Omega-3 Fatty Acid Biochemistry: Perspectives from Human Nutrition

Sheila M. Innis, MSc, PhD

ABSTRACT The possibility that western diets poor in omega-3 and rich in omega-6 fatty acids contribute to the increasing burden of chronic diseases including neurological problems is becoming recognized. Modern, westernized diets provide 80 to 90% of polyunsaturated fatty acids as omega-6 linoleic acid (LA) and are depleted in omega-3 fatty acids, giving a distorted balance of LA to α-linoleic acid, and to eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). LA intakes exceed Δ-6 desaturase needs for maximal activity. LA accumulates in blood and tissue lipids with increasing intake, and this exacerbates competition between LA and limited omega-3 fatty acids for metabolism and acylation into tissue lipids. Increasing EPA and DHA intake decreases tissue omega-6 fatty acids while also providing EPA and DHA. However, strategies for EPA and DHA supplementation do not address potential underlying problems of omega-6 and omega-3 fatty acid imbalance in the food supply.

INTRODUCTION

The omega-3 fatty acids are essential nutrients throughout the animal kingdom explained by the enzymatic inability to insert a double bond at the Δ-15 position of an 18-carbon chain fatty acid. Three major omega-3 fatty acids in human diets are the 18-carbon chain α-linolenic acid (ALA; 18:3 omega-3) and its elongation and desaturation products eicosapentaenoic acid (EPA; 20:5 omega-3), and docosahexaenoic acid (DHA; 22:6 omega-3). The omega-6 fatty acids comprise a second series of essential fatty acids for which the major dietary fatty acid is linoleic acid (LA; 18:2 omega-6), with smaller amounts of its elongation and desaturation product arachidonic acid (ARA; 20:4 omega-6). The distribution of omega-3 and omega-6 fatty acids in plant and animal foods differs, and in recent history has been modified through agriculture, aquaculture, and food processing. This has relevance to human health not simply because of the changes in omega-3 fatty acids but also because of the changes in omega-6 fatty acids and their inseparable links to the metabolism and needs for omega-3 fatty acids.

Epidemiological and intervention studies have provided evidence that humans consuming diets rich in omega-3 fatty acids have reduced risk of cardiometabolic diseases, immune and inflammatory disorders, and a variety of neurological problems.[1–6] These findings coupled with abundant evidence that the amount, type, and balance of omega-3 and omega-6 fatty acids in the diet impacts plasma and tissue omega-3 fatty acids sets the stage to consider how recent changes in human fatty acid nutrition may have contributed to an increased incidence of a broad array of chronic diseases, extending to the central nervous system. This review summarizes dietary sources of the omega-3 and omega-6 fatty acids as a background to consideration of the implications of current dietary omega-3 and omega-6 fatty acids intakes to their metabolism and ultimately dietary needs for EPA and DHA.

Sources of Dietary Sources of Omega-3 Fatty Acids

The omega-3 and omega-6 polyunsaturated fatty acids (PUFAs) arise from the activity of Δ-15 and the Δ-12 desaturases.[7,8] These enzymes are present in plants and confer the ability to form double bonds at the Δ^{15} and Δ^{12} positions of an 18-carbon chain fatty acid leading to synthesis of ALA (18:3 $\Delta^{9,12,15}$, 18:3 omega-3) and LA (18:2 $\Delta^{9,12}$, 18:2 omega-6), respectively. The most widely studied membrane-bound plant fatty acid desaturases act on oleic acid (18:1 omega-9) esterified to the glycerol of phosphatidylcholine. These desaturases appear to be ubiquitous in higher plants and give rise to ALA and LA, the major PUFAs in plant lipids.[7,8] Although Δ-15 and Δ-12 desaturases are absent in animal cells, animals have Δ-6 (FAD2) and Δ-5 (FAD1) desaturases and elongases that enable modification of ALA and LA obtained in their diet by desaturation and chain elongation.[9] Thus, ALA and LA are transferred up the food chain from plants to animals, with potential desaturation and elongation to omega-3 and omega-6 fatty acids with 20 or more carbons and three or more double bonds, although the amounts formed depend on the species and diet. Several microorganisms also have the ability to form EPA and DHA. These include aquatic phytoplankton with the result that wild fish, shellfish, and molluscs are rich sources of EPA and DHA, and also low in omega-6 fatty acids. This dichotomy between the distribution of shorter 18-carbon chain omega-6 and omega-3 fatty acids ALA and LA in plants and mixtures of 18-, 20-, and 22-carbon chain omega-3 and omega-6 fatty acids in animals, with more omega-6 fatty acids in terrestrial animals and more omega-3 fatty acids in fish and seafood means that the types of plant and animal fats determines the amounts, types and balance of the different omega-6 and omega-3 fatty acids in the diet.[9–11]

Nutrition Research Program, Child and Family Research Institute, Department of Paediatrics, University of British Columbia, 950 West 28th Avenue, Vancouver, British Columbia V5Z 4H4, Canada.

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Adding complexity, plants vary widely in LA and ALA. ALA and LA are present in green leaves and modified flower structures (curd), such as the Brassica family cauliflower and broccoli, with an LA to ALA ratio well below 1. The amount of fat in leaves and flower structures, however, is low. Seeds, including hard-shelled seeds, and some unusual fruits and legumes such as olives, avocados, and peanuts are rich in fat, but also have variable amounts of ALA and LA, and hence variable ratios of LA to ALA. Many common refined vegetable oils such as soybean, corn, safflower, and peanut oil contain high amounts of LA, with LA representing over 30% of the total fatty acids. The omega-3 and omega-6 fatty acids in animals, poultry and fish also vary, not only because of the species but also because of the fat composition of the feed.[12–17] Because of relative abundance of omega-6 LA and ARA and low omega-3 fatty acids in many meats, poultry, and eggs in the modern food supply, diets with little or no fish are likely to be high in LA with high ratios of ARA to EPA and DHA.[10–11]

Historical Perspectives

Several reports have highlighted a potential discrepancy between human metabolism and the rapid increase in omega-6 fatty acids in the westernized food supply over the last two centuries.[18–22] Before the development of modern agriculture, human diets were limited to plant foods, naturally grazing animals, and fish. Consequently, the availability of omega-6 LA was limited. Because of the lower intakes of LA, omega-6 and omega-3 fatty acids were present in the diet in roughly similar amounts, with a ratio of 2 to 1 or lower.[19,23] Also important, a much greater proportion of the PUFAs were consumed as the 20- and 22-carbon chain omega-3 and omega-6 fatty acids, rather than as LA.[18,20] Over time, dietary fatty acid intakes changed, beginning slowly with the commencement of agriculture, the Industrial Revolution and increasing urbanization, although the impact on omega-3 and omega-6 fatty acid nutrition appears to have been small until about 100 years ago.[18,22] Available data based on knowledge of the food supply and some early dietary studies indicates that LA provided less than 2 to 3% dietary energy until the late 19th to early 20th century.[23,24] Fatty acid nutrition then began to change dramatically, with several prominent events contributing to the prolific increase in LA at many levels of the food supply. Notable among these events were the development of commercial refining of LA-rich seed oils, the invention of hydrogenation processes in 1897 that enabled vegetable oils to be solidified for use in shortenings and margarines, the replacement of animal fats with LA-rich oils in numerous foods, and dietary recommendations calling for increased consumption of LA-rich oils.

Illustrative of the recent introduction of LA-rich vegetable oils into the human diet, corn refining with processes for corn starch hydrolysis, and the first commercial corn oil production in the United States did not occur until 1889.[25] Similarly, the first successful processing of edible oil from soybeans using hydraulic pressing appears to have been around 1911, and by 1945 soybean oil was the leading edible oil in the United States.[26] Dietary intakes of PUFA-rich oils were then promoted primarily for the purpose of serum cholesterol lowering, with a further upward trend in dietary intakes following the 1961 recommendation from the American Heart Association and 1977 Dietary Goals for Americans that called for 10% dietary energy from PUFAs and 10% from saturated fatty acids, with a limitation on dietary cholesterol to less than 300 mg/d.[26–29] Of note, in his 1957 article, Keys[30] wrote that "even tremendous dosages of cholesterol added to the daily diet of man – up to 30 Gm. per day –have only trivial effect unless the dietary cholesterol is accompanied by additional fats." In the same year, Keys et al[31] explained that "to lower the serum-cholesterol adequately a decrease in the intake of saturated fats seems to be more important than an increase in linoleic." However, the implications of replacing saturated fats with LA-rich oils with respect to metabolism and needs for omega-3 fatty acids, or loss of nutrients such as DHA in cholesterol-containing foods, were neither considered nor at that time fully appreciated.

At the same time as LA intakes were increasing, omega-3 EPA and DHA appear to have been lost from the human diet. This is a more complex problem that includes loss of EPA and DHA from animal lipids because of feeding with grains high in LA and low in omega-3 fatty acids, and decreasing consumption of animal products such as eggs, which also provide EPA and DHA.[12–22,32] Although grass fat has 14% LA and 46 to 49% ALA giving ad libitum grass-fed cattle only about 2% LA and a omega-6 to omega-3 fatty acid ratio of 2.3:1 in muscle fatty acids, similar to that of wild ruminants, meat from modern domesticated cattle has negligible EPA and DHA and ratios of ARA to EPA plus DHA well over 50:1.[14,15] Similarly, loss of omega-3 fatty acids and an increase in LA has occurred in poultry and eggs, but as is well known, this can be reversed by feeding chicken and laying hens with feeds high in ALA, EPA, and DHA.[12,13,16,17]

To summarize, the evidence that the production and consumption of LA has risen over the last 150 years is irrefutable.[19,22,23,29] LA now represents 80 to 90% of all the PUFA in western-style diets, with average intakes of about 6.7% energy from LA in the United States, and 5% energy in Canada and Europe.[28,32–34] As average intakes, half of the population has intakes of LA above these values. These high intakes of LA are possible only through the wide-spread use of LA-rich grains and oils at multiple levels of the food supply. ALA provides about 0.5 to 0.7% dietary energy, which gives an average dietary LA to ALA ratio of about 10:1, although again this ratio is much higher in the diet of many individuals.[10,19,24,35] The median intake of EPA plus DHA in many western nations is 0.1 to 0.3 g/d, with about 0.1 to 0.2 g/d from ARA.[35] This means that less than 0.5% of dietary PUFA are now consumed in the form of the long-chain EPA, DHA, and ARA. Whereas nutrient deficiencies

have historically occurred because of inadequate food availability or poor food choices, modern humans are now faced with the possibility of omega-3 fatty acid deficiency and fatty acid imbalances because of manipulation of fatty acids in the food supply, not only in fats, oils, and processed foods, but also in meats, poultry, and eggs.

Dietary Omega-3 and Omega-6 Fatty Acids and the Implications for Omega-3 Fatty Acids Metabolism and Requirements

The relationship between dietary omega-3 fatty acids and tissue omega-3 fatty acids is complex and depends on how much and which omega-3 fatty acids are consumed, the concurrent intake of omega-6 fatty acids and several other modifying factors, such as disease and genotype. Major questions with respect to diet center on two points: whether humans with low intakes of EPA and DHA are able to form adequate EPA and DHA from ALA, and whether high intakes of LA interfere with synthesis of EPA and DHA from ALA, or dietary EPA and DHA utilization.

Currently, the most well-accepted pathway for EPA and DHA synthesis involves the sequential actions of Δ-6 and Δ-5 desaturase, with several elongases.[9,36] The Δ-6 desaturase is considered rate-limiting and acts both at the initial desaturation of ALA and at the third desaturation in which 24:5 omega-3 is converted to 24:6 omega-3 before chain shortening to DHA. The same Δ-6 desaturase is required for desaturation of omega-6 LA, both at the initial and third desaturation in which 22:4 omega-6 is desaturated for synthesis of 22:5 omega-6.[9] Several important features characterize omega-3 and omega-6 fatty acid desaturation and these include regulation by Δ-6 desaturase activity, not the availability of fatty acid substrate, the low amounts of omega-6 and omega-3 fatty acid needed to achieve maximal enzyme activity (saturation), and the apparent use of a single enzyme for four potential desaturation reactions; the initial and third desaturation of both LA and ALA.[9] The fact that Δ-6 desaturase requires as little as 1% dietary energy or less from LA plus ALA for maximum activity, and that omega-6 and omega-3 fatty acids compete for the enzyme and influence each other's metabolism[37–41] is a key point when considering modern diets with LA well in excess the enzyme's needs.

The need for de novo synthesis of ARA from LA, or EPA and DHA from ALA in adults in energy balance is expected to be equal to the irreversible loss of ARA, EPA, or DHA because of oxidation; further metabolism (e.g., to eicosanoids); or excretion; such as in sloughed cells. ARA and DHA in contrast to their 18-carbon chain precursors are generally considered poor substrates for β-oxidation.[42–44] Studies published in 1967 using [^{14}C] LA as a tracer in diets with 4% or 18% energy as LA concluded that the low amounts of LA converted to ARA indicated replacement of quantitatively small, irreversible ARA turnover.[45] More recent studies using stable isotope tracers confirm that only a small proportion, about 1%, of a tracer dose of LA appears in plasma ARA, with similar low amounts of labeled ALA appearing in plasma EPA and DHA in adults and children.[46–48] Interpretation is complicated when the amounts of EPA, DHA, and ARA actually needed are not known. More importantly, several lines of evidence are consistent with saturation of Δ-6 desaturase in humans following usual westernized diets. Saturation of enzyme activity rather than low activity is a key point to addressing the question of whether much lower LA and total LA plus ALA would better enable desaturase activity through the terminal steps of the pathway leading to DHA.

Numerous studies have shown that increasing dietary LA leads to a progressive increase in LA in plasma and tissue lipids, including blood cells, adipose, heart, and other tissue lipids in humans.[49–54] The increase in LA in blood and adipose lipids with increasing dietary LA intake is often exploited to validate measures of dietary LA intake, or monitor compliance to changes in LA intake.[55–59] More importantly, increasing LA over the range of 2 to 10% dietary energy does not increase ARA in blood lipids in humans, although as noted, it does increase LA.[49–52,60,61] Minor allele variants in FADS1 and FADS2, which encode Δ-5 and Δ-6 desaturase, respectively, are also associated with higher blood lipid LA and lower ARA,[62–64] a result possible only if desaturase activity, not LA availability, controls conversion to ARA in healthy individuals.

Direct experimental evidence for saturation of Δ-6 desaturase in humans is difficult to address experimentally because of the high body burden of LA in adipose tissues and challenges of preparing diets with less than 2.5% energy from LA but adequate protein, a problem related to the high omega-6 fatty acids in meats, poultry, eggs, and soy products.[49] Studies in pigs, considered a good model of human lipid metabolism, however, showed that an evolutionary diet with 1.2% energy from LA and 1.1% energy ALA supported much higher EPA and DHA, but lower ARA in tissue lipids than a diet with the same amount of ALA but 10.7% energy from LA,[65,66] similar to the upper end of the current recommended intake for PUFA.[33] In these studies, the total LA plus ALA intake was low, below 2.5% dietary energy, addressing the need to avoid inhibition of the second Δ-6 desaturase step leading from EPA to DHA.[9] With the exception of the brain, tissue, and blood levels of 22:5 omega 6 remained low at <0.25% fatty acids even in the liver of pigs fed 10.7% energy from LA and with low tissue DHA.[65] These results show the expected increase in ALA desaturation when dietary LA was low, as expected because of competition between LA and ALA for Δ-6 desaturase, as well as inhibition at the second for Δ-6 desaturation when LA intake was high. This is consistent with studies in humans to show that increasing dietary ALA or decreasing LA results in an increase in blood lipid EPA, but not DHA.[49,50,52]

The implications of the increase in LA in blood and tissue lipids with increasing dietary LA intake[49–54] also need to be

considered. The omega-3 and omega-6 fatty acids are typically esterified at the sn-2 (center) position of the glycerol backbone of phospholipids, raising the possibility of competition for acylation. Acylation of EPA into phospholipids compared to triglycerides seems to differ with the amount of LA in the diet. Studies in animals suggest that high dietary LA may interfere with EPA and DHA accretion not simply through competition and inhibition of desaturation, but also by blocking available acylation sites in tissue and blood phospholipids.[42–44] Consistent with this, LA shows a strong inverse relationship with ARA and EPA in human plasma and RBC phospholipids.[49,61,67] Numerous observation and intervention studies provide compelling evidence that higher intakes of EPA and DHA lead to higher EPA and DHA in plasma and tissue lipids.[1,68–72] The competitive interactions of EPA and DHA with LA is also evident from the decrease not only in ARA, but also in LA in serum or plasma phospholipids of adults given fish oils or purified EPA and DHA.[71,72]

DISCUSSION

The possibility that western diets poor in omega-3 fatty acids and rich in omega-6 fatty acids contribute to chronic cardiometabolic and inflammatory diseases and mental health problems is becoming increasing questioned. An increase in omega-6 and decrease in omega-3 fatty acids in human diets has occurred during the last century because of the development of seed oil production, changes in food production, and animal husbandry and dietary practices. At the same time, and increase in the dietary LA to saturated fatty acid ratio has occurred, but the implications of this, if any, for n-3 fatty acid metabolism and requirements is unknown. Emphasis has been placed on insufficient activity of the Δ-6 and Δ-5 desaturase enzymes to convert ALA to EPA and DHA. However, LA intakes now exceed needs of the fatty acid desaturases, and LA accumulates in blood and tissue lipids with increasing intake. This in turn may lead to competition and inhibition of omega-3 fatty acid metabolism and omega-6 fatty acid retention in tissue lipids. Increasing dietary EPA and DHA effectively increases EPA and DHA in blood and tissue, although it does not address potential underlying issues of omega-6 and omega-3 fatty acid imbalance in the modern food supply.

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