid-rich diets on the acute phase of coronary occlusion in conscious rats: influence of indomethacin and aspirin. *Journal of Cardiovascular Pharmacology* **3**, 847–853.

Lewis, G. P. (1983). Immunoregulatory activity of metabolities of arachidonic acid and their role in inflammation. *British Medical Bulletin* **39**, 243–248.

Liu, C. F., Carlson, S. E., Rhodes, P. G., Rao, V. S. & Meydrech, E. F. (1987). Increase in plasma phospholipid docosahexaenoic and eicosapentaenoic acids as a reflection of their intake and mode of administration. *Pediatric Research* **22**, 292–296.

McCarren, T., Hitzemann, R., Smith, R., Kloss, R., Allen, C. & Glueck, C. J. (1985). Amelioration of severe migraine by fish oil (omega-3) fatty acids. *American Journal of Clinical Nutrition* **41**, 874.

MacDonald, M. L., Rogers, Q. R. & Morris, J. G. (1983). Role of linoleate as an essential fatty acid for the cat, independent of arachidonate synthesis. *Journal of Nutrition* **113**, 1422–1433.

MacDonald, M. L., Rogers, Q. R., Morris, J. G. & Cupps, P. T. (1984). Effects of linoleate and arachidonate deficiencies on reproduction and spermatogenesis in the cat. *Journal of Nutrition* **114**, 719–726.

MacGregor, J. T., Wilson, R. E., Neff, W. E. & Frankel, E. N. (1985). Mutagenicity tests on lipid oxidation products in *Salmonella typhimurium*: monohydroperoxides and secondary oxidation products of methyl linoleate and methyl linolenate. *Food Chemistry and Toxicology* **23**, 1041–1047.

Mahley, R. W. (1982). Atherogenic hyperlipoproteinemia. The cellular and molecular biology of plasma lipoproteins altered by dietary fat and cholesterol. *Medical Clinics of North America* **66**, 375–402.

Manku, M S., Horrobin, D. F., Morse, N., Kyte, V. & Jenkins, K. (1982). Reduced levels of prostaglandin precursors in the blood of atopic patients: defective delta-6-desaturase function as a biochemical basis for atopy. *Prostaglandins, Leukotrienes and Medicine* **9**, 615–628.

Marsden, J. R. (1987). Effect of dietary fish oil on hyperlipidaemia due to isoretinoin and etretinate. *Human Toxicology* **6**, 219–22.

Mattson, F. H. & Grundy, S. M. (1985). Comparison of effects of dietary saturated, monounsaturated, and polyunsaturated fatty acids on plasma lipids and lipoproteins in man. *Journal of Lipid Research* **26**, 194–202.

Mattson, F. H., Hollenbach, E. J. & Kligman, A. M. (1975). Effect of hydrogenated fat on the plasma cholesterol and triglyceride levels of man. *American Journal of Clinical Nutrition* **28**, 726–731.

Mertin, J. & Stackpoole, A. (1981). Anti-PGE antibodies inhibit in vivo development of cell-mediated immunity. *Nature* **294**, 456–458.

Miller, D. G., Williams, S. K., Palombo, J. D., Griffin, R. E., Bistrian, B. R. & Blackburn, G. L. (1987). Cutaneous application of safflower oil in preventing essential fatty acid deficiency in patients on home parenteral nutrition. *American Journal of Clinical Nutrition* **46**, 419–423.

Moncada, S. & Vane, J. R. (1979). Arachidonic acid metabolites and the interactions between platelets and blood vessel walls. *New England Journal of Medicine* **300**, 1142–1147.

Morson, L. A. & Clandinin, M. T. (1986). Diets varying in linoleic and linolenic acid content alter liver plasma membrane lipid composition and glucagon-stimulated adenylate cyclase activity. *Journal of Nutrition* **116**, 2355–2362.

Mueller, R. W., Schiefer, H. B., Laxdal, V. A. & Ackman. R. G. (1982). Aortic changes in Cynomolgus monkeys fed high-fat diets (long-term study). *Artery* **11**, 174–191.

Mullane, K. M., Read, N., Salmon, J. A. & Moncada, S. (1984). Role of leukocytes in acute myocardial infarction in anaesthetized dogs: relationship to myocardial salvage by anti-inflammatory drugs. *Journal of Pharmacology and Experimental Therapeutics* **228**, 510–522.

Myant, N. B. (1981). *The Biology of Cholesterol and Related Steroids*. London: W. Heinemann Medical Books.

Nestel, P. J. (1986). Fish oil attenuates the cholesterol-induced rise in lipoprotein cholesterol. *American Journal of Clinical Nutrition* **43**, 752–757.

Neuringer, M. & Connor, W. E. (1987). The importance of dietary *n*-3 fatty acids in the development of the retina and nervous system. In *Proceedings of the AOCS Short Course on Polyunsaturated Fatty Acids and Eicosanoids*, pp. 301–311 [W. E. M. Lands, editor]. Champaign, Illinois: American Oil Chemists' Society.

Norris, P. G., Jones, C. J. H. & Weston, M. J. (1986). Effect of dietary supplementation with fish oil on systolic blood pressure in essential hypertension. *British Medical Journal* **ii**, 104–105.

Nugteren, D. H. & Kivits, G. A. A. (1987). Conversion of linoleic and arachidonic acid by skin epidermal lipoxygenases. *Biochimica et Biophysica Acta* **921**, 135–141.

Nugteren, D. H. & Christ-Hazelhof, C. (1987). Naturally occurring conjugated octadecatrienoic acids are strong inhibitors of prostaglandin biosynthesis. *Prostaglandins* **33**, 403–417.

O'Dea, K. & Sinclair, A. J. (1982). Increased proportion of arachidonic acid in plasma liquids after two weeks on a diet of tropical seafood. *American Journal of Clinical Nutrition* **36**, 868–872.

Olsen, S. F., Hansen, H. S., Jensen, B., Sommer, S., Soerensen, T. I. A., Secher, N. J. & Zachariassen, P. (1987). Duration of pregnancy and intake of marine fat in two populations, one with low and one with high consumption of marine fat. In *Proceedings of the AOCS Short Course on Polyunsaturated Fatty Acids and Eicosanoids*, pp. 268–269 M. Lands, editor]. Champaign, Illinois: American Oil Chemists' Society.

Phillipson Rothrock, D. W., Connor, W. E., Harris, W. S. & Illingworth, D. R. (1985). Reduction of plasma poproteins, and apoproteins by dietary fish oils in patients with hypertriglyceridemia. *New England Journal of Medicine* **19**, 1210–1216.

Poyser, N. L. (1981). *Prostaglandins in Reproduction*. Chichester: John Wiley.

Press, M., Kikuchi, H., Shimoyama, T. & Thompson, G. R. (1974). Diagnosis and treatment of essential fatty acid deficiency in man. *British Medical Journal* **ii**, 247–250.

Pudelkewicz, C., Seufert, J. & Holman, R. T. (1968). Requirements of the female rat for linoleic and linolenic acids. *Journal of Nutrition* **94**, 138–146.

Putnam, J. C., Carlson, S. E., DeVoe, P. W. & Barness, L. A. (1982). The effects of variations in dietary fatty acids on the fatty acid composition of erythrocyte phosphatidylcholine and phosphatidylethanolamine in human infants. *American Journal of Clinical Nutrition* **36**, 106–114.

Reddy, B. S. & Tanaka, T. (1986). Interactions of selenium deficiency, vitamin E, polyunsaturated and saturated fat on azoxymethane-induced colon carcinogenesis in male F344 rats. *Journal of the National Cancer Institute* **76**, 1157–1162.

Ritter, J. M. & Taylor, G. W. (1988). Fish oil in asthma. *Thorax* **43**, 81–83.

Rivers, J. P. W. & Frankel, T. L. (1981). Essential fatty acid deficiency. *British Medical Bulletin* **37**, 59–64.

Robinson, D. R., Colvin, R. B., Hirai, A. & Steinberg, A. D. (1985). Dietary marine lipids modify autoimmune diseases. In *Health Effects of Polyunsaturated Fatty Acids in Seafoods*, pp. 247–259 [A. P. Simopoulos, R. R. Kifer and R. E. Martin, editors]. New York: Academic Press.

Roshanai, F. & Sanders, T. A. B. (1984). Assessment of fatty acid intakes in vegans and omnivores. *Human Nutrition: Applied Nutrition* **38A**, 345–354.

Sanders, T. A. B. (1983). Dietary fat and platelet function. *Clinical Science* **65**, 343–350.

Sanders, T. A. B. (1987). Fish and coronary artery disease. *British Heart Journal* **57**, 214–219.

Sanders, T. A. B., Ellis, F. R. & Dickerson, J. W. T. (1978). Studies of vegans: the fatty acid composition of plasma choline phosphoglycerides, erythrocytes, adipose tissue, and breast-milk and some indicators of susceptibility to ischemic heart disease in vegans and omnivore controls. *American Journal of Clinical Nutrition* **31**, 805–813.

Sanders, T. A. B. & Naismith, D. J. (1979). A comparison of the influence of breast-feeding and bottle-feeding on the fatty acid composition of erythrocytes. *British Journal of Nutrition* **41**, 619–623.

Sanders, T. A. B. & Roshanai, F. (1983). The influence of different types of ω3 polyunsaturated fatty acids on blood lipids and platelet function in healthy volunteers. *Clinical Science* **64**, 91–99.

Sanders, T. A. B. & Younger, K. M. (1981). The effects of dietary supplements of ω3 polyunsaturated fatty acids on the fatty acid composition of platelets and plasma choline phosphoglycerides. *British Journal of Nutrition* **45**, 613–616.

Schiefer, H. B. (1982). Studies in non-hominid primates. In *Nutritional Evaluation of Long-Chain Fatty Acids in Fish Oil*, pp. 215–243 [S. M. Barlow and M. E. Stansby, editors]. New York: Academic Press.

Schiefer, H. B., Hancock, D. S. & Loew, F. M. (1982). Long-term effects of partially hydrogenated herring oil on rat myocardium. *Drug–Nutrient Interactions* **1**, 89–102.

Seybeth, H. W., Oez, O, Kennedy, T., Sweetman, B. J., Danon, A., Frolich, J. C., Heimberg, M. & Oates, J. A. (1975). Increased arachidonate in lipids after administration to man: effects on prostaglandin biosynthesis. *Clinical Pharmacology and Therapeutics* **18**, 521–529.

Shamberger, R. J., Andreone, T. L. & Willis, C. E. (1974). Antioxidants and cancer. IV. Initiating activity of malonaldehyde as a carcinogen. *Journal of the National Cancer Institute* **53**, 1771–1773.

Sherertz, E. F. (1986). The skin in essential fatty acid deficiency. In *Nutrition and the Skin*, pp. 117–130 [D. A. Roe, editor]. New York: Alan R. Liss.

Sinclair, H. M. (1956). Deficiency of essential fatty acids and atherosclerosis, etcetera, *Lancet* **i**, 381–383.

Spector, A. A., Mathur, S. N., Kaduce, T. L. & Hyman, B. T. (1982). Lipid nutrition and metabolism of cultured mammalian cells. *Progress in Lipid Research* **19**, 155–186.

Sullivan, D. R., Sanders, T. A. B., Trayner, I. M. & Thompson, G. R. (1986). Paradoxical elevation of LDL

apoprotein B levels in hypertriglyceridaemic patients and normal subjects ingesting fish oil. *Atherosclerosis* **61**, 129–134.

Svaar, H. (1982). The long-term heart lesion phenomenon in animals and humans. In *Nutritional Evaluation of Long-Chain Fatty Acids in Fish Oil*, pp. 163–184 [S. M. Barlow and M. E. Stansby, editors]. New York: Academic Press.

Talesnik, J. (1986). Arachidonic acid induced coronary reactions and their inhibition by docosahexaenoic acid. *Canadian Journal of Physiology and Pharmacology* **64**, 77–84.

Ten Hoor, F., de Deckere, E. A. M., Haddeman, E., Hornstra, G. & Quadt, J. F. A. (1980). Dietary manipulation of prostaglandin and thromboxane synthesis in heart, aorta and blood platelets of the rat. *Advances in Prostaglandin and Thromboxane Research* **8**, 1771–1781.

Terano, T., Hirai, A., Hamazaki, T., Kobayashi, S., Fujita, T., Tamura, Y. & Kumagai, A. (1983). Effect of oral administration of highly purified eicosapentaenoic acid on platelet function, blood viscosity and red cell deformability in healthy human subjects. *Atherosclerosis* **46**, 321–331.

Thomas, L. H., Jones, P. R., Winter, J. A. & Smith, H. (1981). Hydrogenated oils and fats: presence of chemically modified fatty acids in human adipose tissue. *American Journal of Clinical Nutrition* **34**, 877–886.

Thomas, L. H., Olpin, S. O., Scott, R. G. & Wilkins, M. P. (1987). Coronary heart disease and the composition of adipose tissue taken at biopsy. *Human Nutrition: Food Science and Nutrition* **41F**, 167–172.

Thomson, M., Fulton, M., Elton, R. A., Brown, S., Wood, D. A. & Oliver, M. F. (1988). Alcohol consumption and nutrient intake in middle-aged Scottish men. *American Journal of Clinical Nutrition* **47**, 139–45.

Tinoco, J. (1982). Dietary requirements and functions of α-linolenic acid in animals. *Progress in Lipid Research* **21**, 1–45.

Vergroesen, A. J. & Gottenbos, J. J. (1975). The role of fats in human nutrition. An introduction. In *The Role of Fats in Human Nutrition*, [A. J. Vergroesen, editor]. New York: Academic Press.

Von Schacky, C. & Weber, P. C. (1985). Metabolism and effects on platelet function of the purified eicosapentaenoic and docosahexaenoic acids in humans. *Journal of Clinical Investigation* **76**, 2446–2450.

Wertz, P. W., Swarzendruber, D. C., Abraham, W., Madison, K. C. & Dowing, D. T. (1987). Essential fatty acids and epidermal integrity. *Archives of Dermatology* **123**, 1381–1384.

Wheeler, T. G., Benolken, R. M. & Anderson, R. E. (1975). Visual membranes: specificity of fatty acid precursors for the electrical response to illumination. *Science* **182**, 1312–1314.

Wong, S. & Nestel, P. J. (1987). Eicosapentaenoic acid inhibits the secretion of triacylglycerol and of apoprotein B and the binding of LDL in Hep G2 cells. *Atherosclerosis* **64**, 139–146.

Wood, D. A., Butler, S., MacIntyre, C., Riemersma, R. A., Thomson, M., Elton, R. A. & Oliver, M. F. (1987). Linoleic and eicosapentaenoic acids in adipose tissue and platelets and risk of coronary heart disease. *Lancet* **i**, 117–183.

Wright, S. & Burton, J. L. (1982). Oral evening primrose oil improves atopic eczema. *Lancet* **ii**, 1120–1122.

Yamamoto, N., Saitoh, M., Moriuchi, A., Nomura, M. & Okuyama, H. (1987). Effect of dietary α-linolenate linoleate balance on brain lipid compositions and learning ability of rats. *Journal of Lipid Research* **28**, 144 151.

Yamanaka, W. K., Clemans, G. W. & Hutchinson, M. L. (1980). Essential fatty acids deficiency in humans. *Progress in Lipid Research* **19**, 187–215.

Young, M. R., Ellis, N. K., Young, M. E. & Wepsic, H. T. (1987). Stimulation of hematopoiesis and bone marrow suppressor cells by the subcutaneous injection of linoleic acid. *Cellular Immunology* **107**, 238–248.

Zevenbergen, J. L. (1987). Biological effects of *trans* fatty acids. In *Fat Production and Consumption. Technologies and Nutritional Implications*, pp. 157–167 [C. Galli and E. Fedeli, editors]. New York: Plenum Press.

Printed in Great Britain