Prostaglandins, Leukotrienes and Essential Fatty Acids xxx (xxxx) xxx–xxx

Contents lists available at ScienceDirect

Prostaglandins, Leukotrienes and Essential Fatty Acids

journal homepage: www.elsevier.com/locate/plefa

The Omega-6:Omega-3 ratio: A critical appraisal and possible successor

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ARTICLE INFO

Keywords:
Omega-3 fatty acids
Omega-6 fatty acids
Eicosapentaenoic acid
Docosahexaenoic acid
Linoleic acid
Arachidonic acid
Biomarkers
Omega-3 index

ABSTRACT

The well-known health effects of the long-chain, marine omega-3 (n-3) fatty acids (FAs) has led to a growing interest in the prognostic value that blood levels of these FAs might have vis-à-vis cardiovascular and neurocognitive diseases. The measurement and expression of n-3 FA levels is not straight-forward, however, and a wide variety of means of expression of n-3 FA status have been used in research and clinical medicine. This has led to considerable confusion as to what "optimal" n-3 FA status is. The n-6:n-3 ratio has enjoyed relatively widespread use, but this apparently simple metric has both theoretical and practical difficulties that have contributed to misunderstandings in this field. Just as the once-popular polyunsaturated:saturated FA ratio has largely disappeared from the nutritional and medical literature, it may be time to replace the n-6:n-3 ratio with a newer metric that focuses on the primary deficiency in Western diets – the lack of eicosapentaenoic and docosahexaenoic acids (EPA and DHA). The Omega-3 Index (red blood cell EPA + DHA) has much to recommend it in this regard.

1. Introduction

There is a growing interest in exploring the relationships between fatty acid (FA) status and clinically important health outcomes [1]. These include cardiac disease [2–4], stroke [5], diabetes [6], cognitive function [7], and aging [8–10]. However, analysis of FAs is much more complicated than it is for other biomarkers like cholesterol or glucose. The latter analytes circulate in plasma as single molecular species whose concentrations can be easily measured by long-ago standardized enzymatic methods, and optimal levels are clearly defined after decades of research, either as a risk factor for a disease [e.g., coronary heart disease (CHD), the former] or as a diagnostic for disease (e.g., diabetes, the latter). There are several reasons why FA testing is more challenging. First, there are many different FA species, typically organized into groups based on the *number of double bonds* they contain [0, saturated; 1, monounsaturated; >1, polyunsaturated (PUFA); or >2, highly unsaturated (HUFA)]. FAs within the latter 2 groups are further segregated based on the *position of the terminal double bond* into the omega (n)-6 and n-3 groups. But even these are not homogeneous groupings as the physiological functions of FAs within each class may differ depending on *carbon chain length* and *orientation* of the double bonds (*cis* vs. *trans*). Beyond the differences in molecular species, FA status can be measured in *multiple lipid pools* – from whole blood to blood cells (red, white or platelet) to whole plasma or plasma lipid classes or even subclasses. In general, the same FAs are found in all lipid pools but always in unique relative proportions peculiar to that pool [11]. Like cholesterol and glucose, FA levels can be expressed in molar or mass units. Finally, regardless of the pool analyzed, FA status can be expressed as *composition* (each as a percent of total) or as *concentration* (mass/volume or cell count). Partly because of these challenges, defining a "high risk" FA level that can be used clinically has been difficult.

In the 1960s the "P:S ratio" became popular as the ratio of dietary/plasma polyunsaturated vs. saturated FAs was inversely related to serum cholesterol levels [12]. This metric became obsolete as Mensink et al. demonstrated the illogic of its underlying assumptions [13]. These included physiologic differences between two FAs from the same class (e.g., one saturated FA – palmitic – raised cholesterol but another – stearic – did not), and physiological similarities between two FAs from different classes (e.g., *trans* monounsaturated FAs proved to have even more detrimental effects on serum lipids than saturated FAs). What's more, *cis* monounsaturated FAs had clear beneficial effects on a classic CHD risk marker – the ratio of total to high density lipoprotein cholesterol – and they were not included in the P:S ratio. The confluence of these advances in science led to the eventual demise of the P:S ratio.

The discovery in the late 1970s of the potential health benefits of the marine n-3 FAs (eicosapentaenoic and docosahexaenoic acids, EPA and DHA) by Bang and Dyerberg in Greenland Inuits [14] sparked an avalanche of studies on these novel FAs. The realization that EPA and arachidonic acid (AA, n-6) competed as substrates for several enzymes critical to hemostasis, vascular reactivity, and inflammation suggested that some kind of ratio of the n-6 to n-3 FAs in both the diet and the

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https://doi.org/10.1016/j.plefa.2018.03.003
Received 7 March 2018; Accepted 9 March 2018

blood might be a convenient way to conceptualize the overall "omega" status. This comported well with the pioneering studies of Holman and colleagues showing that the shorter chain n-6 and n-3 FAs (linoleic and alpha-linolenic acids, LA and ALA) also competed with each other for desaturase and elongase enzymes in the production of AA and EPA/DHA. These findings further supported a "competition" mindset and provided a conceptual underpinning for the n-6:n-3 ratio. The discovery that aspirin blocked the cyclo-oxygenase mediated conversion of AA to a variety of pro-inflammatory eicosanoids fed into the idea that AA was itself pro-inflammatory, and soon, ALL n-6 FAs were painted with that brush. So the n-6 PUFAs became "bad" and the n-3 PUFAs "good," and a ratio seemed a reasonable way to simply represent the body's potential inflammatory response to an insult [15]. However, in recent years, the complex biochemistry of the eicosanoids (and docosanoids and octadecanoids) has become clearer, with some n-6 FA metabolites being pro- and others anti-inflammatory [16], so the class itself can no longer be so simply regarded as pro-inflammatory. In addition, beneficial (not detrimental) effects of the primary dietary n-6 FA, LA, have been repeatedly observed [17–19]. These new findings began to erode the view that PUFA biology could be summed up in one simple ratio. Now, based on a further appreciation of both its conceptual problems and flawed assumptions [20], calls to abandon the use of this ratio have become more frequent [20–23]. These concerns are discussed below, and an alternative approach to expressing PUFA status is suggested.

2. The n-6:n-3 ratio is imprecise and non-specific

> *"What is the usefulness of the ratio of n-6 to n-3, which is good divided by good?"* [23]

The components of the n-6:n-3 ratio are rarely defined. The ratio is formed by summing all of the n-6 FAs in either the diet or a biological sample and dividing it by the sum of all the n-3 FAs. As simple as this sounds, even here we find ambiguity since "all" depends on how many FAs are actually quantified in a given study. The major n-6 FA in the diet and plasma is LA followed by AA. There are trace amounts of gamma-linolenic (GLA) in the diet, but its blood levels are very low. Then there are a variety of other n-6 metabolites not in the diet, but present in the blood, again at low levels. These would include dihomo-gamma linolenic acid (DGLA), adrenic acid (ADA), eicosadienoic acid (EDA) and docosapentaenoic acid (DPAn-6). The major n-3 FA in the western diet is ALA, but the n-3 FA in greatest abundance in blood and most tissues is DHA, followed by DPAn-3 and EPA. Depending on the analytical conditions, greater or fewer individual FA species may be measured. Using RBC data from a random set of 50 individuals measured in our laboratory, if one includes the seven n-6 FAs listed above and the four n-3 FAs, one gets a ratio of 7.8 in RBCs. If instead only the "important" or "major" n-6 FAs (LA and AA) and n-3 FAs (ALA, EPA and DHA) are included, the ratio is 9.3. The lack of a standardized definition of which FAs constitute both the numerator and the denominator of the ratio is an obvious weakness.

The lipid pool in which the ratio is calculated is not defined. Again using data from our laboratory as an example, when the ratio is measured in RBCs and in plasma cholesteryl esters from the same 50 people, the ratio ranges from 3.5% in RBCs to nearly 29% in plasma (Table 1). The ratio in RBCs also differs considerably from that in platelets (2.7 vs. 6.3) [24], and to get even more granular, each different class of phospholipids present in cell membranes has its own characteristic FA composition, and thus n-6:n-3 ratio. RBC phosphatidyl-choline, -ethanolamine, -serine and -inositol have ratios of 12, 2, 2, and 4.5, respectively [25]. A further challenge on this point has to do with measurements made in whole plasma/serum since this matrix contains an undefined mixture of 4 lipid classes (phospholipids, triglycerides, cholesteryl esters, and free FAs) each with its own FA signature [11], and except for the free FAs, these are all carried in unique proportions in 3 different lipoprotein particles (very low-, low- and high-density lipoproteins).

Table 1
Differences in the n-6:n-3 ratio by lipid pool measured in the same 50 random blood samples in the author's laboratory.

Lipid compartment	n-6:n-3 ratio (wt%)
Whole blood	7.8
Red blood cell	3.5
Whole plasma/serum	9.1
Non-esterified FAs	4.0
Cholesteryl esters	19.7
Phospholipids	4.9
Triglycerides	6.5

The n-6:n-3 ratios in patients with various dyslipidemias can thus be affected by variations in serum levels of each lipid class [11].

The means of expression of FA abundance is not defined. FA status can be expressed in molar or mass terms (whether as concentration or percent compositions). For example, the RBC ratio is 11.2 based on mol % expression and 10.2 on weight%.

Identical ratios can be calculated from an endless variety of individual FA levels. This weakness, raised earlier [20], can be illustrated by considering a RBC membrane with 30% LA + AA and 8.3% EPA + DHA. (The latter value is called the Omega-3 Index, and a value of 8.3% is within the optimal cardioprotective zone [26,27]). The n-6:n-3 ratio of this sample would be 3.6. Virtually the same ratio would be calculated for a sample containing 18% LA + AA and 5% EPA + DHA (which is near the undesirable zone of < 4% for the Omega-3 Index). Hence, both high and low risk status could have the same ratio. (To be fair, this is only a theoretical concern since in human biology, the RBC membrane PUFA content is held constant, so when the n-3 FA level increases, the n-6 level decreases; they cannot both decrease to any appreciable extent [28]).

Coherent dietary advice cannot be given based only on the n-6:n3 ratio. Based on NHANES data, the average n-6:n-3 ratio of the American diet is about 10 [29]. Some have advocated the consumption of a diet with a ratio of 1 – the presumed ratio of the ancient human diet [30]. Putting aside for a moment the problem of the implicit presumption of metabolic equivalence of each FA within each class (discussed below), there are at least five ways to lower a ratio that is "too high" (Table 2), and the physiological consequences of each approach differs. For example, lowering *both* n-3 and n-6, the latter more than the former (approach 5 in the Table) is clearly less healthy than simply raising the n-3 intake (approach 2). Ratio-thinking distracts from the almost universal need for individuals with a "high" ratio to simply raise their EPA + DHA intake, not lower their n-6 intake. Thus, it can be challenging for a clinician who is "ratio-focused" to make rational and healthy dietary recommendations.

3. Use of the n-6:n-3 ratio is based on invalid assumptions

In addition to problems of imprecision and non-specificity, there are at least four assumptions underlying the use of the n-6:n-3 ratio that are, if not completely false, at least highly debatable. This thin evidentiary foundation contributes to the disutility of the metric.

Assumption 1: Omega-6 FAs have adverse effects on cardiovascular health. More precisely, LA – because it can be converted to AA which can then be metabolized to pro-inflammatory eicosanoids – increases the chronic inflammatory status of the body which predisposes to

Table 2
Five ways to lower the n-6:n-3 ratio [19].

Approach	1	2	3	4	5
n-3 FAs	↑	↑	↑↑	→	↓
n-6 FAs	↓	→	↑	↓	↓↓
Ratio	↓	↓	↓	↓	↓