Review Article

The omega-6/omega-3 fatty acid ratio, genetic variation, and cardiovascular disease

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A high omega-6/omega-3 ratio, as is found in today's Western diets, promotes the pathogenesis of many chronic diseases, including cardiovascular disease. Increased dietary intake of linoleic acid (LA) leads to oxidation of low-density lipoprotein (LDL), platelet aggregation, and interferes with the incorporation of essential fatty acids (EFA) in cell membrane phopholipids. Both omega-6 and omega-3 fatty acids influence gene expression. Omega-3 fatty acids have strong anti-inflammatory effects, suppress interleukin 1β (IL-1β), tumor necrosis factor-α (TNFα) and interleukin-6 (IL-6), whereas omega-6 fatty acids tend to be pro-inflammatory. Because inflammation is at the base of many chronic diseases, including coronary heart disease, dietary intake of omega-3 fatty acids plays an important role in the manifestation of disease, particularly in persons with genetic variation, as for example in individuals with genetic variants at the 5-lipoxygenase (5-LO). Increased dietary arachidonic acid (AA) significantly enhances the apparent atherogenic effect of genotype, whereas increased dietary intake of omega-3 fatty acids eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) blunts this effect. The diet-gene interaction further suggests that dietary omega-6 fatty acids promote, whereas marine omega-3 fatty acids EPA and DHA inhibit leukotriene-mediated inflammation that leads to atherosclerosis in this subpopulation.

Key Words: omega-6/omega-3 ratio, genetic variation, gene expression, inflammation, cardiovascular disease

INTRODUCTION

The interaction of genetics and environment, nature, and nurture is the foundation for all health and disease. In the last two decades, using the techniques of molecular biology, it has been shown that genetic factors determine susceptibility to disease and environmental factors determine which genetically susceptible individuals will be affected.[1-6] Nutrition is an environmental factor of major importance. Using the tools of molecular biology and genetics, research is defining the mechanisms by which genes influence nutrient absorption, metabolism and excretion, taste perception, and degree of satiation; and the mechanisms by which nutrients influence gene expression. Whereas major changes have taken place in our diet over the past 10,000 years since the beginning of the Agricultural Revolution, our genes have not changed. The spontaneous mutation rate for nuclear DNA is estimated at 0.5% per million years. Therefore, over the past 10,000 years there has been time for very little change in our genes, perhaps 0.005%. In fact, our genes today are very similar to the genes of our ancestors during the Paleolithic period 40,000 years ago, at which time our genetic profile was established.[7] Genetically speaking, humans today live in a nutritional environment that differs from that for which our genetic constitution was selected. Studies on the evolutionary aspects of diet indicate that major changes have taken place in our diet, particularly in the type and amount of essential fatty acids and in the antioxidant content of foods.[7-18]

The beneficial health effects of omega-3 fatty acids, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) were described first in the Greenland Eskimos who consumed a high seafood diet and had low rates of coronary heart disease, asthma, type 1 diabetes mellitus, and multiple sclerosis. Since that observation, the beneficial health effects of omega-3 fatty acids have been extended to include benefits related to cancer, inflammatory bowel disease, rheumatoid arthritis, and psoriasis.[19-22]

Imbalance of omega-6/omega-3 fatty acids

Food technology and agribusiness provided the economic stimulus that dominated the changes in the food supply.[23-25] It has been estimated that the present Western diet is "deficient" in omega-3 fatty acids with a ratio of omega-6 to omega-3 of 15-20/1, instead of 1/1 as is the case with wild animals and presumably human beings.[7-11, 13, 26-28]

An absolute and relative change of omega-6/omega-3 in the food supply of Western societies has occurred over the last 150 years. A balance existed between omega-6 and omega-3 for millions of years during the long evolutionary history of the genus Homo, and genetic changes occurred partly in response to these dietary influences. During evolution, omega-3 fatty acids were found in all foods consumed: meat, wild plants, eggs, fish, nuts and berries.[29-38]

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Because of the increased amounts of omega-6 fatty acids in the Western diet, the eicosanoid metabolic products from AA, specifically prostaglandins, thromboxanes, leukotrienes, hydroxy fatty acids, and lipoxins, are formed in larger quantities than those formed from omega-3 fatty acids, specifically EPA.[8] The eicosanoids from AA are biologically active in very small quantities and, if they are formed in large amounts, they contribute to the formation of thrombus and atheromas; to allergic and inflammatory disorders, particularly in susceptible people; and to proliferation of cells.

Thus, a diet rich in omega-6 fatty acids shifts the physiological state to one that is prothrombotic and proaggregatory, with increases in blood viscosity, vasospasm, and vasocontriction and decreases in bleeding time. Bleeding time is decreased in groups of patients with hypercholesterolemia, hyperlipoproteinemia, myocardial infarction, other forms of atherosclerotic disease, and diabetes (obesity and hypertriglyceridemia). Bleeding time is longer in women than in men and longer in young than in old people. There are ethnic differences in bleeding time that appear to be related to diet.

The balance of omega-6/omega-3 fatty acids is important for health: the evidence from gene transfer studies

Further support for the need to balance the omega-6/omega-3 EFA comes from the studies of Kang *et al*.,[39-44] which clearly show the ability of both normal rat cardiomyocytes and human breast cancer cells in culture to form all the omega-3's from omega-6 fatty acids when fed the cDNA encoding omega-3 fatty acid desaturase obtained from the roundworm *Caenorhabditis elegans* (*C. elegans*). The omega-3 desaturase efficiently and quickly converted the omega-6 fatty acids that were fed to the cardiomyocytes in culture to the corresponding omega-3 fatty acids. Thus, omega-6 LA was converted to omega-3 ALA and AA was converted to EPA, so that at equilibrium, the ratio of omega-6 to omega-3 PUFA was close to 1/1. Further studies demonstrated that the cancer cells expressing the omega-3 desaturase underwent apoptotic death whereas the control cancer cells with a high omega-6/omega-3 ratio continued to proliferate.[41] More recently, Kang, *et al*. showed that transgenic mice and pigs expressing the *C. elegans fat-1* gene encoding an omega-3 fatty acid desaturase are capable of producing omega-3 from omega-6 fatty acids, leading to enrichment of omega-3 fatty acids with reduced levels of omega-6 fatty acids in almost all organs and tissues, including muscles and milk, with no need of dietary omega-3 fatty acid supply.[42-44] This discovery provides a unique tool and new opportunities for omega-3 research, and raises the potential of production of *fat-1* transgenic livestock as a new and ideal source of omega-3 fatty acids to meet the human nutritional needs.

Omega-3 fatty acids and gene expression

Previous studies have shown that fatty acids released from membrane phospholipids by cellular phospholipases, or made available to the cell from the diet or other aspects of the extracellular environment, are important cell signaling molecules. They can act as second messengers or substitute for the classical second messengers of the inositide phospholipid and the cyclic AMP signal transduction pathways. They can also act as modulator molecules mediating responses of the cell to extracellular signals. Recently it has been shown that fatty acids rapidly and directly alter the transcription of specific genes.[45] In the case of genes involved in inflammation, such as IL-1β, EPA and DHA suppress IL-1β mRNA whereas AA does not, and the same effect appears in studies on growth-related early response gene expression and growth factor.[45] In the case of vascular cell adhesion molecule (VCAM), AA has a modest suppressing effect relative to DHA. The latter situation may explain the protective effect of fish oil toward colonic carcinogenesis, since EPA and DHA did not stimulate protein kinase C. PUFA regulation of gene expression extends beyond the liver and includes genes such as adipocyte glucose transporter-4, lymphocyte stearoyl-CoA desaturase 2 in the brain, peripheral monocytes (IL-1β, and VCAM-1) and platelets [platelet derived growth factor (PDGF)]. Whereas some of the transcriptional effects of PUFA appear to be mediated by eicosanoids, the PUFA suppression of lipogenic and glycolytic genes is independent of eicosanoid synthesis, and appears to involve a nuclear mechanism directly modified by PUFA.

Linoleic acid and arachidonic acid increase atherogenesis: evidence from diet-gene interactions: genetic variation and omega-6 and omega-3 fatty acid intake in the risk for cardiovascular disease

Leukotrienes are inflammatory mediators generated from AA by the enzyme 5-lipoxygenase. Since atherosclerosis involves arterial inflammation, Dwyer *et al*. hypothesized that a polymorphism in the 5-lipoxygenase gene promoter could relate to atherosclerosis in humans, and that this effect could interact with the dietary intake of competing 5-lipoxygenase substrates.[46] The study consisted of 470 healthy middle-aged women and men from the Los Angeles Atherosclerosis study, randomly sampled. The investigators determined 5-lipoxygenase (5-LO) genotypes, carotid-artery intima thickness, markers of inflammation, CRP, IL-6, dietary AA, EPA, DHA, LA, and ALA with the use of six 24-hour recalls of food intake. The results showed that 5-LO variant genotypes were found in 6.0 percent of the cohort. Mean intima-media thickness adjusted for age, sex, height and racial or ethnic group was increased by 80 ± 19 μm from among the carriers of two variant alleles as compared with the carrier of the common (wild-type) allele. In multivariate analysis, the increase in intima-media thickness among carriers of two variant alleles (62 μm, $p < 0.001$) was similar in this cohort to that associated with diabetes (64 μm, $p < 0.01$) the strongest common cardiovascular risk factor. Increased dietary AA significantly enhanced the apparent atherogenic effect of genotype, whereas increased dietary intake of omega-3 fatty acids EPA and DHA blunted this effect. Furthermore, the plasma level of CRP of two variant alleles was increased by a factor of 2, as compared with that among carriers of the common allele. Thus, genetic variation of 5-LO identifies a subpopulation with increased risk for atherosclerosis. The diet-gene interaction further suggests that dietary omega-6 fatty acids promote, whereas marine omega-3 fatty acids EPA and DHA

inhibit leukotriene-mediated inflammation that leads to atherosclerosis in this subpopulation.

The prevalence of variant genotypes did differ across racial and ethnic groups with higher prevalence among Asians or Pacific Islanders (19.4%), blacks (24.0 percent) and other racial or ethnic groups (18.2 percent) than among Hispanic subjects (3.6 percent) and non-Hispanic whites (3.1 percent). Increased intima-mediated thickness was significantly associated with intake of both AA and LA among carriers of the two variant alleles, but not among carriers of the common alleles. In contrast, the intake of marine omega-3 fatty acids was significantly and inversely associated with intima-media thickness only among carriers of the two variant alleles. Diet-gene interactions were specific to these fatty acids and were not observed for dietary intake of monounsaturated, saturated fat, or other measured fatty acids. The study constitutes evidence that genetic variation in an inflammatory pathway – in this case the leukotriene pathway, can trigger atherogenesis in humans. These findings could lead to new dietary and targeted molecular approaches for the prevention and treatment of cardiovascular disease according to genotype, particularly in the populations of non-European descent.[6]

CONCLUSIONS AND RECOMMENDATIONS

- Western diets are characterized by high omega-6 and low omega-3 fatty acid intake. Genetically speaking, humans today live in a nutritional environment that differs from that for which our genetic constitution was selected.
- The balance of omega-6/omega-3 fatty acids is an important determinant in decreasing the risk for coronary heart disease, both in the primary and secondary prevention of coronary heart disease.
- Increased dietary intake of LA leads to oxidation of LDL, platelet aggregation, and interferes with the incorporation of EPA in cell membrane phospholipids.
- Both omega-6 and omega-3 fatty acids influence gene expression. EPA and DHA have the most potent anti-inflammatory effects. Inflammation is at the base of many chronic diseases, including coronary heart disease, diabetes, arthritis, cancer, osteoporosis, mental health, dry eye disease and age-related macular degeneration. Dietary intake of omega-3 fatty acids may prevent the development of disease, particularly in persons with genetic variation, as for example in individuals with genetic variants at the 5-LO and the development of coronary heart disease.
- Chronic diseases are multigenic and multifactorial. It is quite possible that the therapeutic dose of omega-3 fatty acids will depend on the degree or severity of disease resulting from the genetic predisposition.
- In carrying out clinical intervention trials, it is essential to increase the omega-3 and decrease the omega-6 fatty acid intake in order to have a balanced omega-6 and omega-3 intake in the background diet. Both the dietary intake and plasma levels should be determined before and after the intervention study.

AUTHOR DISCLOSURES

Artemis P Simopoulos, no conflicts of interest.

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