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The role of dietary n-6 fatty acids in the prevention of cardiovascular disease

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n-6 Fatty acids, like n-3 fatty acids, play essential roles in many biological functions. Because n-6 fatty acids are the precursors of proinflammatory eicosanoids, higher intakes have been suggested to be detrimental, and the ratio of n-6 to n-3 fatty acids has been suggested by some to be particularly important. However, this hypothesis is based on minimal evidence, and in humans higher intakes of n-6 fatty acids have not been associated with elevated levels of inflammatory markers.

n-6 Fatty acids have long been known to reduce serum total and low-density lipoprotein cholesterol, and increases in polyunsaturated fat intake, mostly as n-6 fatty acids, were a cornerstone of dietary advice during the 1960s and 1970s. In the United States, for example, intake of n-6 fatty acids doubled and coronary heart disease (CHD) mortality fell by 50% over a period of several decades. In a series of relatively small, older randomized trials, in which intakes of polyunsaturated fat were increased (even up to 20% of calories), rates of CHD were generally reduced. In a more recent detailed examination of fatty acid intake within the Nurses' Health Study, greater intake of linoleic acid, up to about 8% of energy, has been strongly related to lower incidence of myocardial infarction or CHD death. Because n-3 fatty acids were also related inversely to risk of CHD, the ratio was unrelated to risk. n-6 Fatty acids reduce insulin resistance, probably by acting as a ligand for peroxisome proliferator-activated receptors-γ, and intakes have been inversely related to risk of type 2 diabetes.

Adequate intakes of both n-6 and n-3 fatty acids are essential for good health and low rates of cardiovascular disease and type 2 diabetes, but the ratio of these fatty acids is not useful. Reductions of linoleic acid to "improve" this ratio would likely increase rates of cardiovascular disease and diabetes. *J Cardiovasc Med* 8 (suppl 1):S42–S45

Journal of Cardiovascular Medicine 2007, **8 (suppl 1)**:S42–S45

Keywords: coronary heart disease, diet, dietary fats, linoleic acid, n-6 fatty acids, polyunsaturated fatty acids

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The author reports no conflicts of interest.

Dietary n-6 fatty acids in the prevention of cardiovascular disease

Alpha-linolenic acid (ALA, the primary 18-carbon member of the n-3 series) and linoleic acid (LA, the primary 18-carbon member of the n-6 series) are the most abundant essential dietary fatty acids. These fatty acids can be desaturated and elongated through common enzymatic pathways to form arachidonic acid (a 20-carbon n-6 fatty acid derived from LA) and eicosapentaenoic and docosahexaenoic acids (20- and 22-carbon n-3 fatty acids derived from ALA). Through cyclooxygenase, arachidonic acid can be converted to prothrombotic and proinflammatory prostaglandins, and long-chain n-3 fatty acids can be converted to antithrombotic anti-inflammatory eicosanoids. By competition in these parallel pathways, greater intake of LA could potentially inhibit the conversion of ALA to the long-chain n-3 fatty acids, and higher concentrations of arachidonic acid could favor the production of proinflammatory molecules. Because of this potential competition, some have believed that higher dietary intake of LA is proinflammatory and increases the risk of cardiovascular disease, and that the dietary ratio of n-6 to n-3 fatty acids is an important risk factor for coronary heart disease (CHD) risk [1].

The belief that LA intake adversely affects the risk of heart disease derives primarily from this hypothesized mechanism of competition, which is partly based on the assumption that dietary LA substantially increases blood and tissue levels of arachidonic acid. However, because levels of arachidonic acid are under close homeostatic regulation, changes in dietary LA within the usual dietary range do not appreciably alter arachidonic acid levels [2,3].

Furthermore, dietary factors can influence risk of heart disease through many different pathways. These include effects on blood lipid fractions, blood pressure, thrombotic tendency, insulin resistance, oxidative stress, endothelial function, and the likelihood of ventricular arrhythmias. Because LA has multiple physiological influences unrelated to the arachidonic acid pathway, higher intake of LA would not necessarily increase CHD risk (even if there were some increase in inflammatory factors) if its effects on

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other pathways favored a net reduction in risk. The net effect of any specific dietary factor on risk of heart disease is the result of the balance of positive and negative influences on all of the pathways related to cardiovascular disease, possibly including additional mechanisms that are not fully appreciated at this time.

Effects of linoleic acid on blood lipids

The role of polyunsaturated fat (which is primarily LA) in reducing blood total cholesterol levels became well recognized in the 1960s. This effect was documented in dozens of controlled feeding studies that were summarized by the Keys and Hegsted equations [4,5]. These equations demonstrated nearly identical results, with saturated fat positively related to serum cholesterol and polyunsaturated fat inversely related to serum cholesterol. Primarily on the basis of the Keys and Hegsted equations, dietary recommendations in the United States and other western countries between the 1960s and early 1980s included advice to replace saturated fat with polyunsaturated fat, because this would have a double benefit in reducing total serum cholesterol. As a result of these recommendations, consumption of polyunsaturated fat (primarily LA) in the United States increased from ~3% of energy in the 1950s to ~6–7% of energy by the late 1980s [6,7]. This dietary change was confirmed by large increases in the LA content of adipose samples; concentrations increased from about 8.5% in 1962 and about 10% in 1966 [8] to 17% to 19% in the 1980s [9–11].

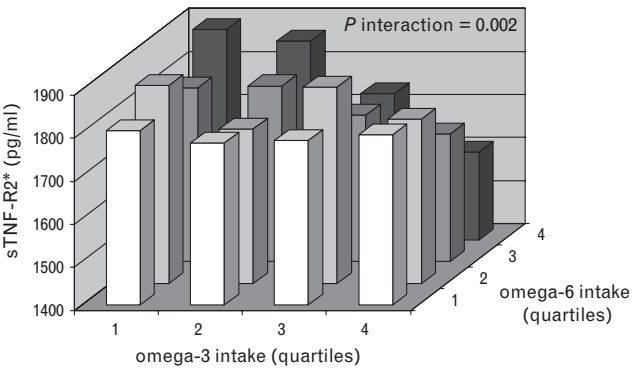
More recently, using data from 61 controlled feeding studies, Mensink *et al.* [12] assessed the effects of dietary fats on not only blood total cholesterol, but also low-density lipoprotein (LDL) cholesterol, high-density lipoprotein (HDL) cholesterol, and triglycerides. This analysis demonstrated that compared to carbohydrate, polyunsaturated fat reduces LDL cholesterol, increases HDL cholesterol, and reduces triglycerides [12]; each of these effects favors lower risk of CHD. Indeed, compared to all other classes of fatty acids, LA produces the most favorable lipid changes, as reflected by the lowest ratio of LDL to HDL cholesterol.

Effects on inflammatory factors

Several lines of evidence indicate that n-6 fatty acids have anti-inflammatory effects, mediated by pathways that do not involve cyclooxygenase [13]. Both n-3 and n-6 fatty acids inhibit the production of inflammatory factors in endothelial cells by down-regulating nuclear factor-κB, a nuclear transcription factor considered the master regulator of the inflammatory response in vascular cells; this effect appears to be mediated by multiple mechanisms [14,15].

Relatively few studies have actually examined in humans the effects of both n-3 and n-6 fatty acid intake on inflammatory factors. In a cross-sectional analysis among

Fig. 1



Association between different amounts of omega-3 and omega-6 polyunsaturated fatty acid consumption and systemic inflammation as measured by levels of soluble tumor necrosis factor receptor-2 (sTNF-R2). *Mean levels adjusted for age, gender, smoking status, physical activity, alcohol use, nonsteroidal anti-inflammatory medication use, body mass index, energy intake, and consumption of protein, saturated fat, monounsaturated fat, and cholesterol ($n = 859$) [16].

859 men and women, Pischon *et al.* [16] cross-classified individuals by consumption of both n-3 and n-6 fatty acids and measured plasma levels of soluble tumor necrosis factor (TNF) receptors, a stable indicator of TNF activity. The lowest levels of soluble TNF receptor-2 were among persons with the highest intakes of both n-6 and n-3 fatty acids (Fig. 1). Similar findings were seen in relation to soluble TNF receptor-1 levels. Reduction in TNF-α by n-6 and n-3 fatty acids is likely a consequence of their inhibiting the activation of nuclear factor-κB. In a randomized trial among healthy men comparing 4% to 10% of energy consumption from LA, there was no effect on plasma levels of C-reactive protein or interleukin-6 [17]. Thus, in contrast to hypothesized increases in inflammation, consumption of n-6 fatty acids does not appear to increase inflammatory factors, and may actually reduce some indicators of systemic inflammation.

Effects on insulin resistance and risk of type 2 diabetes

In controlled feeding studies, increases in polyunsaturated fat (primarily LA) consumption improves insulin resistance [18,19]. Consistent with this, intake of LA has been inversely related to incidence of type 2 diabetes in large prospective studies [19]. The most detailed examination of LA intake and risk of type 2 diabetes was conducted by Salmeron *et al.* [20] within the Nurses' Health Study cohort. In this analysis, women with the highest intake of LA had a 25% lower risk of type 2 diabetes, compared to those with the lowest intake ($P = 0.0002$). In rats, LA, consumed as corn oil, reduced insulin resistance and fasting insulin levels when compared to monounsaturated or saturated fat [21], although the molecular mechanism was not clear. Thus, the overall evidence strongly suggests that LA consumption

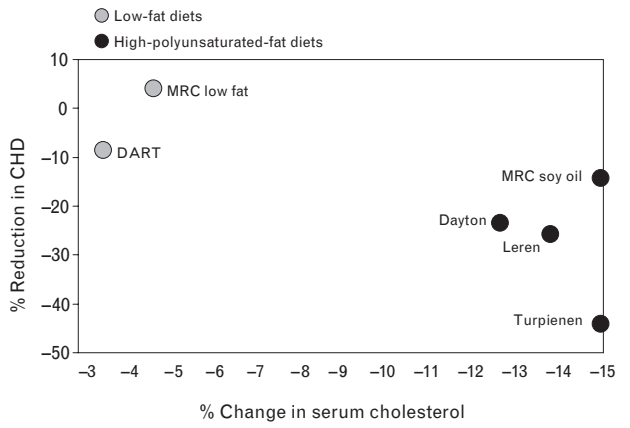
improves insulin resistance and decreases the incidence of type 2 diabetes.

Risk of coronary heart disease

Although each of the studies of the individual metabolic effects of n-6 fatty acids described above suggest that LA consumption would lower CHD risk, the best test of the balance of risks and benefits due to intake of n-6 fatty acids is their relation to risk of CHD events. As described, average consumption of LA greatly increased from the 1960s through the 1980s in the United States, and in the United Kingdom and Australia somewhat later. During this time period, CHD mortality declined by nearly 50% in both men and women. In a time trend analysis, Dwyer and Hetzel [22] concluded that increased intake of polyunsaturated fats, mainly LA, was probably the primary factor underlying these large decreases in CHD mortality, which have resulted in several additional years of life expectancy in these countries.

Also during this period, several intervention trials were conducted using high intakes of polyunsaturated fat in the prevention of CHD [6,23,24]. These trials used either corn oil or soybean oil, both of which contain large amounts of LA. Although soybean oil also contains about 7% ALA, the ratio of n-6 to n-3 fatty acids is about 10 : 1. In four different trials in which n-6 consumption was increased (British Medical Research Council Soy Oil Study [25], the Dayton study of Veterans [26] in the United States, the study by Leren [27] in Norway, and the Turpeinen study [28] in Scandinavia), serum total cholesterol declined by 12–15%. Most importantly, incidence of CHD was reduced by 10–45% (Fig. 2). Notably, in the Dayton Study, which documented a benefit in a composite atherosclerotic outcome, the intake of LA in the intervention group was nearly 20% of calories [26].

Fig. 2



Randomized controlled clinical trials evaluating the effect of either a low-fat diet (grey circles) or a high-polyunsaturated-fat diet (black circles) on coronary heart disease (CHD) risk [24].

The relation between total polyunsaturated fat or LA intake and risk of CHD has also been examined in several large prospective studies (summarized in *Nutritional Epidemiology* [6]). Significant inverse associations were seen in five of the 13 studies [29–33], and in no study was a positive association between LA consumption and CHD risk observed.

Detailed analyses of n-6 polyunsaturated fat intake and risk of CHD have been conducted within the Nurses' Health Study cohort [33]. In this study, nearly 90 000 women were followed for 14 years, during which time nearly 1000 women died of CHD or were hospitalized for acute myocardial infarction. Dietary intake was assessed by repeated, validated food frequency questionnaires that focused on specific types of dietary fat. Greater consumption of polyunsaturated fat (an increment of 5% energy replacing equivalent calories from carbohydrate) was associated with a nearly 40% lower risk of CHD. This robust association was stronger than would be expected from effects on LDL and HDL cholesterol alone, suggesting beneficial effects of LA consumption on other physiologic pathways. In the 20-year follow-up of the Nurses' Health Study [34], the strong inverse association persisted. Consumption of LA ranged from approximately 2.5% to 8.5% of total energy (median intake ~5%), and increasing LA consumption was linearly associated with lower risk of CHD, with ~30% lower risk comparing the highest to the lowest level of consumption ($P < 0.01$) and no suggestion of higher risk at higher intakes. In a further analysis within this cohort, Hu *et al.* [35] examined ALA and the ratio of ALA to LA intake in relation to risk of fatal CHD. Both ALA and LA consumption were associated with lower incidence of fatal CHD. Thus, the ratio of ALA to LA was not associated with risk of fatal CHD, because both the numerator and denominator were beneficial. In a recent analysis within a large cohort of men, higher intake of LA also did not reduce the strong inverse relation between consumption of marine n-3 fatty acids and risk of sudden death [36].

Conclusion

Both n-3 and n-6 fatty acids are essential, and greater intakes of both are related to lower risk of CHD. Thus, the ratio of n-6 to n-3 fatty acids is not a useful concept. Further, at any ratio, both n-6 and n-3 fatty acid intake could be deficient, and for this reason they are better considered individually rather than as a ratio. Because increases in LA consumption in US and some other populations have been achieved for the prevention of CHD, insufficiency of n-3 fatty acids is much more common than inadequate intake of n-6 fatty acids. However, reduction in n-6 fatty acid intake to 'improve' the n-6 : n-3 ratio is likely to increase incidence and mortality rates of CHD and type 2 diabetes and potentially reverse the major reductions in cardiovascular

mortality that have been achieved in the last 40 years. The optimal intake and the safe upper limit of dietary LA are not clear, but intake up to ~10% of energy appears to be progressively beneficial and also safe. Intakes above this level should be evaluated in more detail with regard to incidence of heart disease and other long-term outcomes.

Acknowledgments

The author acknowledges the helpful suggestions of Frank Sacks and assistance of Hilary Farmer in the preparation of this manuscript.

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