Omega-3 Polyunsaturated Fatty Acids and Cardiovascular Health

A review of epidemiological, clinical and basic science data

February 25, 2008

Reinhard Grzanna, Ph.D.
RMG Biosciences, Inc.

Introduction

In the United States heart disease is the leading cause of death accounting for one of every 2.8 deaths (American Heart Association, 2005). Preventive measures such as exercise, life-style changes and diets low in cholesterol have been proven to offer protection against cardiovascular disease. Now there is another preventive measure that appears to be just as effective: the consumption of fish and the long-chain omega-3 fatty acids eicosopentaenoic acid (EPA) and docosahexaenoic acid (DHA) in form of fish oil. This review summarizes data from epidemiological and clinical studies of the effects of fish and fish oil consumption on cardiovascular health. The results of these studies led to two conclusions: (1) most Americans do not get enough EPA and DHA from their diet to maintain optimal cardiac health; and (2) inadequate intake of EPA and DHA increases the risk for heart disease and premature death from cardiovascular disease.

Systematic investigations of the health effects of omega-3 fatty acids started with the observation of the very low mortality from coronary heart disease among Greenland Inuits despite their high fat diet. This finding led investigators to suspect that the high fish consumption of Inuits accounts for the cardioprotective effect. This hypothesis prompted many epidemiological and clinical studies of the relationship between fish consumption and heart disease (Dyerberg et al., 1975). Most, though not all, clinical studies have confirmed the beneficial effects of fish consumption on cardiovascular health. From these studies, the omega-3 polyunsaturated fatty acids EPA and DHA emerged as the ingredients principally responsible for the cardioprotectiv effects. The beneficial effects of EPA and DHA on heart disease were found to be substantial and have been compared to the positive effects of cholesterol-lowering drugs.

In reviewing the evidence from the large number of clinical trials, the American Heart Association concluded that omega-3 fatty acids can reduce the risk of cardiovascular disease. In 2002, the Association issued a set of guidelines for fish, fish oil and omega-3 fatty acids consumption (Kris-Etherton et al., 2002). Healthy individuals free of heart disease should eat fish at least twice a week. Individuals who already have heart disease should consume about 1 g/ day of EPA + DHA in capsule form. Patients with high plasma triglycerides should consume 2-4 g of EPA + DHA provided as capsules http://www.americanheart.org/presenter.jhtml?identifier=4632). Health organizations in other countries around the word have made similar recommendations. Rarely has a dietary supplement received such ringing endorsement from so many national and international health organizations.

It appears that the public is responding to calls to consume more omega-3 fatty acids. There are now dozens of omega-3 fatty acid supplements available on the market. Foods fortified with omega-3 fatty acids can be found in products ranging from beverages to sliced meat. There are eggs that contain as much as 100 mg of omega-3 fatty acids. Wegmans Food Markets introduced an omega-3 fatty acids enriched bread. Nestle launched a strawberry flavored drink for kids fortified with omega-3 fatty acids. There are snack bars fortified with omega-3 fatty acids that offer a tasty in-between-meals alternative to fish. Kellogg is said to have teamed up with Martek Biosciences for use of their algae omega-3. The introduction of the wide range of such products shows that there is a growing market for products fortified with omega-3 fatty acids.

Structural features of fatty acids

Fatty acids are long-chain carboxylic acids with 12 or more carbon atoms. Fatty acids may be saturated or unsaturated. Saturated fatty acids do not contain any double bonds between their carbon atoms. Unsaturated fatty acids contain carbon-carbon double bonds. They are either monounsaturated, i.e., they contain one carbon-carbon double bond or they are polyunsaturated, i.e., they contain more than one carbon-carbon double bond. In this review, polyunsaturated fatty acids are referred to as PUFAs. Some fatty acids cannot be synthesized by the body. They must be supplied through the diet and are referred to as essential fatty acids.

In their free (unesterified form), fatty acids are nearly insoluble in water. In blood, they are bound to albumin for transport to different sites within the body. The bulk of fatty acids, including polyunsaturated fatty acids, is stored in two main classes of lipids (see Figure 1 below): neutral triacylglycerol (storage pool) and in the more polar glycerophosholipids (structural and metabolic pool). Triacylglycerols constitute more than 90% of adipose tissue in mature animals. Glycerophospholipids contribute between 10-40% of the total fatty acids in muscle. Triglycerides contain three fatty acids connected to a glycerol backbone. Omega-3 and omega-6 polyunsaturated fatty acids are usually linked to the middle carbon atom of the glycerol backbone (*sn-2* position).

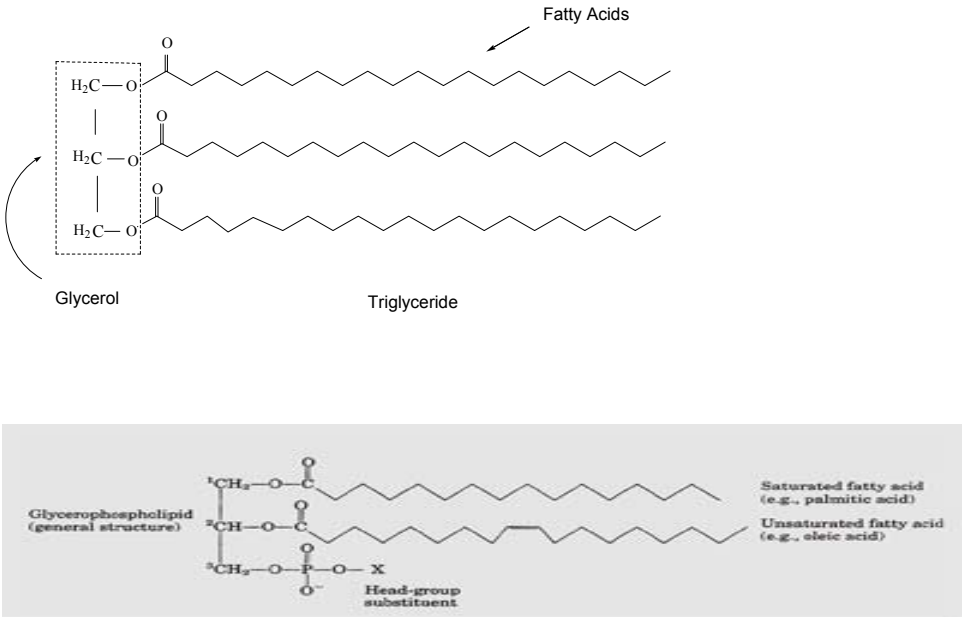

Figure 1: Fatty acids are linked via an ester bond to glycerol yielding triglycerides which are sometimes referred to as triacylglycerol (top). In contrast to triglycerides, glycerophospholipids contain only two fatty acids (bottom) with a polar phosphate group occupying the position at the third carbon atom of glycerol.

Omega-3 and omega-6 polyunsaturated fatty acids – structural differences

Depending on how many carbon atoms separate the terminal methyl carbon (the omega end) of PUFAs from the carbon atom of the first double bond, biochemists distinguish omega-6 and omega-3 polyunsaturated fatty acids (see Figure 2). In omega-6 fatty acids (sometimes written as ω-6 or n-6), the first double bond starts at the sixth carbon atom from the terminal methyl carbon of the molecule. In omega-3 fatty acids, the first double bond starts at the third carbon atom. The precursor of fatty acids in the omega-6 family is linoleic acid while the precursor of fatty acids in the omega-3 family is α-linolenic acid. EPA and DHA, the PUFAs most important for cardiovascular health, belong to the omega-3 family.

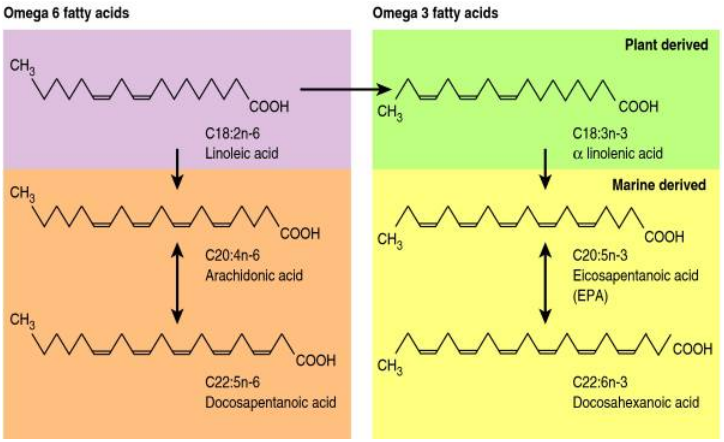

Figure 2: Omega-3 and omega-6 polyunsaturated fatty acids differ in the relative position of the first double bond from the methyl terminal end. From: Din et al., 2004

Observational studies provided the first clue about the health benefits of omega-3 fatty acids

Most observational studies have shown that populations with high fish consumption have a reduced risk of heart disease. For example, among Japanese the incidence of sudden cardiac death is 7.8 per 100,000 person years (Iso et al., 2006), whereas among European the incidence is 122 for men and 41 for women (Moore et al., 2006). Non-fatal cardiovascular events are less frequent in Japanese with high fish consumption compared to those with low fish consumption (Iso et al., 2006). A systematic review of 11 prospective cohort studies concluded that fish consumption at 40-60 g per day is associated with markedly reduced coronary heart disease mortality (Marckmann and Gronbaek, 1999).

An often cited study in discussions of the beneficial effects of fish consumption is the Chicago Western Electric Study. It included 1822 men who were 40-55 years old and free of cardiovascular disease at the beginning of the study in 1957. Participants were divided into four groups based on the amount of daily fish intake and followed over a 30 years period. Men with the highest fish consumption per day had a 42% lower risk of fatal coronary heart disease compared to those who consumed no fish at all (Daviglus et al., 1997). A more recent study reached a similar conclusion (Mozaffarian et al., 2005a). This study

followed 45,722 men free of known cardiovascular disease over a 14 year period. The results showed that omega-3 fatty acids from seafood reduced their risk of coronary heart disease by 40-50%.

The beneficial effect of fish consumption on cardiovascular health is not restricted to coronary heart disease but also extends to stroke. A group of 4775 adults 65 years and older without cerebrovascular disease at baseline was followed for 12 years. Those individuals who consumed fish were found to have a 27% lower risk of suffering an ischemic stroke (Mozaffarian et al., 2005b). Using data from the World Health Organization on 36 countries, Zhang et al. showed an inverse relationship between fish intake and mortality from all causes, coronary heart disease and stroke (Zhang et al., 1999). Thus, it is a general observation of the epidemiological and cohort studies that the beneficial effect of eating fish is more pronounced in populations and individuals with low fish consumption. For example, no protective effect of fish intake was observed in 11,000 Norwegian men, a population with relatively high fish consumption (Vollset et al., 1985).

Clinical intervention trials with omega-3 fatty acids targeting cardiovascular disease

The observations made in the epidemiological studies led to intervention trials to determine whether omega-3 PUFAs can reduce mortality and sudden cardiac deaths in patients with pre-existing heart disease. Table 1 lists 5 widely cited clinical studies on the cardioprotective effects of fish and fish oil. One of the first large intervention trials was the "Diet and Re-Infarction Trial" (DART-1) conducted in

Investigators	Subjects	Design	P value	Comments
Diet and Reinfarction Trial (DART-1) – 1989	2033 men with recent MI	Randomized controlled trial	<0.05	29% reduction in 2-year all-cause mortality
Diet and Reinfarction Trial (DART-2) - 2003	3114 men with angina	Randomized controlled trial		Increased risk of cardiac death
The Lyon Diet Heart Study – 1994 and 1999	605 men with recent MI	Secondary prevention trial	<0.0002	Three endpoints examined
GISSI-Prevenzione trial 1999	11,324 patients with recent MI	Open label RTC	0.01	20% reduction in overall mortality and 30% reduction cardiovascular mortality after 3.5 years
JELIS (Japanese EPA Lipid Intervention Trial) 2007	18,645 patients with hyperlipidemia	Open randomized trial with EPA alone	<0.011	Significant (19%) reduction in non-fatal coronary events

Table 1: Clinical intervention trials with omega-3 fatty acids

England in the early 1970s. The study followed 2033 men with prior myocardial infarction (MI) over a 2-year period. Those who consumed fish or fish oil showed a 29% reduction in all-cause mortality primarily due to a reduction in deaths from coronary heart disease.

The Lyon Diet Heart Study is a secondary prevention trial involving 605 men. All were patients who had survived a first heart attack and were subsequently followed for 4 years. One group received a diet rich in omega-3 and omega-9 fatty acids and low in omega-6 fatty acids and saturated fats (the Mediterranean Diet). Patients in the second group received no dietary directions but were asked by their

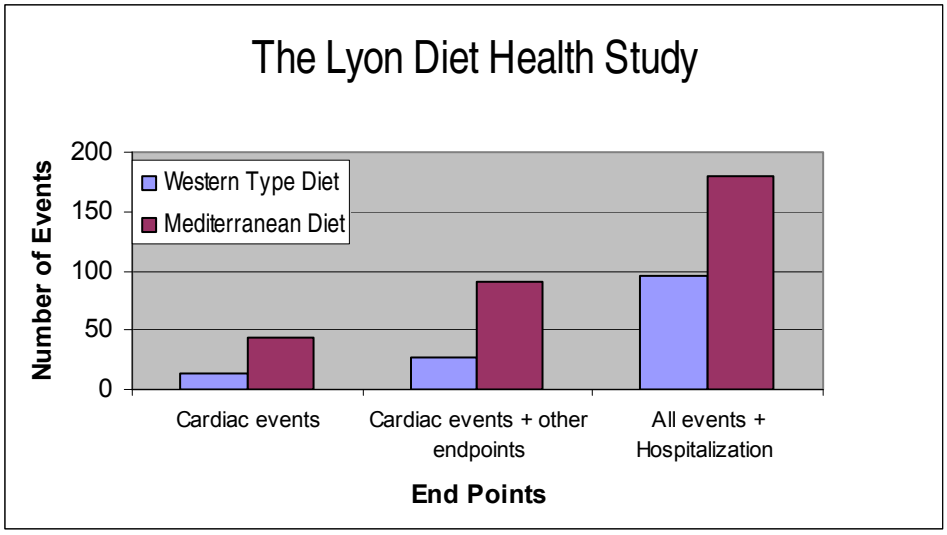

Figure 3: Results of the Lyon Diet Health Study. Adapted from GISSI-Prevenzione Investigators (1999) and redrawn from Schwalfenberg (2006).

physician to follow a "prudent" diet. This diet was described as similar to an American diet or Western diet. After almost 4 years, patients following the Mediterranean-style diet had a 50-70% lower risk of recurrent heart disease. This risk was determined by a combination of three outcome measures (see Figure 3).

The GISS Prevenzione Trial was a large study involving patients who had survived a recent myocardial infarct. They were randomly assigned to one of four groups: one group was given Omacor (1g/day), a second was given vitamin E, a third was given both Omacor and vitamin E. A fourth group received no treatment and served as control. More than two-thirds of patients were already eating fish once or more per week. After 3.5 years, participants randomized to fish oil capsules had a risk reduction for total mortality by 20%. The relative risk of cardiovascular death was reduced by 30% and the risk of sudden cardiac death was reduced by 45% (see Figure 4). Vitamin E had no effect.

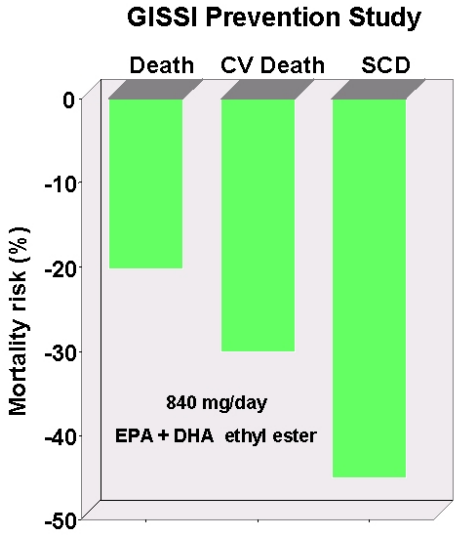

Figure 4: Results of the GISSI Prevention Study. Adapted from: GISSI-Prevenzione Investigators, Lancet 354: 447-455 (1999). Death = total mortality; CV Death = cardiovascular death; SCD = Sudden cardiovascular death.

The JELIS study (Japan EPA Lipid Intervention Study) is unique in that it tested the effect of EPA in patients with high cholesterol levels. All of the participants were taking cholesterol-lowering statins and all of them were on a diet rich in fish. Half of the participants took 1800 mg of EPA from fish oil in addition to their standard dose of statins. The other half took only statins and served as the control group. The duration of the study was 4.5 years. The study found a 19% decrease in the number of non-fatal coronary events. This was a striking outcome considering that the benefits were seen over and above the benefits due to the statin treatment.

In 2002, Bucher et al. published a meta-analysis of the clinical data on the cardiovascular effects of omega-3 fatty acids (Bucher et al., 2002). Analyzing data from 11 clinical trials involving 15,806 patients they reached the following conclusion: highly significant reductions in overall mortality ($p< 0.0001$) and fatal MI ($p<0.001$), and a significant reduction in sudden cardiac death ($p<0.01$).

Trials that did not find a beneficial effect of omega-3 fatty acids on cardiovascular disease

There are a number of clinical studies that failed to observe any beneficial effects of omega-3 PUFAs on various endpoints of heart disease. One those studies prompted a great deal of controversy because its negative results weighed heavily in a later meta-analysis of the clinical trials. The study was headed by the same investigators who conducted the DART-1 study and is referred to as the DART-2 study (see Table 1). DART-2 included 3114 men with stable angina. The goal of the study was to determine whether coronary mortality and morbidity can be reduced by an intake of oily fish or fish oil that provides 3 g of EPA each week. A group that was given no specific dietary advice served as control. The study observed an increase rather that a decrease in sudden and total cardiac deaths in men who eat fish or took fish capsules compared to patients who were given no specific dietary instructions. The highest death rate was observed in the group of patients who took fish capsules.

The publication of this study led to a great deal of criticism about the designs and conduct of the trial (von Schacky and Harris, 2007). Nevertheless, the study had a major impact in a meta-analysis of all the clinical data on the cardiovascular benefits of omega-3 fatty acids. The results of this meta-analysis were published in a 168 page paper in the Cochrane Reviews (Hooper et al., 2004). Data from 48 randomized controlled clinical trials (36,913 patients) were analyzed to assess whether omega-3 fats from oily fish and plants are beneficial in preventing and treating cardiovascular disease. The authors concluded that "…it is not clear that dietary or supplemental omega-3 fats alter total mortality, combined cardiovascular events in people with, or at high risk of, cardiovascular disease or in the general population". Two years later, the same authors published a shorter version of their analysis in the British Medical Journal (Hooper et al., 2006). Both reviews state in their summary that the evidence for a reduction in cardiovascular events and mortality is not conclusive. In both publications, it was the DART-2 study that shifted the balance toward a null effect of omega-3 fatty acids. Many prominent researchers have voiced their concerns about the reviews. Some of the critical comments on the reviews by Hooper et al. can be found at the website of the British Medical Journal (http://www.bmj.com/cgi/eletters/332/7544/739). A particularly outspoken critique has been published by von Schacky and Harris (von Schacky and Harris, 2007).

The American Heart Association and the US Food and Drug Administration now endorse n-3 PUFA consumption for cardiac health

The American Heart Association was not swayed in its assessment of the available data. It conducted its own reviews through workshops with prominent scientists and clinicians working the field. In 1999, such a workshop was held at the National Institutes of Health in Bethesda, Maryland to review the "Essentiality of and Recommended Dietary Intake for Omega-6 and Omega-3 Fatty Acids". A consensus was reached on the importance of reducing omega-6 PUFAs while increasing omega-3 PUFAs in the diet of adults and newborns (Simopoulos et al., 2000). The workshop concluded that an increase in α-linolenic acid together with EPA and DHA and a reduction of vegetable oils with high linoleic acid content are necessary to achieve a healthier diet in Western industrialized countries.

In 2002, the American Heart Association issued new guidelines on fish, fish oil and omega-3 fatty acids which were published in the journal *Circulation*. Different doses of omega-3 fatty acids were recommended for healthy individuals (two portions of fish per week), for patients with existing cardiovascular disease (about 1 g of EA + DHA provided as supplement), and for people who have elevated triglyceride levels (2-4 g of EPA + DHA per day provided as supplement). In 2004, FDA issued its second "qualified health claim" for omega-3 fatty acids stating that "supportive but not conclusive research shows that consumption of EPA and DHA ω-3 fatty acids may reduce the risk of coronary heart disease" (http://www.fda.gov/bbs/topics/news/2004/NEW01115.html). This approval was based on the review of thousands of scientific reports and the large body of clinical evidence indicating that consumption of fish and fish oils has beneficial effects on human health in general and cardiovascular health in particular.

Issues beyond the clinical data

Are omega-3 polyunsaturated fatty acids from fish and plants the same?

No, they are not. Omega-3 fatty acids from plants and marine sources differ in their chemical structure and their biological activity. This is an important issue since they are often lumped together as if they were equivalent. Terrestrial plants (not including algae) such as flaxseed contain the omega-3 fatty acid α-linolenic acid (ALA). They do not contain EPA or DHA. ALA is structurally different from EPA and DHA. It can serve as a precursor for EPA and DHA, but in humans the conversion is inefficient and insufficient (see next paragraph). Thus, while plants are a good source of omega-3 fatty acids, they are not an adequate source of EPA and DHA. Whether ALA by itself has beneficial effects on cardiovascular health is still controversial. It is therefore important to verify the identity of the omega-3 fatty acids used in fortification of various food products.

The omega-3 fatty acid α-linolenic acid in plants is not a good substitute for EPA and DHA in humans

For the majority of Americans, α-linolenic acid is the major source of long-chain omega-3 fatty acids. It is found in leafy green vegetables, flaxseeds, and walnuts. Certain vegetable oils, notably flaxseed oil and to a lesser extent canola oil, contain α-linolenic acid. There are repeated references in the literature that ALA can be converted in the body to EPA and DHA. This has led to the view that EPA and DHA can be supplied through a plant diet rich in ALA. A study of α-linolenic acid metabolism in adult healthy volunteers showed that only about 0.2 percent of ALA is converted to EPA (Pawlosky et al., 2001). It is noteworthy that the study found that 63% of the EPA derived from ALA was subsequently available for conversion to DHA indicating that EPA is a fairly good precursor of DHA in humans.

A study by Finnegan et al. reached a similar conclusion (Finnegan et al., 2003). These investigators compared ALA and EPA + DHA consumption in 50 mildly hyperlipidemic subjects over a period of 6 months. They found that dietary intake of 9.5 g/d of ALA failed to reduce plasma triglycerol concentrations despite increases in plasma EPA comparable with those observed with increased dietary EPA+ DHA. In fact, ALA was found to have a hypertriglyceridemic effect in contrast to the hypotriglyceridemic effect of EPA+DHA. This finding indicates that the plant-derived omega-3 fatty acid α-linolenic acid and marine-derived omega-3 fatty acids EPA and DHA have different physiologic effects. Together these studies show that ALA is an inadequate substrate for enhancing EPA and DHA production in healthy subjects.

A report by the UK Food Standard Agency also concluded that there is little, if any, benefit of ALA on risk factors for cardiovascular disease (Sanderson et al., 2002). This is despite the fact that ALA supplementation can increase EPA. A comprehensive review of the effects of ALA on cardiovascular disease outcomes concluded that ALA does not reduce the rate of all-cause mortality, cardiac and sudden death, and stroke (Wang et al., 2006). But other studies suggested that ALA may have value as a rescue remedy for those who do not regularly eat fish (Mozaffarian et al., 2005a) since a small portion is converted to EPA. Obviously, this issue is still controversial and is likely to continue to receive attention because ALA is the traditional vegetarian source of omega-3 fatty acids.

EPA versus DHA – are they the same?

Both EPA and DHA are recognized as important modulators of biochemical events that account for the beneficial effects of fish and fish oil on cardiovascular health. Fish and fish oil contain EPA and DHA in varying ratios depending on the type of fish. Nearly all clinical studies have used a mixture of EPA and DHA which prevents any identification of their individual contributions. EPA and DHA have been reported to have similar triacylglycerol-lowering effects but divergent effects on serum lipoprotein and fatty acid metabolism in humans (Grimsgaard et al., 1997). A study conducted through Kaiser Permanente concluded that DHA alone (from Martek Biosciences) is as good as DHA + EPA in lowering triglycerides in patients with coronary artery disease (Schwellenbach et al., 2006). In a recent review, Mori and Woodman concluded that for many endpoints relating to cardiovascular disease, DHA and EPA may not be the same (Mori and Woodman, 2006). They found that DHA but not EPA increased HDL cholesterol which is suspected to have antiatherogenic effects. DHA was also found to be more effective in reducing blood pressure and in decreasing heart rate. But the authors did not make any recommendations about the intake of one specific omega-3 fatty acid over another. More studies will be needed before a definitive statement can be made. It is quite possible that EPA and DHA differ in their effects on different outcome measures. It has already been shown, that DHA but not EPA is critical in brain development and for proper brain functions. The Massachusetts General Hospital is currently conducting a Phase III clinical trial (NCGT00361374) to compare the effect of 1 g/d EPA and 1 g/d of DHA, respectively, versus 1 g placebo/d in treating major depressive disorders. Future studies of this type may show that EPA and DHA have differential effects. This would be good news for manufacturers of EPA and DHA from algae since separation of the two omega-3 fatty acids from fish oil is difficult.

Bioavailability of EPA and DHA from triglycerides versus ethyl esters

Knowledge about the bioavailability of omega-3 fatty acids is important in establishing optimal formulations and optimal dosing schedules. In fish, EPA and DHA are present in the form of triglycerides. On average, fish triacylglycerols contain only 1 molecule of EPA or DHA along with 2 other fatty acids. Therefore, the omega-3 content does not exceed 33% of the fish extract. A higher concentration can be achieved through transesterification with ethanol which converts the omega-3 fatty acids into ethyl esters. Subsequent purification of this mixture yields a nearly homogeneous preparation of EPA and DHA (e.g., Omacor).

Studies that have conducted direct comparisons regarding the bioavailability of omega-3 fatty acids from triglycerides and ethyl esters have produced conflicting results. One study found omega-3 fatty acids from ethyl esters to be 50% less bioavailable than from triglycerides. Another study reported the same bioavailability of either source (Hansen et al., 1993). A study at the University of Trondheim in Norway concluded that absorption of EPA and DHA is as good from synthetic ethyl esters as it is from natural triacylglycerol (Krokan et al., 1993). In this study, fasting levels of EPA and DHA were measured in total lipids and phospholipids after administration for 14 days to obtain more relevant information about total absorption. If EPA and DHA from ethyl esters and triglycerides have the same bioavailability, the ethyl ester formulation would be advantageous since it contains the omega-3 fatty acids in the more concentrated form.

Bioavailability of EPA and DHA from fish versus fish oil

It is widely assumed that the bioavailability of EPA and DHA from fish or fish oil is the same. This has been confirmed in a recent study (Harris et al., 2007) which showed that consumption of equal amounts of EPA and DHA from oily fish or from fish oil capsules is equally effective at enriching blood lipids with omega-3 fatty acids. The study was conducted over a 16 week period to account for initially different rates of increases in EPA and DHA in plasma phospholipids and erythrocytes from the two sources. The authors concluded that "the consumption of equal amounts of EPA and DHA from oily fish on a weekly basis or from fish-oil capsules on a daily basis is equally effective at enriching blood lipids with omega-3 fatty acids".

Which is better – eating fish or taking fish oil supplement?

Since the data described in the section above show that omega-3 fatty acids from fish and fish oil are equally bioavailable, it may simply be a matter of preference. Anyone who likes sushi or fresh baked fish would not want to substitute fish for fish capsules. People who don't like fish can get the benefits simply by taking a fish oil capsule. However, there is concern about pollutants such as mercury which is present in fish in much higher concentrations than in fish oil (see section on Safety and Benefits below). But, at the doses recommended by the American Heart Association, the amounts of fish needed to supply up to 4 g of EPA and DHA per day are considered safe. This may not be true for pregnant women and infants. But fish is more than just a rich source of omega-3 fatty acids. It is also a rich source of proteins, vitamins and minerals. The much criticized DART-2 study reported that men with angina who consumed fish capsules had a higher risk of cardiac death than those who were advised to eat oily fish (Burr et al., 2003). If one believes the results of the DART-2 study (though most experts don't!), fish consumption may have had a more beneficial effect than taking fish oil capsules.

Long-chain omega-3 fatty acids from algae

A number of arguments can be made in favor of algae as a preferable source of omega-3 fatty acids. The percentage and ratio of EPA and DHA from fish oil varies depending on the fish source. Moreover, there are concerns about contamination of fish oils by environmental pollutants. Some experts fear that the supply of omega-3 fatty acids from fish is unlikely to meet future requirements considering the increasing demands (Sijtsma and de Swaaf, 2004). Perhaps the strongest argument in favor of omega-3 PUFAs from algae can be made if it turns out that EPA and DHA need to be available individually for specific pharmaceutical applications. Separation of EPA from DHA in fish oils is difficult to achieve on a processing scale. Consequently, alternative and cost-effective sources would be attractive. Certain microalgae produce high levels of EPA and DHA, and are already being used for the production of omega-3 PUFAs (Sijtsma and de Swaaf, 2004).

Safety: Risks and benefits

Fish is not only a good source of omega-3 fatty acids, it is also a source of mercury and other contaminants. Large, carnivorous fish that are high in the food chain, such as swordfish and shark, have the highest tissue concentration of mercury (1 μg/g) whereas tuna, trout, pike and bass have intermediate levels (0.1– 0.5 μg /g). Invertebrates such as shellfish have low concentration of mercury. The mean daily intake of mercury in the United States is 3.5 μg. Blood mercury levels in non-occupationally-at-risk individuals are about 2 μg/L in subjects who do not eat fish and 8.4 μg/L in individuals who eat 2-4 fish meals per week. Mercury levels of 300 μg/L produce overt symptoms of acute toxicity. A 100 g portion of sword fish would provide as much as 100 μg of mercury. While this amount is still far from the toxic level, it is obvious that high mercury levels in some fish are justifiably a safety concern.

Fish oils do not share the safety concerns – at least not with respect to mercury. Foran et al. measured mercury levels in 5 over-the-counter brands of fish oil (Foran et al., 2003). None of them contained significant amounts of mercury. As shown in the table below, levels of mercury in these brands of fish oil were negligible. This led the authors to conclude that consumption of fish oil may be preferable to consuming fish.

Mercury Content of 5 Preparations of Fish Oil	
Fish Oil Brand Name	Mercury Level in μg /L
CVS	10
Kirkland	<6
Nordic Ultimate	<6
Omega Brite	12
Sundown	<6

Table 3: From Foran et al., 2003

Mercury contained in fish is suspected to counteract the beneficial effects of n-3 PUFAs. A study published in the New England Journal of Medicine directly addressed this issue and concluded that high levels of mercury content may diminish the cardioprotective effect of fish intake (Guallar et al., 2002). Nevertheless, there appears to be a general consensus that the risk posed by contaminants does not outweigh the benefits of fish consumption. In 1997, the US Food and Drug Administration (FDA) granted "generally recognized as safe (GRAS)" status to refined menhaden fish oil (Administration, 1997). The agency also stated that consumption of up to 3 g/day of EPA + DHA from all sources can be considered safe for American adults. But, FDA has advised pregnant women and women who may become pregnant not to eat sword fish, king mackerel, tile fish, shark, or fish from locally contaminated areas.

Dosing

Mozaffarian and Rimm generated a diagram (see Figure 5) from a pooled analysis of data obtained in prospective and randomized clinical trials. The graph depicts the relationship between fish and fish oil intake and the relative risk of coronary heart disease (Mozaffarian et al., 2005a). Compared with little or no intake, modest consumption of 250-500 mg/d of EPA and DHA lowers the risk by 25% or more. Higher intake does not appear to provide any noticeably greater risk reduction suggesting a threshold effect. It appears that a dose of 1 g/day provides maximal protection. International recommendations for the consumption of long-chain omega-3 fatty acids reflect this relationship (see Table 1 in Appendix).

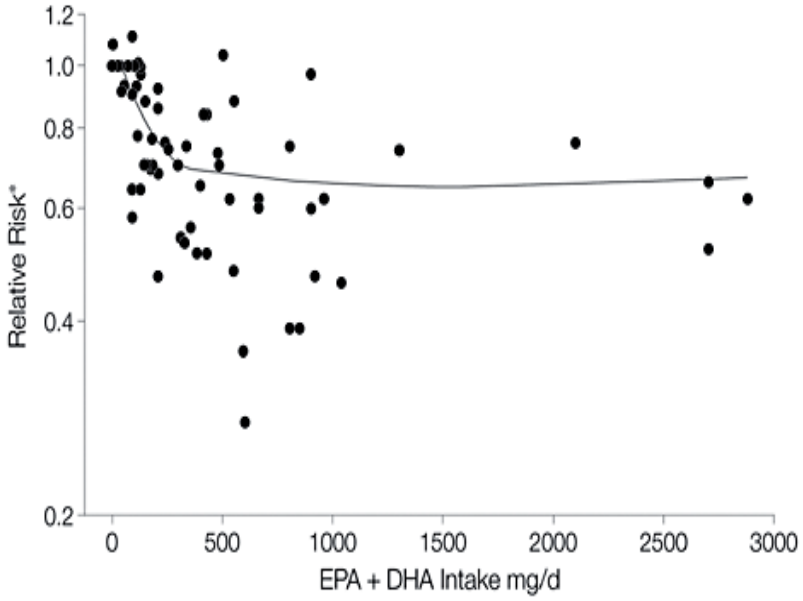

Figure 5: The relationship between fish or fish oil intake and the relative risk of death from coronary heart disease in prospective cohort studies and randomized clinical trials. From: Mozaffarian and Rimm, 2006

The American Heart Association has divided its recommendations into 3 categories: (A) persons without any coronary artery disease (2 fish meals per week, preferably oily fish), (B) persons with coronary artery disease (1 g EPA + DHA per day from oily fish or fish capsules), and (C) persons with elevated serum triacylglycerol levels (2-4 g EPA + DHA per day from fish capsules) (Kris-Etherton et al., 2002). Studies of the beneficial effects of omega-3 fatty acids on different outcome measures have shown noticeable differences in the amounts of EPA + DHA that are required. Some investigators have referred to this observation as the stratification effects of omega-3 fatty acid supplementation. At typical dietary intake of EPA + DHA the antiarrthymic effects pre-dominate. Higher to very high doses are required to achieve significant triglyceride-lowering effects (see Figure 6).

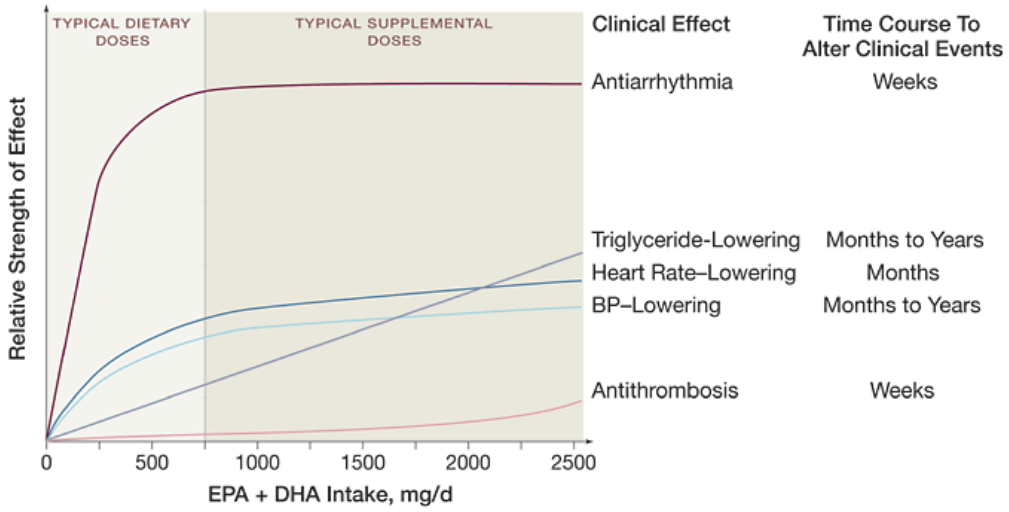

Figure 6: Stratification of the effects of omega-3 fatty acids consumed at various doses. From Mozaffarian and Rimm, 2006

The amount of EPA and DHA needed for cardiovascular risk reduction can be determined for everyone via the omega-3 index

Several studies reported a direct relationship between cardiovascular risk factors and the fraction of EPA + DHA expressed as the percentage of total fatty acids in plasma. Subjects with the highest percentile of EPA + DHA (>8%) in blood had a much lower risk of sudden cardiac death compared to subjects with the lowest percentile (<4%). This percentile value has been named the Omega-3 Index and has been proposed as a measure to assess the level of risk for death from coronary heart disease (Harris and von Schacky, 2004). Using data from clinical trials with omega-3 fatty acids, it was shown that an intake of 900 mg/day of EPA + DHA produces an Omega-3 Index of about 9.5.

Basic science studies are providing a scientific rationale for the beneficial health effects of omega-3 polyunsaturated fatty acids

The epidemiological and clinical studies on the cardioprotective effects of PUFAs prompted a large number of basic science studies attempting to identify the biological mechanisms by which EPA and DHA modify cardiovascular risks. The data that have emerged from these studies indicate that the EPA and DHA affect nearly every risk factor that has been linked to cardiovascular disease. Many studies have now identified mechanisms at the molecular level by which EPA and DHA influence the formation of atherosclerotic plaques, the most important pathologic process for cardiovascular disease (De Caterina et al., 2006).

Cardiovascular effect	Mechanism
Anti-arrhythmic	affect myocyte excitability; prevents atrial fibrillation
Anti-thrombotic	inhibits platelet aggregation reduces production of pro-inflammatory cytokines and eicosanoids,
Improves endothelial function	reduces adhesion molecule expression promotes nitric oxide induced endothelial relaxation
Improves blood lipid profile and cardiovascular risk factors	lowers triglycerides levels
Retards growth of atherosclerotic plaques	due to reduction in blood lipids and pro-inflammatory cytokines

Table 4: Omega-3 fatty acids influence a wide range of mechanisms to produce their beneficial effects on the cardiovascular system.

In recent years, much has been written about the link between inflammation and a host of chronic diseases, especially heart disease. Many studies now have documented that a diet rich in EPA and DHA results in a decrease in endogenously generated pro-inflammatory molecules. Inflammation is an important pathogenic feature in the evolution of atherosclerotic lesions. Vulnerable plaques are rich in inflammatory cells including monocytes and macrophages which secrete cytokines and other pro-inflammatory mediators that play a key role in acute coronary events.

As illustrated in Figure 7, omega-6 fatty acids give rise to pro-inflammatory prostaglandins and leukotrienes whereas omega-3 fatty acids give rise to anti-inflammatory prostaglandins and leukotrienes. Therefore, a shift in the balance between the two will significantly influence that mixture of pro- and anti-inflammatory bioactive molecules. Many chronic diseases such as cardiovascular disease and diabetes are associated with an increased production of thromboxane A_2, leukotriene B_4, interleukin-1β (IL-1β), interleukin-6, tumor necrosis factor-α (TNF-α) and C-reactive protein (CRP). All these factors increase when omega-6 fatty acid intake increases and decrease when omega-3 fatty acid intake increases. Many scientists now believe that this shift in the diet is responsible for the increase in chronic inflammatory diseases such as heart disease, diabetes and even Alzheimer's disease in industrialized countries. Supplementation with 750 mg DHA per day decreased the omega-6 to omega-3 ratio by 60% (Conquer and Holub, 1998). The optimal ratio is likely to vary between different diseases. A 4:1 ratio n-6 to n-3 appears to be optimal for maximum benefit for cardiovascular disease (Simopoulos, 2006).

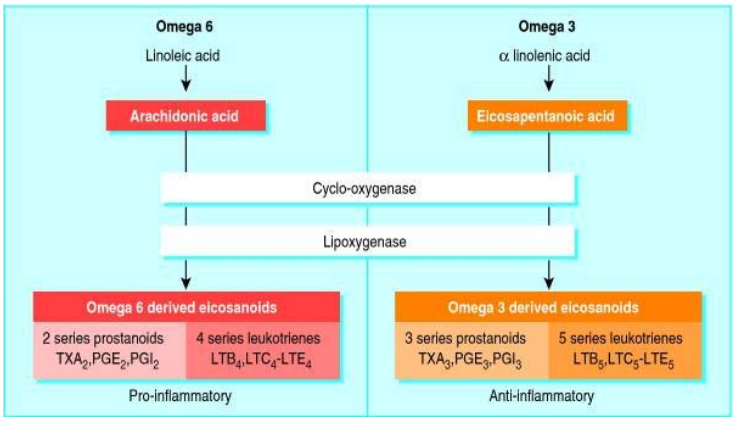

Figure 7: Synthesis of eicosanoids from omega-6 and omega-3 fatty acids. From (Din et al., 2004)

Effect of omega-3 fatty acids on cytokine production

Pro-inflammatory cytokines such as tumor necrosis factor α (TNF-α) and interleukin-1 (IL-1) are released during the early course of ischemic heart disesase. These two peptide molecules are key regulators of inflammatory responses and have been implicated as mediators of the cellular pathology of atherosclerosis. EPA and DHA consumption can suppress the production of TNF- and IL-1β (Caughey et al., 1996; James et al., 2000). The observation that EPA and DHA can strongly affect cytokine production in monocytes points to an important site of action of omega-3 PUFAs. Dietary fish-oil supplementation resulted in significant reductions in TNF-α and IL-6 production by peripheral blood monocytes. This effect was significant at a dose of 300 mg of EPA+ DHA and was maximal at a dose of 1 g/day (Trebble et al., 2003). EPA and DHA have been shown to inhibit the production of IL-1, IL-2 and TNF-α both *in vitro* and *in vivo* (Novak et al., 2003; De Caterina et al., 2004).

SUMMARY

1. Epidemiological and clinical studies strongly suggest that consumption of fish and fish oil have a positive effect on heart health and reduce the number of deaths from heart disease.

2. Americans do not get enough EPA and DHA from their diet to maintain optimal cardiovascular health.

3. The American Heart Association urges everyone to eat fish, preferably fatty fish, at least twice a week. Individuals with existing cardiovascular disease are advised to consume 1 g of EPA + DHA in form of supplements. For individuals with elevated triglyceride levels, the agency recommends 2-4 g of EPA + DHA supplements per day.

4. The omega-3 fatty acid α-linolenic acids from plants is not an adequate substitute for EPA and DHA from marine sources since its conversion in humans is inefficient.

5. Fish and fish oil are considered bioequivalent as sources of EPA and DHA.

6. EPA and DHA from algal sources are as beneficial as omega-3 from fish.

7. In contrast to some fish, fish oils contain only trace amounts of mercury.

8. Safety concerns led the American Heart Association and the FDA to issue warnings on fish consumption for pregnant women, women who are nursing, and for infants.

9. For healthy adults, the benefits of fish consumption outweigh the safety risk posed by contaminants in fish.

10. Basic science studies of the effects of EPA and DHA provide a scientific rationale for the observed positive effects of these omega-3 fatty acids on heart health.

11. Omega-3 fatty acids fortification of a wide range of food products is a rapidly expanding business.

REFERENCES

Administration FaD (1997) Substances affirmed as generally recognized as safe: menhaden oil. Fed Regist:30751-30757.

Bucher HC, Hengstler P, Schindler C, Meier G (2002) N-3 Polyunsaturated fatty acids in coronary heart disease: A meta-analysis of randomized controlled trials. Am J Med 112:298-304.

Burr ML, Ashfield PAL, Dunstan FDJ, Fehily AM, Breay P, Zotos PC, Haboubi NAA, Elwood PC (2003) Lack of benefit of dietary advice to men with angina: results of a controlled trial. Eur J Clin Nutr 57:193-200.

Caughey GE, Mantzioris E, Gibson RA, Cleland LG, James JA (1996) The effect on human tumor necrosis factor α and interleukin 1β production on diets enriched in n-3 fatty acids from vegetable oil or fish oil. Am J Clin Nutr 63:116-122.

Conquer JA, Holub BJ (1998) Effect of supplementation with different doses of DHA on the levels of circulating DHA as non-esterified fatty acids in subjects of Asian Indian background. J Lipid Res 39:286-292.

Daviglus ML, Stamler J, Orencia AJ, Dyer AR, Liu K, Greenland P, Walsh MK, Morris D, Shekelle RB (1997) Fish consumption and the 30-year of fatal myocardial infarction. N Engl J Med 336:1046-1053.

De Caterina R, Madonna R, Massaro M (2004) Effects of omega-3 fatty acids on cytokines and adhesion molecules. Curr Artheroscler Rep 6:485-491.

De Caterina R, Zampolli A, Del Turco S, Madonna R, Massaro M (2006) Nutritional mechanisms that influence cardiovascular disease. Am J Clin Nutr 83:421S-426S.

Din JN, Newby DE, Flapan AD (2004) Omega 3 fatty acids and cardiovascular disease - fishing for a natural treatment. Br J Med 328:30-35.

Dyerberg J, Bang HO, Hjorne N (1975) Fatty acid composition of the plasma lipids in Greenland Eskimos. Am J Clin Nutr 28:958-966.

Finnegan YE, Minihane AM, Leigh-Firbank EC, Kew S, Meijer GW, Muggli R, Calder PC, Williams CM (2003) Plant- and marine-derived n-3 polyunsaturated fatty acids have differential effects on fastingh and postprandial blood lipid concentrations and on the susceptibility of LDL to oxidative modification in moderately hyperlipidemic subjects. Am J Clin Nutr 77:783-795.

Foran SE, Flood JG, Lewandrowski KB (2003) Measurement of mercury levels in concentrated over-the-counter fish oil preparations. Is fish oil healthier than fish? Arch Pathol Lab Med 127:1603-1605.

Grimsgaard S, Bonaa KH, Hansen J-B, Nordoy A (1997) Highly purified eicosopentaenoic and docosahexaenoic acid in humans have similar triacylglycerol-lowering effects but divergents effects on serum fatty acids. Am J Clin Nutr 66:649-659.

Guallar E, Sanz-Gallardo MI, van't Veer P, Bode P, Aro A, Gomez-Aracena J, Kark JD, Riemersma RA, Martin-Moreno JM, Kok FJ, Group ftHMaMIS (2002) Mercury, fish oils, and the risk of myocardial infarction. N Engl J Med 347:1747-1754.

Hansen J-B, Olsen JO, Wilsgard L, Lyngmo V, Svensson B (1993) Comparative effects of prolonged intake of highly purified fish oils as ethyl ester or triglyceride on lipids, haemopstasis and platelet function in normolipaemic men. Eur J Clin Nutr 47:497-507.

Harris WS (2004) Fish oil supplementation: Evidence for health benefits. Cleveland Clin J Med 71:208 - 221.

Harris WS (2007) International recommendations for consumption of long-chain omega-3 fatty acids. J Cardiovasc Med 8:S50-S52.

Harris WS, von Schacky C (2004) The Omega-3 Index: a new risk factor for death from coronary heart disease? Preventive Medicine 39:212-220.

Harris WS, Pottala JV, Sands SA, Jones PG (2007) Comparison of the effects of fish and fish-oil capsules on the n-3 fatty acid content of blood cells and plasma phospholipids. Am J Clin Nutr 86:1621-1625.

Hooper L, Thompson RL, Harrison RA, Summerbell CD, Ness AR, Moore HJ, Worthington Hv, Durrington PN, Higgins JPT, Capps NE, Riemersma RA (2006) Risks and benefits of omega 3 fats for mortality, cardiovascular disease, and cancer: systematic review. Br J Med 332:752-760.

Hooper L, Thompson RL, Harrison RA, Summerbell CD, Moore H, Worthington HV, Durrington PN, Ness AR, Capps NE, Davey Smith G, Riemersma RA, Ebrahim SBJ (2004) Omega-3 fatty acids for prevention and treatment of cardiovascular disease. Cochrane Database of Systematic Reviews 2004, Issue 4 Art No: CD003177DOI: 101002/14651858CD003177pub2.

Iso H, Kobayashi M, Ishihara J, Sasaki S, Okada K, Kita Y, Kokubo Y, Tsugane S, Group ftJS (2006) Intake of fish and n3 fatty acids and risk of coronary heart disease among Japanese. The Japan Public Health Center-based (JPHC) Study Cohort I. Circulation 113:195-202.

James MJ, Gibson RA, Cleland LG (2000) Dietary polyunsaturated fatty acids and inflammatory mediator production. Am J Clin Nutr 71:343s-3438s.

Kris-Etherton PM, Harris WS, Appel LJ, Committee ftN (2002) Fish consumption, fish oil, omega-3 fatty acids, and cardiovascular disease. Circulation 106:2747-2757.

Krokan HE, Bjerve KS, Mork E (1993) The enteral bioavailability of eicosapentaenoic acid and docosahexaenoic acid is as good from ethyl esters as from glyceryl esters in spite of lower hydrolytic rates by pancreatic lipase in vitro. Biochim Biophys Acta 1168:59-67.

Marckmann P, Gronbaek M (1999) Fish consumption and coronary heart diseae mortality. A systematic review of prosective cohort studies. Eur J Clin Nutr 53:585-590.

Moore MJ, Glover BM, McCann CJ, Cromie NA, Ferguson P, Catney DC, Kee F, Adgey AAJ (2006) Demographic and temporal trends in out of hospital sudden cardiac death in Belfast. Heart 92:311-315.

Mori TA, Woodman RJ (2006) The independent effects of eicosapentaenoic acid and docosahexaenoic acid on cardiovascular risk factors in humans. Curr Opin Clin Nutr Metab Care 9:95-104.

Mozaffarian D, Ascherio A, Hu FB, Stampfer MJ, Willett WC, Siscovick DS, Rimm EB (2005a) Interplay between different polyunsaturated fatty acids and risk of coronary heart disease in men. Circulation 111:157-164.

Mozaffarian D, Longstreth J, W.T., Lemaitre RN, Manolio TA, Kuller LH, Burke GL, Siscovick DS (2005b) Fish consumption and stroke risk in elderly individuals. The cardiovascular health study. Arch Intern Med 165:200-206.

Novak TE, Babcock TA, Jho DH, Helton WS, Espat NJ (2003) NF-κB inhibition by ω-3 fatty acids modulates LPS-stimulated macrophage TNF-α transcription. Am J Physiol Lung Cell Mol Physiol 284:L84-L89.

Pawlosky RJ, Hibbeln JR, Novotny JA, Salem J, N. (2001) Physiological compartmental analysis of α-linolenic acid metabolism in adult humans. J Lipid Res 42:1257-1265.

Sanderson P, Finnegan YE, Williams CM, Calder PC, Burdge GC, Wootton SA, Griffin BA, MillwardN.C., Bemelmans WJE (2002) UK Food Standards Agency α-linolenic acid workshop report. Br J Nutr 88:573-579.

Schwellenbach LJ, Olson KL, McConnell KJ, Stolcpart RS, Nash JD, Merenich JA, Group CPCRSS (2006) The triglyceride-lowering effects of a modest dose of docosahexaenoic acid alone versus in combination with low dose eisocapentaenoic acid in patients with coronary artery disease and elevated triglycerides. J Am Coll Nutr 25:480-485.

Sijtsma L, de Swaaf ME (2004) Biotechnological production and application of the ω-3 polyunsaturated fatty acid docosahexaenoic acid. Appl Microbiol Biotechnol 64:146-153.

Simopoulos AP (2006) Evolutionary aspect of diet, the omega-6/omega-3 ratio and genetic variation: nutritional implications for chronic diseases. Biomed Pharmacother 60:502-507.

Simopoulos AP, Leaf A, Salem Jr. N (2000) Workshop statement on the essentiality of and recommended dietary intakes for omega-6 and omega-3 fatty acids. Prostaglandins Leukot Essent Fatty Acids 63:119-121.

Trebble T, Arden NK, Stroud MA, Wootton SA, Burdge GC, Miles EA, Ballinger AB, Thompson RL, Calder PC (2003) Inhibition of tumour necrosis factor-α and interleukin-6 production by mononuclear cells following dietary fish-oil supplementation in healthy men and response to anti-oxidant co-supplementation. Br J Nutr 90:405-413.

Vollset SE, Heuch I, Bjelke E (1985) Fish consumption and mortality from coronary heart disease. New Engl J Med 313:820-821.

von Schacky C, Harris WS (2007) Cardiovascular benefits of omega-3 fatty acids. Cardiovascular Res 73:310-315.

Wang C, Harris WS, Chung M, Lichtenstein AH, Balk E, M., Kupelnick B (2006) n-3 Fatty acids from fish or fish-oil supplements, but not α-linolenic acid, benefit cardiovascular disease outcomes in primary- and secondary-prevention studies: a systematic review. Am J Clin Nutr 84:5-17.

Zhang J, Sasaki S, Amano K, Kesteloot H (1999) Fish consumption and mortality from all causes, ischemic heart disease, and stroke: an ecologicla study. Prev Med 28:520-529.

APPENDIX

International recommendations for consumption of omega-3 fatty acids

Source	Date	EPA + DHA per day	n6-n3 ratio
NATO workshop	1989	800 mg	4:1
ISSFAL workshop	1999	650 mg	
Eurodiet	2000	200 mg	
Martin (France)	2001	450 mg (DHA 110-120 mg)	5:1
Health Council of the Netherlands	2001	200 mg	7.5:1
American Heart Association	2002	1000 mg (secondary prevention)	
US National Academy of Science	2002	90-160 mg	
European Society of Cardiology	2003	1000 mg (secondary prevention)	
WHO/FAO	2003	400-1000 mg (1-2 fish meals/week)	
ISSFAL	2004	at least 500 mg	
UK Scientific Advisory Committee on Nutrition	2004	2 fish portions per week (assumes 450 mg)	
Australia and New Zealand	2006	adequate: 90-160 mg; target: 430-610 mg	

Table 1: From (Harris, 2007)

Informal survey of fish oil capsules for sale in retail outlets in Kansas City in spring/summer 2003*

NAME	EPA/DHA (MG/CAPSULE)	CAPSULES TO PROVIDE 1 G	COST TO PROVIDE 1 G
Kirkland Signature fish oil concentrate	180/120	3	$0.07
Member's Mark omega-3 fish oil	180/120	3	$0.07
GNC Liquid Norwegian CLO (teaspoons)†	460/370	1	$0.11
Spring Valley MaxEPA	180/120	3	$0.13
Origin Natural fish oil concentrate	180/120	3	$0.15
Walgreen's fish oil concentrate	180/120	3	$0.15
Your Life fish oil concentrate	180/120	3	$0.17
GNC fish body oils 1000	180/120	3	$0.19
Natrol omega-3	180/120	3	$0.20
VitaSmart fish oil concentrate	180/120	3	$0.20
Sav-On Fish Oil Concentrate	180/120	3	$0.21
Sundown fish oil	180/120	3	$0.21
Nature Made Fish Oil	216/144	3	$0.22
Omega-3 Enteric Coated	180/120	3	$0.23
Nature's Bounty Natural Fish Oil	180/120	3	$0.25
Nature Made Fish Oil	216/144	3	$0.27
Rexall cholesterol-free fish oil	180/120	3	$0.31
GNC triple cod liver oil (CLO) caps	173/120	3	$0.30
TwinLab emulsified Norwegian CLO† (tablespoons)	516/344	1	$0.44

*This is not an exhaustive list, and periodic sale prices may be lower than these retail prices. Sales tax is not included. Many other products are available via the Internet, but shipping and handling charges must be considered in determining overall cost.
†This is a liquid; therefore, doses are in teaspoons or tablespoons instead of capsules.

From: (Harris, 2004)

Fish	EPA	DHA	EPA+DHA
Salmon (farmed)	0.587	1.238	1.825
Herring (Pacific)	1.056	0.751	1.807
Mackerel (Pacific)	0.555	1.016	1.571
Trout (farmed)	0.284	0.697	0.981
Tuna (canned)	0.198	0.535	0.733
Flounder	0.207	0.219	0.426
Shrimp	0.145	0.122	0.267
Haddock	0.065	0.138	0.203
Catfish	0.042	0.109	0.151
Cod	0.003	0.131	0.134

Fish sources of EPA and DHA in grams of fatty acids per 3 oz cooked fish (adapted from US Environmental Protection and Agency and US Department of Agriculture)