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Review Article

Animal studies of the functional consequences of suboptimal polyunsaturated fatty acid status during pregnancy, lactation and early post-natal life

J. Thomas Brenna

Division of Nutritional Sciences, Cornell University, Ithaca, New York, USA

Abstract

Scores of animal studies demonstrate that seed oils replete with linoleic acid and very low in linolenic acid fed as the exclusive source of fat through pregnancy and lactation result in visual, cognitive, and behavioural deficits in the offspring. Commodity peanut, sunflower, and safflower oils fed to mother rats, guinea pigs, rhesus monkeys, and baboons induce predictable changes in tissue polyunsaturated fatty acid composition that are abnormal in free-living land mammals as well as changes in neurotransmitter levels, catecholamines, and signalling compounds compared with animals with a supply of ω3 polyunsaturated fatty acid. These diets consistently induce functional deficits in electroretinograms, reflex responses, reward or avoidance induced learning, maze learning, behaviour, and motor development compared with ω3 replete groups. Boosting neural tissue docosahexaenoic acid (DHA) by feeding preformed DHA enhances visual and cognitive function. Though no human randomized controlled trials on minimal ω3 requirements in pregnancy and lactation have been conducted, the weight of animal evidence compellingly shows that randomizing pregnant or lactating humans to diets that include high linoleate oils as the sole source of fat would be frankly unethical because they would result in suboptimal child development. Increasing use of commodity ω3-deficient oils in developing countries, many in the name of heart health, will limit brain development of the next generation and can be easily corrected at minimal expense by substituting high oleic acid versions of these same oils, in many cases blended with small amounts of α-linolenic acid oils like flax or perilla oil. Inclusion of DHA in these diets is likely to further enhance visual and neural development.

Keywords: PUFA, animal studies, evidence-based nutrition, malnutrition, linoleic acid, linolenic acid.

Correspondence: J. Thomas Brenna, Division of Nutritional Sciences, Cornell University, Savage Hall, Ithaca, NY 14853, USA. E-mail: jtb4@cornell.edu

Introduction

Animal studies and polyunsaturated fatty acid (PUFA) maternal and child nutrition

Animal studies form the basis of most of the detailed knowledge of human nutrition, including maternal and child fatty acid (FA) nutrition. The vast majority of maternal and child FA nutrition studies have focused on FA with more than one double bond [PUFA, long chain polyunsaturated fatty acid (LCPUFA)/highly unsaturated fatty acid]. Remarkably, little attention has focused on the other classes of fatty acids; particularly surprising is the dearth of studies with *trans* FA, which human studies have associated with reproductive difficulties (Brenna & Lapillonne 2009) and which have been the subject of great attention for their influence on blood biomarkers for cardiovascular disease.

PUFA are essential nutrients known to be required for reproductive success in animals and are at high concentration in specific membranes of every cell; some cell types are especially rich in LCPUFA. PUFA are not only best known and understood as signalling molecules but also play very specific biophysical roles which have only been partially elucidated. Of specific interest is docosahexaenoic acid (DHA), which accumulates in the human central nervous system (CNS) rapidly *in utero* (Martinez 1992) and post-natally up to the age of 18 years (Carver *et al*. 2001). Scores of studies from the 1960s onwards in animals and humans have documented perinatal effects of PUFA nutrition for mother and child on CNS function. They take the form of feeding studies in which experimental groups are deprived of all-fat or all ω3 fatty acids, or given only linolenic acid (ALA) as a source of ω3, or given DHA in addition to ALA. The roles of PUFA have been gleaned almost exclusively from basic research in animals and in cells derived from animals, and confirmed with measurements accessible in human studies. For example, the early work developing models of fat deficiency was ineffective until the development of the water-deprived rat model of Thomasson (1953). That model exploited the water permeability of the skin attendant with fat-free feeding that halts growth. Growth rates measured for rats on diets with various FA as the sole source of fat enabled rapid discovery of what today are called 'omega-6' PUFA as essential for skin integrity, in contrast to saturated and monounsaturated FAs which are inactive, and omega-3 PUFAs which are active only at a trace level. Extension of these results to humans could then be confirmed in limited, ethically acceptable studies that had immediate influence on the composition of total enteral or parenteral feeds, many of which were, in the 1970s, fat free (Wene *et al*. 1975).

Collectively, PUFA nutrition studies during pregnancy and lactation are highly relevant to maternal and child nutrition in the developing world precisely because they are easily achieved with widely available foods. Oils such as commodity sunflower, peanut or safflower oils, which are very high quality in their production and taste, supply tiny amounts of ALA. In countries such as India, an increasingly wealthy middle class has perceived their high quality and has adopted them as basic cooking oils. The results of animal studies leave no room to doubt that negative developmental consequences are in store for a developing fetus of a pregnant woman chronically consuming high amounts of these oils. Delayed and eventually suboptimal retinal and cognitive developments will result, with likely negative effects on behaviour as well as negative neuropsychiatric effects for the mother. This is a clear case where the evidence from animal studies goes far beyond mere hypothesis generation (Smit *et al*. 2009) to provide clear and convincing evidence that randomization of a pregnant or lactating woman to, for instance, a diet containing sunflower, safflower or other oils with negligible ALA as the only source of fat, would be frankly unethical. Here, the evidence from animal studies is unequivocal and sufficient to set dietary guidelines. The animal data lead to the specific guideline that pregnant and

Key messages

- Studies show convincingly that high LA/low ALA (high ω6/low ω3) seed oils fed as the exclusive source of fat to pregnant and lactating primates, mice, rats, and other species leads to biochemical, neural, visual, and behavioral abnormalities in the offspring.
- Diets with fat dominated by linoleic acid (LA), such as those in which nearly all of the fat comes from seeds (e.g. cereal grains and peanuts) and seed oils (e.g. corn, peanut, safflower and sunflower oils), fail to support optimal omega-3 long chain polyunsaturated fatty acid status.
- The developing world is particularly vulnerable to widespread adoption of high LA oils because of the low cost and high quality taste and food performance of such oils.
- A strategy to normalize human PUFA levels is to replace high LA commodity oils with high oleic acid oils, such as have been developed for peanut and soy oil, which have dramatically reduced LA content and fatty acid compositions resembling that of olive oil.

lactating women should not consume any of the commodity seed oils with high linoleic acid (LA) and negligible ALA as a major dietary component. This concept is described in more detail in the *Developmental studies of ω3 deficiency* section and in the tables.

For context, we briefly contrast the relative roles of human and animal studies in the development of guidelines for human nutrition. The application of human studies to nutrition, in a framework borrowed but distinct from evidence-based medicine (EBM), evidence-based nutrition (EBN), has recently been taken up (Blumberg *et al*. 2010; Mann 2010).

Animal vs. human studies in nutrition and biomedicine

The most direct evidence for medical and nutritional practice is direct study of humans in studies designed to match participants between control and experimental groups and randomize unknown factors so as to isolate the particular treatment under investigation, including masking/blinding of investigators and participants to their respective study groups. Apart from the obvious ethical limitations on human medical research compared with animal research, a number of factors are unique to nutrition studies.

- Human studies cannot reduce nutrient intake of a control human group to zero (Von Schacky 2008; Blumberg *et al*. 2010) and rely instead on raising nutrient intake in a vulnerable segment of participants to a level where function is measurably improved (Innis & Friesen 2008).
- Human double blind randomized controlled trials (RCT) require that control and experimental treatments are indistinguishable to investigators and participants. They are thus limited to supplementation trials, and double-blinded studies of food substitutions, e.g. fish for beef, cannot be conducted in humans. They can, however, be conducted in animals.
- The vast majority of multigenerational phenomena carried over three or more generations, such as nutritional programming (Barker 2007), have been investigated using animal models. Human lifespan and ethics limit RCT to partial transgenerational studies (e.g. parent–progeny), and even here, the ability to control and reproduce results, both vital to developing firm scientific results, is severely limited.
- Human double-blind RCTs are limited in their duration as a practical matter. For instance, it is impractical to investigate long-term outcomes, such as brain function in adults and the elderly because of perinatal effects, because of the many decade timescales and because blinding cannot be maintained indefinitely.
- Human RCTs that rely upon sample participants who are expected to develop certain characteristics similar to broad demographics are risky. An example comes from a recent DHA trial in ageing, designed to investigate a possible treatment to '. . . slow the decline of cognitive and retinal function in older people by increasing daily dietary intake of n-3 LCPs' (Dangour *et al*. 2006). The trial failed to show a decline in cognitive function in the control group (Dangour *et al*. 2010), and thus was unable to address the original question.

An overzealous drive to formalize a universal algorithm for the acceptance or rejection of novel medical therapies can lead to an overemphasis on EBM, summarily rejecting common experience and declaring other approaches, such as animal research, of minimal value. Requiring all health-related practices to pass an EBM test or be abandoned leads to a sort of *reductio ad absurdum* (Smith & Pell 2003). A demand for RCT in the face of overwhelming evidence from animal studies or human practice is harmful if it perpetuates unsafe practices.

Human studies in maternal and child nutrition are particularly prone to difficulties because of practical constraints on experimentation with humans in these vulnerable physiological circumstances as well as the long-term nature of the outcomes for the child. Careful use of animal studies can lead to a clear picture of the implications for humans without recourse to impractical human studies. On the basis of animal studies and a consideration of basic, non-EBM research in humans, many trials would be blatantly unethical or impossible to conduct in humans. An ethical sense has been broached as a guide for health practice (James & Cleland 2004).

Fractured food and fragmented health priorities

Importantly, changes in the nutrient mix of the food supply have never been subject to evidentiary oversight except in special cases such as infant formula. That is, evidence is not required to introduce and expand the use of novel foods. Food components are typically derived from chemical or physical fractionation of foods in their natural form into components that have specific functional roles in food preparation, processing or palatability. While safe and nutritious as minor components of a diet, their adoption as staples becomes the base of epidemic deficiency conditions. A familiar example is wheat milling in which the germ, middling and bran, containing the vitamins, minerals and fibre, are removed. This leaves matter that makes a versatile baking flour, giving products that are high quality in their taste and easy to chew and digest. The scourge of pellagra, beriberi and a spectrum of deadly deficiency conditions of the 19th century was, in part, a direct result of the adoption of milled flour as a staple, as discussed in a highly recommended paper by Brody (1945). Recent consumer demand for whole grains is encouraging; though at least in the United States, the separation of commodity corn and soy into dozens of fractions for later recombination into processed foods raises similar concerns.

From the perspective of fats and FAs, the rise of seed oils from near zero intake in 1900 to greater than 20% of calories is a remarkably unrecognized phenomenon even among nutritional scientists and virtually transparent to the medical community. In the 1970s, corn oil was advocated for its high 'PUFA' content. With increasing recognition that corn is nearly devoid of ALA and with the introduction of herbicide-resistant genetically modified soy, soy oil production skyrocketed to now dominate US edible oil intake and indeed the fat intake of Americans. This has all occurred in the laudable industry-driven trend to reduce the cost of food. As shown by animal studies, soy oil is a crucial improvement over ω3-deficient oils, and animal studies also point to more gains that can be made by lower ω6 oils.

Biomedical research measures outcomes that are related to specific conditions, diseases, organs or functions. Following this, medicine is naturally fragmented into specialties: cardiology, psychiatry, neurology, oncology, and each concerns itself with conditions relevant to its subspecialty. From the perspective of fats and FAs, outcomes relevant to heart health have overwhelmed other outcomes, in part because dietary fat and cardiovascular disease are linked via cholesterol-laden vascular plaques, and in part because of the obvious symptomatology of cardiovascular disease. Although some of the basic tenets that food fats are linked to heart disease remain controversial (Lands 2008), there is good evidence for the effect of polyunsaturated fats on lowering of disease markers, though not necessarily mortality (Skeaff & Miller 2009; Ramsden *et al*., 2010). It is largely for cardiovascular benefits that high ω6 oils have been promoted. However, there is no question from animal studies that high ω6 oils reduce anabolic conversion of ALA to ω3 long-chain metabolites [eicosapentaenoic acid (EPA) and DHA] and reduce incorporation of EPA and DHA into membrane phospholipids. Neural tissue is especially rich in DHA. An unqualified focus on heart health presumed to be enhanced by a fractured food, e.g. high ω6/low ω3 seed oils as discussed later (Table 2), fails to account for the health of the whole person and the future of the species. Animal studies show that heart health and neural health can be achieved with the same recommendations.

Animal studies in human nutrition

There is nothing new about the application of animal data to set human requirements. Most fundamental knowledge about human nutrient requirements rest upon a vast literature on animal nutrition studies, and confirmed in humans. Indeed, scientific progress in metabolism and nutrition, and especially establishing the chemical compounds and elements required for health, did not start in earnest until the principles of animal experimentation were established. Seminal milestones in this progress were recognition of animal conditions that are analogous with human conditions. The latter 19th century saw the germ hypothesis of Pasteur occupy such a central role in human medicine that virtually all human conditions were ascribed to infectious agents, whether or not

they satisfied Koch's postulates (Swazey & Reeds 1978). Hindsight reveals reports scattered through the medical literature showing that intake of particular foods alleviates epidemic diseases – a notable example is scurvy, which was reported as early as 1720 to be cured by green vegetables (Kramers 1720; Bethke 1934). This and many other similar observations of what turned out to be nutrient deficiencies were ignored by the scientific community until demonstrations in animal studies, confirmed in humans, showed in dramatic fashion that trace dietary components, in addition to macronutrients, were required for proper growth and reproduction.

Even after ample compelling scientific data were available, resistance to adoption of nutritional causes for widespread human diseases was strong. The case of pellagra, niacin deficiency, is especially well documented and dramatic. The legendary experiments of Joseph Goldberger are widely credited as the final compelling demonstrations of the nutritional origin of pellagra (Kraut 2010). As late as 1912, the origins of pellagra were unknown, but variously held to be related to a microbe, a fly of field origin because of its prevalence in rural areas, sunlight, corn made toxic by a fungus and myriad other factors as discussed in *A Treatise on Pellagra*. This book focused on observations in humans primarily with a public health perspective as practised at the time and was little informed by animal studies; its author was able to say 'I can deny that bad nourishment in the sense of insufficient nourishment is a cause.' (p 55), and in response to a colleague's observation that pellagra is dramatically improved by a 'change in food', he countered that 'the same wonderful change is often brought about . . . without any change of food at all and by the use of arsenic . . .' (p. 74) (Wood 1912). Notably, by this time Hopkin's concept of accessory food factors [Funk's 'vitamines' (Reed 1922)] based on observations a half century earlier (Hopkins 1906), including the observations of Eijkmann of the cure of beriberi in pigeons, was long established (Hopkins 1929). Goldberger famously injected into himself and his assistants several millilitres of blood, and they inoculated themselves with the secretions, and swallowed scabs, of pellagra sufferers. Though neither he nor his assistants developed pellagra, resistance to the nutritional theory of pellagra remained (Kraut 2010). Eventual acceptance of the nutritional origins of pellagra came with Goldberger's own studies identifying black tongue in dogs as canine pellagra (Goldberger & Wheeler 1928), as well as the work of many as purifications eventually yielded chemically pure crystalline niacin, an effort aided indispensably by animal bioassays. It is through this lens of documented historical resistance to animal data that modern notions of medical evidence should be viewed.

EBM vs. EBN

Contrasting issues between drug-based and procedure-based medicine vs. nutrition have led to very recent discussions of the separate issues facing nutrition research (Blumberg *et al*. 2010; Mann 2010).

EBM as a means for adopting novel therapies seeks to develop a hierarchy for the compelling nature of results. There is much written on the topic, with considerable diversity of opinion, as a Google search will reveal. Important to the present discussion is the consensus that multiple, consistent RCTs in humans that can be put into a meta-analysis framework constitute the most convincing evidence, with observational studies less compelling (prospective, retrospective), and animal studies often said to be unacceptable. EBM via RCT applies most appropriately to synthetic drugs of exogenous origin or to novel practices that improve patient (human) outcomes. All factors other than those under study are controlled (e.g. equal genders) or assumed to be as evenly distributed as possible among groups by randomization. Importantly, the nature of the control group is a factor that limits interpretation of the effects of the experimental treatment. Moreover, the types of studies that can be done are limited by ethical considerations that are driven both by common experience (Smith & Pell 2003) and by the unambiguous results of animal studies. The latter are relevant for maternal and child FA nutrition, particularly in developing countries.

Attempts to apply these principles to nutrition are imperfect for reasons related to the differences between drug intake/medical intervention and food intake in humans. Human intake of drugs, or use of

medical services, is inherently optional and can be discontinued in the case of any intervention that is eligible for an RCT, namely any that does not have a recognized benefit. In contrast, human intake of food and natural nutrients cannot be discontinued whether or not it has been shown to be efficacious for any particular outcome. As discussed next, these issues are of particular relevance for FA requirements. Evidence for a requirement for ALA is compelling and a prime example where animal studies suffice to inform general requirements for humans, if not precise numbers, even in the absence of human studies. Indeed, the human studies would be unethical by virtue of the very animal studies that suggest them, as discussed next.

Developmental studies of ω3 deficiency

The role of animal studies for informing general PUFA nutrition was reviewed 10 years ago (Innis 2000). It has long been known that rats deprived of dietary fat are incapable of reproduction. Studies in the 1960s established that females deprived of PUFA abort or pups die soon after birth, and males are sterile and refuse to mate, and later studies of low-fat diets showed impairments in reproductive success as well (e.g. Pudelkewicz *et al*. 1968; Sinclair & Crawford 1972 and references therein). Dietary requirements for ALA were estimated at about 0.5% of calories for both genders, and for LA they were estimated at 0.5% for females and 1.3% of calories for males. Titration of LA in maternal rat diets to establish the level at which arachidonic acid plateaued in organs of male pups yielded an estimated requirement of 1.2 g/100 g food intake, or about 1% by weight excluding fibre and moisture (Bourre *et al*. 1990). Importantly, as observed in later rat studies focused on growth and biochemical measures (Cunnane & Anderson 1997), LA and ALA can substitute for one another in the absence of the other for growth, skin barrier function and other functional outcomes to a degree that tends to be unappreciated. However, they are neither biochemically interconvertible in mammals [as opposed to worms, for instance (Spychalla *et al*. 1997)], nor are they interchangeable in all molecular processes in which they are involved. Tissue AA (arachidonic acid), derived from LA, is most closely associated with growth, inflammation, vascular and immune function, blood clotting, and parturition, whereas DHA, derived from ALA, is associated with visual and neural function, and recently the resolution of inflammation. The two PUFA families mutually participate in most functions, with competitive and complementary roles both well characterized.

LA and ω3 deficiencies are usually studied in maternal and child nutrition for their effects on neural function because of the high concentration of DHA in the CNS. Table 1 presents more than 50 studies in which female animals were deprived of a dietary source of ω3 PUFA during pregnancy and/or lactation, and which reported functional or biochemical effects as well as a few related studies. Multigenerational deprivation of ω3 was used to induce reduction of tissue ω3 PUFA because of earlier observations that adult animals avidly retain ω3 once replete (Tinoco *et al*. 1971). Subsequent studies evaluated the effects of depleting ω3 with fat-free or ω3-deficient diets on retina composition and function. The deficiency of ω3 because of sunflower oil feeding resulted in similar rates of pregnancy, pups per litter, birth weight, food intake and weight of pregnant or lactating females, and pup growth during suckling; however, compared with soy oil control, ω3 deficiency resulted in threefold greater perinatal mortality through three generations (Guesnet *et al*. 1986). Nevertheless, ω3 deficiency as an isolated nutritional factor in ω6-replete developing animals could be studied using sunflower or other ω3-deficient oils. It is this experimental paradigm that provides data directly on the functional consequences of ω3-deficient oil consumption through pregnancy and lactation. Importantly, the range of studies in Table 1 shows that safflower, sunflower, peanut (groundnut) or cottonseed oil, all with very high LA and very low ALA, induce neurological abnormalities consistent with studies of purified LA.

Brain and retina of diverse animal species have long been known to be rich in ω3 PUFA and DHA in particular (Lesch 1969; Anderson 1970). That DHA was actively conserved by species was shown in stark fashion in mammals of widely divergent natural habitats and dietary habits that apparently induce equally

Table 1. Animal studies of ω3 or PUFA deficiency in pregnancy and/or lactation with emphasis on functional (F) and/or function-specific biochemical (B) outcomes in ω3 deficiency studies

	Reference	Experimental	Control	Species	Outcome
1	Tinoco *et al*. 1971	LA-ME	+ALA-ME	Rat	F: Neither growth nor reproduction affected through two generations. No overt deficiency symptoms. B: 22:4ω6 and 22:5ω6 replaced 22:5ω3 and 22:6ω3 in tissues.
2	Benolken *et al*. 1973	Fat-free + LA	'Lab chow'	Rat	F: Reduced ERG responses, a, b wave amplitudes.
3	Messeri *et al*. 1975	Fat-free	Corn or sunflower or olive	Rat	F: Maternal deficiency during gestation reduced conditioned learning (PUFA deficient ω3 deficient)
4	Wheeler *et al*. 1975	Fat-free	18:1ω9 or LA or ALA	Rat	F: Reduced ERG responses, a, b wave amplitudes. Effects ascribed to ω3 deficiency.
5	Lamptey & Walker 1978b	Hydrogenated coconut	Corn	Rat	F: Maternal diet influenced audicular startle, cliff-drop aversion, negative geotaxis, and bar holding. Deficiency in late gestation less effective than late gestation and +lactation. (PUFA deficient vs. ω3 deficient)
6	Lamptey & Walker 1978a	Hydrogenated coconut	Corn	Rat	F: Maternal PUFA deficiency-impaired learning assessed by Y-maze discrimination in pups. Deficiency in late gestation less effective than late gestation and +lactation. (PUFA deficient vs. ω3 deficient)
7	Neuringer *et al*. 1984	Safflower	Soy	Rhesus	F: Visual acuity reduced by preferential looking method in ω3 deficient neonates
8	Neuringer *et al*. 1986	Safflower	Soy	Rhesus	F: Subnormal visual response; prolonged ERG recovery time.
9	Yamamoto *et al*. 1987	Safflower	Perilla	Rat	F: Longer learning time and lower frequency of correct responses in the ω3-deficient group.
10	Watanabe *et al*. 1987	Safflower	Perilla	Rat	F: ERG a- and b-wave amplitudes lower in ω3 deficient pups. B: Reduced number of phagosomes in the retinal pigment epithelium.
11	Bourre *et al*. 1989	Sunflower	Soy	Rat	F: ERG abnormalities; no effect on motor activity; serious effects on learning tasks (shuttle box); faster mortality in response to intraperitoneal neurotoxin injection. B: Nerve terminal NaK-ATPase reduced by 40%; whole brain 5′-nucleotidase reduced by 20%; 2′3′-cyclic nucleotide 3′-phosphodiesterase reduced by 12%
12	Reisbick *et al*. 1990	Safflower	Soy	Rhesus	F: Polydipsia when ω3 deficient pre + post-natal
13	Reisbick *et al*. 1991	Safflower	Soy	Rhesus	F: Polydipsia when ω3 deficient pre + post-natal
14	Stinson *et al*. 1991	Safflower	Flax	Rat	B: ROS 22:6 turnover was not measurable in ω3 deficient compared with 19-day turnover in flax-fed.
15	Bush *et al*. 1991	Safflower	Soy	Rat	F: ω3 deficient less susceptible to light damage than soy.
16	Beauge *et al*. 1992	Sunflower	Soy	Rat	F: ω3 deficit unable to develop tolerance to alcohol.
17	Ziylan *et al*. 1992	Peanut	Peanut + rapeseed	Rat	B: No differences in transport of α-aminoisobutyric acid or phenylalanine.
18	Reisbick *et al*. 1992	Safflower	Soy	Rhesus	F: Polydipsia when ω3 deficient either prenatal only or post-natal only.
19	Ammouche *et al*. 1993	Sunflower	Peanut + rapeseed or salmon	Rat	B: Cytochrome P450 reduced 11% in ω3 deficiency; NADH-cyt-c-reductase act reduced 23% in ω3 deficiency; cytochromeb5 10% greater in ω3 deficiency; fish oil increased cyt P-450 concentration and NADPH-cytochrome-c reductase, aniline hydroxylase, and aminopyrine N-demethylase.
20	Gerbi *et al*. 1993	Sunflower	Soy	Rat	B: NaK-ATPase from ω3 deficient had three inhibitory processes vs. two for ω3 replete

Table 1. *Continued*

	Reference	Experimental	Control	Species	Outcome
21	Nakashima *et al.* 1993	Safflower	Perilla	Mice	F: Learning (time to solve) Morris water maze was lower in ω3 replete (perilla oil) animals; ω3-deficient animals more sensitive to pentobarbital anaesthesia; increased locomotion by scopolamine injection was lower in ω3 deficit group.
22	Reisbick *et al.* 1994	Safflower	Soy	Rhesus	F: ω3 deficient initiated more bouts of stereotyped behaviour in their home cages; locomotion bouts were also more frequent in ω3 deficient; . . . similar partial social isolates or those whose surroundings have been disrupted.
23	Ammouche *et al.* 1994	Sunflower	Peaunt + rapeseed or fish	Rat	B: NTPase greatest in ω3 deficiency.
24	Wainwright *et al.* 1994	Safflower + Olive	+Flax	Mice	F: No changes in Morris water maze learning; ω3 deficient had longer paw-lick latencies on hot plate.
25	Frances *et al.* 1995	Peanut	Peanut + rapeseed	Mice	F: No effects on anxiety, neophobia, defensive behaviour; ω3 deficient had less efficient learning and poorer escape behaviour.
26	Frances *et al.* 1996a	Peanut	Peanut + rapeseed	Rat	F: Morris maze performance reduced in ω3 deficiency; morphine-induced increase in locomotion occurred faster and was greater in ω3 deficiency; morphine-induced early hypothermia was greater and later hyperthermia was smaller in ω3 deficiency; pain thresholds were not affected.
27	Frances *et al.* 1996b	Peanut	Peanut + rapeseed	Mice	F: Habituation in exploration test, greater time in forced swimming, and trend in escape attempt reduced in ω3 deficiency.
28	Organisciak *et al.* 1996	Cottonseed	Cottonseed + flax	Rat	F: ω3 deficient *less* susceptible to light damage than flax
29	Weisinger *et al.* 1996	Safflower	Canola or fish oil	Guinea pig	F: ERG response amplitudes depressed in ω3 deficiency and in DHA excess ('inverted U-shape' response).
30	Yoshida *et al.* 1997b	Safflower	Perilla	Rat	F: Inferior learning in a brightness discrimination task. B: Reduced synaptic vesicles in terminals of the hippocampus CA1 region.
31	Reisbick *et al.* 1997	Safflower	Soy	Rhesus	F: ω3 deficient gave longer individual looks in both immediate and 24-hour novelty preference.
32	Pawlosky *et al.* 1997	Corn	+20:4+DHA	Cat	F: ω3 deficient had increased ERG implicit times for a- and b-wave. B: ω3 deficient corn oil supported brain and retinal rod outer segment 20:4n-6, but not 22:6n-3.
33	Bourre *et al.* 1999	Peanut	Peanut + rapeseed	Rat	F: No change in wave 1 amplitude or latency; wave 2 amplitude and latency dropped with age for both; wave 3 amplitude and latency dropped with age faster for ALA deficient.
34	Carrie *et al.* 1999	Peanut	Peanut + rapeseed	Rat	F: Decreased learning on a passive avoidance task – ERG normal.
35	Greiner *et al.* 1999	Safflower	+Flax	Rat	F: ω3 deficient made more total errors in a seven-problem, two-odor discrimination task. Escape latency in Morris maze longer.
36	Frances *et al.* 2000	Peanut	Peanut + rapeseed	Rat	F: Preference for a sucrose solution lower in ω3-deficient; morphine-induced place preference conditioned in controls but not ω3-deficient.
37	Carrie *et al.* 2000	Peanut or +DHA PL	Peanut + rapeseed or +DHA PL	Mice	F: Rearing activity reduced in ω3 deficient, restored with DHA; learning reduced in ω3 deficient, restored with DHA; anxiety not fully corrected with DHA; no difference in Morris maze or passive avoidance.

Table 1. *Continued*

	Reference	Experimental	Control	Species	Outcome
38	Ikemoto *et al*. 2000c	Safflower	+0.6% EPA EE or perilla	Rats	B: Nerve Growth Factor (NGF) lower in hippocampus of ω3 deficient and in ω3 + EPA compared with perilla.
39	Ikemoto *et al*. 2000d	Safflower	+9%DHA or perilla	Rats	B: Reduced biosynthesis of lysophosphatidylethanolamine (lyso-EtnGpl) in ω3 deficient
40	Ikemoto *et al*. 2000a	Safflower	Perilla	Rats	B: Reduced activity of retinal phospholipid biosynthetic enzymes. In ω3 deficient
41	Ikemoto *et al*. 2000b	Safflower	Perilla	Rats	B: ω3 deficiency induced a change in retinal but not pineal lysosomal enzymes (beta-glucosidase, beta-glucuronidase, hexosaminidase and acid phosphatase).
42	Ikemoto *et al*. 2001	Safflower	Perilla or 1.2% oleic + 1.2% DHA	Rats	F: Reduced learning in a brightness-discrimination test in ω3 deficiency; DHA-diet reversed restored learning.
43	Yoshida *et al*. 2001	Safflower	Perilla	Rats	F: Reduced learning in a brightness-discrimination test in ω3 deficiency; DHA-diet reversed restored learning. B: Brain microsome oligosaccharides altered in ω3 deficiency.
44	Weisinger *et al*. 2002	Safflower	Safflower + flax + DHASCO	Rat	F: ω3 deficiency reduced ERG retinal sensitivity and increased b-wave implicit times.
45	Carrie *et al*. 2002	Peanut, or +DHA at 8 months	Peanut + rapeseed or +DHA	Mice	F: No differences in motor or exploratory activity; DHA reduced anxiety in light/dark test; ω3 deficiency reduced learning in Morris maze, restored in aged animals by DHA; ERG poorer in ω3 deficiency, DHA improved ERG.
46	Jeffrey *et al*. 2002	Safflower	Soy	Rhesus	F: ERG rod sensitivity reduced by 40% in ω3-deficient 9-year-olds. Rod recovery delayed 20% in ω3 deficient; no effect on cone phototransduction.
47	Jeffrey *et al*. 2002	Safflower	Soy	Rhesus	F: ω3 deficient: rod recovery delayed by 30%, ERG implicit times by 5%; no effect on amplitudes, time to initiation of rod recovery, or phototransduction.
48	Catalan *et al*. 2002	Safflower	+Flax+DHA	Rat	F: ω3-deficient rats acquired most simple two-odor discrimination tasks but were deficient in the acquisition of a 20-problem olfactory learning set.
49	Ahmad *et al*. 2002	Safflower	+Flax+DHA	Rat	B: No differences in volume, density, total number, and cell body size of neurons in CA1-3, granular and hilar layers of the hippocampus except for in cell body size; CA1 pyramidal neurons in the LA group were significantly ($P < 0.04$) smaller than neurons in the ALA/DHA group at the septal location.
50	Takeuchi *et al*. 2002	Safflower + coconut + olive	+Fish oil	Rat	F: EEG fast activities lower in ω3 deficit pups; learning slower in ω3 deficiency. B: ω3-deficient pups had lower noradrenaline in cerebral cortex, hippocampus, striatum; dopamine was lower until 7 days.
51	Moriguchi & Salem 2003	Safflower + hydrogenated coconut	+Flax +flax repletion	Rat	F: Morris water maze learning poorer in ω3 deficiency; repletion reversed some but not all learning deficit.
52	Niu *et al*. 2004	Safflower	+Flax +DHA	Rat	B: Reduced rhodopsin activation, rhodopsin-transducin (G(t)) coupling, cGMP phosphodiesterase activity, and slower formation of meta-rhodopsin II (MII) and the MII-G(t) complex relative to n-3 FA-adequate ROS. ROS membranes from n-3 FA-deficient rats exhibited a higher degree of phospholipid acyl chain order relative to n-3 FA-adequate rats.

Table 1. *Continued*

	Reference	Experimental	Control	Species	Outcome
53	Reinwald *et al.* 2004	Safflower	+Flax+DHA	Rat	F: Bone mechanical properties (energy to peak load) of tibiae showed that (n-3) deficiency diminished structural integrity.
54	Levant *et al.* 2004	Sunflower	Soy or + fish oil	Rat	F: Haloperidol-induced catalepsy was reduced in ω3-deficient pups and remediated by ω3; novel-environment or amphetamine-induced locomotion was higher in ω3 deficiency and remediated by ω3; no difference in response to a thermal stimulus; all these are dopamine-related behaviours.
55	Anderson *et al.* 2005	Safflower in gestation, then soy to 3 years	Soy	Rhesus	F: ERG did not recover completely even by 3 years, though tissue DHA recovered.
56	Lim *et al.* 2005	18:2EE	+18:3EE	Rat	F: ω3 deficiency had poorer escape latency and memory retention in Morris maze.
57	Vancassel *et al.* 2007	Spontaneous		Rat	Spontaneous hyperactivity, an ADD symptom, predicts rat frontal cortex DHA
58	Ozias *et al.* 2007	Safflower	Soy	Rat	F: None B: Reduction in brain PL DHA over multiple litters.
59	Fedorova *et al.* 2007	18:2EE	+18:3+DHA	Rat	F: ω3 deficiency impaired learning in the reference-memory version of the Barnes circular maze.
60	Connor *et al.* 2007	Safflower	+DHA+EPA or FAT-1	Mouse	F: ω3 deficit had abnormal retinal neovascularization
61	Levant *et al.* 2008	Safflower	Soy	Rat	B: Decreased hippocampal BDNF gene expression and increased relative corticosterone response to an intense stressor; BDNF gene expression and increased relative corticosterone response to an intense stressor; serotonin content and turnover in frontal cortex were decreased.
62	Fedorova *et al.* 2009b	Safflower	Safflower + flax + DHASCO	Rat	F: Impairment in Barnes maze during initial learning B: Reduced dopamine in ventral striatum; Enhanced 3,4-dihydrophenylacetic acid in frontal cortex and hypothalamus.
63	Fedorova *et al.* 2009b	Safflower (high oleic)	Safflower(high oleic) + flax or safflower + flax + menhaden	Rat	F: PPI deficit in ω3 deficient and in low ALA compared with high ALA and menhaden; high ALA lower than menhaden.
64	Davis *et al.* 2010	Safflower	Soy	Rat	F: Receptor alterations are similar to those found in several rodent models of depression; decreased density of ventral striatal D2-like receptors B: decreased density of ventral striatal D2-like receptors; higher density of D1-like receptors in the caudate
65	Levant *et al.* 2010	Safflower + low soy	Soy	Rat	F: ω3 deficient pups had higher levels of activity to familiar but not a novel stimulus, similar to human attention-deficit-disorder.

Other than very early works, papers that characterize exclusively responses of tissue fatty acid to intake are excluded.
F, functional outcome; B, biochemical outcome; LA, linoleic acid; ME, methyl ester; ALA, linolenic acid; ERG, electroretinogram; PUFA, polyunsaturated fatty acid; ROS, rod outer segments; DHA, docosahexaenoic acid; DHASCO, docosahexaenoic acid-rich single-cell oil; BDNF, brain derived neurotrophic factor; ADD, attention deficit disorder; DHAPL, DHA phospholid; EEG, electroencephalogram; EPA, eicosapentaenoic acid; PPI, pre-pulse inhibition.

widely different liver FA profiles. In more than 30 species, liver DHA varies over 30-fold, while brain DHA varies over 50% (Crawford *et al*. 1976). Visual, higher CNS functions and reflex will be considered separately, selectively providing an overview of a subset of studies in Table 1.

Vision

The perinatal dependence of retina on a supply of ALA or a DHA precursor was demonstrated in rats in the 1970s. Although the rod outer segments (ROS) containing the photosensitive rhodopsin turn over every 10 days, weanling rats cannot be depleted of DHA by consuming an apparently ω3-free diet. This and similar observations led to the conclusion that DHA is avidly retained by the nervous system in adulthood, and in part led to the necessary use of experimental models of ω3 nutrition in pregnancy and lactation. Depletion of DHA in pup retinas was achieved by feeding a fat-free diet through two generations to enable breeding while supplementing only female rats with LA through breeding and lactation. DHA dropped from 45% of ROS ethanolamine phospholipids to 19%, with an increase in 22:5n-6 to 10%. With a DHA reduction of more than 50%, electroretinogram (ERG) showed dramatic reductions in the amplitude of retinal response for both a-wave (photoreceptors) and b-wave (nerve signal transmission) (Benolken *et al*. 1973).

These results were repeated and extended in seminal primate studies by Neuringer and coworkers, which appear starting in the 1980s (Neuringer *et al*. 1984). Semi-purified diets were prepared with safflower oil as the only source of fat. Safflower oil has ample LA and very low ALA, and thus simulates most other seed oils (sunflower, corn, peanut, sesame, most tree nuts except walnut) as an LA-replete/ω3-deficient fat. Diets were fed to female rhesus monkeys for at least 2 months prior to conception and through pregnancy and lactation. Compared with soy oil-fed controls, preferential looking measures showed slower development of visual acuity in the first weeks of life. By 12 weeks, acuity in Snellen equivalents were about 20/100 and 20/50 for deficient and control neonates, respectively; the visual acuity of deficient animals developed at about half the normal rate. The key for interpreting this and many related results is the recognition that the development of visual acuity in infants is related to nervous system development, not to physical refraction that can be improved with corrective glasses. Plasma phospholipid DHA in the deficit group was less than half that of the control group at birth, and dropped to less than 10% of the control group by 12 weeks of age (Neuringer *et al*. 1984). In a similar, longer-term study, there were no differences between the two groups in food intake or body weight up to 22 months of life. Cerebral cortex and retinal DHA both dropped to less than 20% of control values by 22 months of life. ERG over the first 12 weeks of life showed deficits in b-wave recovery in between flashes, and peak latencies were delayed as well (Connor & Neuringer 1988). A study of baboons showed that preformed DHA consumption via formula or breast milk caused in the neonates significant decreases in a-wave implicit time and an increase in the sensitivity of the initial response ('ä'), a measure of the G-protein cascade intensity that initiates retinal response (Diau *et al*. 2003). Moreover, these responses were related to retinal DHA content, in a manner consistent with biophysical studies suggesting a highly specific interaction between DHA and G-protein-coupled receptors, such as rhodopsin, that is not possible for the ω6 analogue 22:5n-6 (Eldho *et al*. 2003; Gawrisch & Soubias 2008). Biochemical deficits in rhesus retinal DHA induced by ω3 deficiency during gestation were completely reversed in cerebral cortex but not in retina by ALA feeding from 0 to 3 years of life, suggesting that prenatal exposure results in long-term compositional changes that could manifest as subtle functional deficits (Anderson *et al*. 2005).

Nutrition studies in higher primates are particularly informative because of the similarity of retinal microarchitecture, relative brain size among primates and omnivorous natural diets (Su *et al*. 1999). Small laboratory species are of great value in elucidating detailed mechanisms. ERG of guinea pig pups of dams reared on safflower oil had lower minor losses in a-wave and b-wave amplitudes, with the main functional loss in retinal receptor response. After a 10-week repletion with control diets based on canola

oil, functional deficits were reversed (Weisinger *et al*. 1999). Further studies with rats confirmed that amplitude effects were smaller than sensitivity effects, consistent with findings in other species (Weisinger *et al*. 2002). Recent findings in mice (Tanito *et al*. 2010) are consistent with higher sensitivity of the ERG a-wave response with higher DHA, but also show greater vulnerability to photo-oxidative stress, consistent with earlier studies (Weisinger *et al*. 1996).

The picture emerging from these animal studies is consistent with a wealth of data indicating that human infant visual acuity benefits from a dietary supply of DHA (Brenna & Lapillonne 2009). Specific effects appear to be a delay in the development of retina sensitivity, and in more severe deficiency conditions, deficits in response amplitude. Very high DHA intakes appear not only to further potentiate retinal response but also may sensitize the retina to photo-oxidative stress, though recent evaluations do not report elevated incidence of age-related eye diseases or age-related macular degeneration in the Japanese, a population with high DHA intake and status (Wong *et al*. 2006; Kawasaki *et al*. 2010).

Higher order CNS function: learning, mood, motor function

Numerous studies have shown influences of PUFA nutrition on learning, mood and motor skills of developing animals. Apart from the early studies showing the constancy of brain DHA, many others show the influence of ω3 deficiency on specific brain regions (Diau *et al*. 2005), subcellular components (Foot *et al*. 1982; Bourre *et al*. 1984) including the vessels (Homayoun *et al*. 1988), and lipid classes (Carrie *et al*. 2000). Safflower oil ω3-deficient diets, which reduce regional DHA, cause morphological changes that result in learning deficits in mice (Yoshida *et al*. 1997a,b).

An early study was equivalent to the modern 'fetal programming paradigm' in which experimental treatment occurs only during pregnancy, and offspring are placed in identical conditions and tested later. Pregnant dams were fed one of four diets differing only in their fat (corn, olive, safflower, and 'saturated fat' with no LA or ALA) and then switched to 'standard laboratory diet' through lactation. Pups were tested with a shock-avoidance learning paradigm. The corn, olive and sunflower oil groups avoided the shock at approximately twice the rate of the saturated fat group, leading the authors to conclude that the mouse brain is vulnerable to nutrition-induced learning deficit during pregnancy (Messeri *et al*. 1975). Very similar studies that eliminated all dietary PUFA during the last third of gestation of rat dams resulted in permanently impaired learning in a Y-maze, but did not significantly impair learning when PUFAs were withheld during lactation (Lamptey & Walker 1978a).

Learning deficits as a result of ω3 deficiency induced by sunflower oil feeding (compared with soy oil control) were specific to CNS function and independent of retina ERG function as demonstrated in rats (Carrie *et al*. 1999). Rats fed sunflower oil through two generations died from exposure to a neurotoxin more rapidly than soy-fed controls, had ERG deficits and reduced Na^+-K^+-ATPase activity (Bourre *et al*. 1989). The deficit of ω3 was induced by sunflower oil feeding of rat dams through pregnancy and lactation, and pups were weaned to these diets; comparison was with a control diet rich in LA and including DHA, or to an ALA-rich perilla oil diet. Compared with sunflower oil, the perilla-fed animals had a greater percentage of correct responses in a brightness-discrimination task (Yamamoto *et al*. 1987). Learning deficits because of ω3 deficiency in maze escape tasks are found in rats using the Morris water maze; in previously deficient groups, performance is restored to control levels with extended repletion (Moriguchi & Salem 2003). These results have been repeated with the Barnes maze, a less stressful escape maze that does not require swimming. Moreover, the ω3-deficient rats performed much more poorly in the Barnes maze when the escape path was reversed compared with previous trials, a task requiring suppression of previously learned information and reassessment of strategy (Fedorova *et al*. 2009b). These tasks depend on the function of the hippocampus for learning and the frontal cortex for strategy, among other brain structures.

Omega-3 deficiency causes a reduction in dopamine in rat frontal cortex, which is associated with reduced reward-seeking behaviour. Peanut oil feeding was used to induce ω3 deficiency by feeding

to dams prior to breeding and through pregnancy and lactation, and then fed to pups. Compared with a control peanut oil/rapeseed oil diet, ω3-deficient pups had reduced preference for a sucrose solution (Frances *et al*. 2000). Others showed that ω3-deficient diets fed through rat pregnancy and lactation-induced noradrenaline to drop, EEG abnormalities and impaired learning in an avoidance test (Takeuchi *et al*. 2002). Second generation ω3-deficient rat pups performed more poorly than controls in a two-odour olfactory discrimination task (Greiner *et al*. 2001). Sunflower oil-fed ω3-deficient rats had reduced haloperidol-induced catalepsy than soy oil-fed controls, and remediation by feeding fish oil from weaning resulted in similar performance to controls. However, basal and amphetamine-stimulated locomotor activity was equivalent to that of the deficient animals and significantly different from that of controls, showing that some, but not all, behavioural abnormalities are reversed by ω3 repletion (Levant *et al*. 2004). These results are consistent with blunting of sensory-induced learning in ω3-deficient animals.

Rhesus monkeys deficient in ω3 because of safflower oil feeding exhibited stereotypical behaviour similar to that of monkeys that had been raised in partial social isolation, or had surroundings disrupted (Reisbick *et al*. 1994). Consistent with these observations, rats raised on safflower oil had normal spontaneous locomotor activity compared with soy oil controls, but displayed lower exploratory activity in a novel environment (Enslen *et al*. 1991). Recent data show that safflower oil-induced ω3 deficiency causes activity and habituation that is consistent with clinical observations in attention-deficit hyperactivity disorder (Levant *et al*. 2010), following earlier work suggesting that hyperactivity in the rat is related to frontal cortex DHA (Vancassel *et al*. 2007). At the other end of the dietary spectrum, DHA at 1% of FA fed to infant rhesus monkeys from 0 to 1 month of age resulted in improved motor abilities compared with controls receiving formula containing only ALA (Champoux *et al*. 2002). One per cent DHA is at the high end of reported human breast milk DHA levels (Brenna *et al*. 2007); baboon breast milk is similar to human breast milk in that high DHA diets result in high DHA breast milk (Diau *et al*. 2005).

These and other animal studies are consistent with numerous human studies suggesting that higher DHA status has a particular effect on attention (Brenna & Lapillonne 2009). The ability to solve problems requiring attention as well as strategy in 10-month-old human infants was found to be sensitive to DHA inclusion in formula (Willatts *et al*. 1998). Maternal DHA at birth is correlated with less distractibility in infants (Colombo *et al*. 2004). A very recent study showed persistent effects on sustained attention in 5-year-old children of supplementation of their mothers with 200 mg DHA per day during the first 4 months of breastfeeding (Jensen *et al*. 2010).

Reflex, startle

Involuntary movement reflects particular neural pathways and function of the CNS and periphery. An early study showed that omission of all PUFAs during gestation reduced acquisition of audicular startle, cliff drop aversion and other reflexes, and was potentiated if PUFAs were absent during lactation, compared with a corn oil (ω6 replete, ω3 deficient) control (Lamptey & Walker 1978b). Auditory brainstem responses arise from signal transmission stimulated by auditory clicks and reflect transmission velocities and intensities, analogous to visual cortex signals for visual stimuli. Peanut oil-induced ω3 deficiency did not result in significant differences in latency or amplitude in young rats compared with peanut/rapeseed oil controls. However, in ω3 deficiency, amplitude and latency decreased faster as the mice aged, and middle aged (6 months old) rats had similar response as aged (18 months old) rats (Bourre *et al*. 1999).

Recently, pre-pulse inhibition (PPI) on the acoustic startle response has been investigated in ω3-deficient rats. Deficient and low-ALA (0.4%, w/w) diets resulted in a deficit in PPI compared with a diet containing 2% DHA and 2% EPA, and a high ALA group was intermediate. These results showed excellent differentiation between low, high and DHA + EPA effects in a well-characterized paradigm (Fedorova *et al*. 2009a). Baboons and human infants achieve greater brain DHA when preformed DHA is in the diet either in breast milk or as a component of

formula, compared with LCPUFA-free formula (Cunnane *et al*. 2000). As such, rat PPI is a neural function that clearly differentiates between DHA-containing and DHA-free diets; these results thus indicate a functional role for preformed DHA that cannot be filled by ALA.

Implications for maternal and child nutrition in developing countries

A focus of medical recommendations and advertising on the cholesterol-lowering effect of high LA oils, ostensibly for heart health, ignores the impact on the developing fetus, a point made two decades ago (Holman *et al*. 1991). We are not aware of any existing human RCT testing the impact of low ALA oils as the sole source of visible fat in human diets through pregnancy or lactation analogous to the many studies in animals. Sensitive RCT of long-term human health outcomes or psychiatric effects would be prohibitively expensive and lengthy. The key question is: Are animal studies sufficient to set dietary guidelines for ALA? If the weight of animal evidence leads knowledgeable reviewers to conclude that definitive human RCTs would be unethical, then by definition it is sufficient to set dietary guidelines. Animal studies overwhelmingly support a general guideline to avoid ALA-deficient, high LA oils as a sole source of fat during pregnancy and lactation.

For the developing world, adding DHA to diets would be problematic on at least two levels: most sources of DHA are expensive, and DHA is the most oxidizable fatty acid in any food, reducing shelf life especially in situations in which cool dry storage is not possible. However, most evidence suggests that there could be major benefits from reducing excess LA to enable conversion of ALA to DHA. Substitution of oleic acid for LA would, in fact, increase stability and shelf life of foods because oleic acid has half the double bonds of LA (1 vs. 2). We can therefore advocate this strategy for improvement of fat composition in the developing world.

High-quality peanut, sunflower, safflower, rice bran and other oils with negligible ALA and replete with LA are widely available in, for instance, India (Ghafoorunissa 2005). Globally, consumers understandably adopt them as their wealth increases. Oil compositions are shown in Table 2, grouped according to whether they are predominantly saturated, high LA, contain a significant proportion of ALA, or are monounsaturated fatty acid (MUFA) oils that are high oleic/low LA. Notably, a focus on summary composition, particularly the sum of all PUFA, ignores the distinction between LA and ALA. An examination of the last column, total PUFA summing LA and ALA, shows that levels around 50% are common with oils that have 0 or 1% ALA. Specifying ALA content, rather than total generic PUFA, is necessary to judge healthfulness of oils. The MUFA-rich olive, a fruit and not a seed, has small amounts of PUFA and has been used as a plant oil for millennia.

Though the modern food system no longer resembles the subsistence farm, the results of modern nutritional science continue to converge on the conclusion that the composition of its products should resemble natural foods. Modern food systems include novel plants and animals, produced either by traditional breeding or by genetic modification. They are in large measure created to assist with production, processing and transport of desirable food. These practices have led to a global population carrying capacity far beyond that possible with traditional food production a mere century ago. Rather than summarily declaring all progress improper and unnatural, modern nutritional science points to ways in which modern food systems can be exploited to make more healthful food. A reasonable guideline is to use fractured foods to assemble products with compositions similar to natural foods, in an extension of the way that Funk, Hopkins and others reassembled protein, carbohydrate, fat and vitamin/mineral mixtures to establish the required nutrients. Monounsaturated oils with moderate ω6 and ω3 have very similar FA profiles to olive oil.

Specialty-bred oils high in oleic acid and low in LA are now available. High-oleic peanuts with 3% LA, rather than the 25% in commodity peanuts, are widely available in Australian foods such as peanut butter and snacks (PCA 2010). High-oleic sunflower and soy oil are also in production in some countries and under evaluation in others. Traditionally bred high-oleic soy has recently been announced (Pham

Table 2. Fatty acid composition of edible fats and oils, expressed as per cent fatty acid, weight-for-weight (%, w/w); equal to grams fatty acid per 100-g fat

	Saturated				MUFA	PUFA		Total mean ± SD		
	10:0 + 12:0	14:0	16:0	18:0	18:1ω9	18:2ω6	18:3ω3	SFA	MUFA	PUFA
Saturated fats										
Coconut	54	19	9	3	6	2	0	85	6	2
Palm kernel	52	16	8	3	15	2	0	79	15	2
Palm	–	1	44	5	40	9	0	50	40	9
Cocoa	–	–	26	34	33	3	0	60	33	3
Butterfat (cow)	6	11	27	12	29	2	0	56	29	2
Beef tallow	–	3	24	19	43	3	1	46	43	4
Lard (pork fat)	–	2	26	14	44	10	0	42	44	10
							Mean ± SD	60 ± 16	30 ± 15	5 ± 3
PUFA (linoleic)										
Peanut	–	–	11	3	52	28	0	14	52	28
Sesame	–	–	9	5	39	45	0	14	39	45
Safflower	–	–	7	2	19	75	0	9	19	75
Cottonseed	–	1	24	3	18	52	0	28	18	52
Sunflower	–	–	7	5	26	61	0	12	26	61
Corn	–	–	13	2	31	53	1	15	31	54
Rice bran	–	0.5	22	3	43	21	1	26	43	22
							Mean ± SD	17 ± 7	33 ± 13	48 ± 18
PUFA (α-Linolenic)										
Flaxseed (linseed)	–	–	6	3	20	17	53	9	20	70
Canola (rapeseed)	–	–	3	2	60	20	10	5	60	30
Soy (commodity)	–	–	11	4	23	54	7	15	23	61
Walnut	–	–	7	2	18	58	14	9	18	72
							Mean ± SD	10 ± 4	30 ± 20	58 ± 19
MUFA										
Olive	–	–	13	3	71	10	1	16	71	11
Avocado	–	–	14	–	65(16:1,6)	13	1	14	71	14
Soy (high oleic)*	–	–	11	4	75	<9	3	15	75	<12
Peanut (high oleic)	–	–	7	3	76	4	0	10	76	4
							Mean ± SD	14 ± 3	73 ± 5	8 ± 5

*For high oleic soy.

Data are from Firestone (1999) and Butzen & Schnebly (2007).

MUFA, monounsaturated fatty acid; PUFA, polyunsaturated fatty acid; SD, standard deviation; SFA, saturated fatty acid.

et al. 2010), is in field trials, and may be able to be served to those political districts concerned with genetically modified organism foods. Importantly, high-oleic soy also contains 3% ALA, and with <9% LA, compared with >50% LA in commodity soy, has native LA to ALA proportions more similar to pre-industrial diets. This oil, in effect, reproduces olive oil FA composition with higher ALA, in a high oil-yielding soy bean without resort to blending post-extraction. High-oleic oils with negligible ALA, such as high-oleic sunflower or peanut oil, could be blended with 10% of a high-ALA oil such as flax or perilla oil to create an oil with similar LA to ALA proportions. It is well established in animal studies that tissue LCPUFA, especially DHA, rises as LA drops (Mohrhauer & Holman 1963; Lands *et al*. 1990, 1992;Lands 2008), while human studies consistently show that supplementation with the ω3 precursor, ALA, 18:4n-3, or with EPA, fails to enhance DHA status. The provision of dietary DHA, or reduction of LA, both improve DHA status (Brenna *et al*. 2009). The weight of animal studies, consistent with available non-RCT human data, compellingly show that high-LA, low-LA oils severely compromise ω3

LCPUFA status, and with it, neural and visual developments that depend on DHA accretion.

Conflicts of interest

The author declares no conflicts of interest.

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