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A Prospective Study of Plasma Fish Oil Levels and Incidence of Myocardial Infarction in U.S. Male Physicians

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Objectives. **This study evaluated whether increased intake of fish oils (eicosapentaenoic and docosahexaenoic acids) might reduce the risk of coronary heart disease.**

Background. **Observational and clinical studies have suggested that increased intake of fish oils, as reflected in plasma levels of fish oils, may reduce the risk of myocardial infarction.**

Methods. **A nested case-control study was conducted among the 14,916 participants in the Physicians' Health Study with a sample of plasma before randomization. Each participant with myocardial infarction occurring during the first 5 years of follow-up was matched by smoking status and age with a randomly chosen control participant who had not developed coronary heart disease.**

Results. **Mean levels of fish oils (with 95% confidence interval [CI] for paired differences and p values) in case and control participants, expressed as percent of total fatty acids, were, for eicosapentaenoic acid, 0.26 versus 0.25 (95% CI −0.03 to 0.05, p = 0.70) in cholesterol esters and 0.56 versus 0.54 (95% CI −0.04 to 0.09, p = 0.44) in phospholipids, and for docosahexaenoic acid, 0.23 versus 0.24 (95% CI −0.07 to 0.04, p = 0.64) in cholesterol esters and 2.22 versus 2.14 (95% CI −0.10 to 0.27, p = 0.36) in phospholipids. Results adjusted for major cardiovascular risk factors showed a very similar lack of association between fish oil levels and the incidence of myocardial infarction.**

Conclusions. **These results indicate no beneficial effect of increased fish oil consumption on the incidence of a first myocardial infarction. However, the effect of very high levels of fish oils could not be evaluated.**

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The possibility of a beneficial effect of fish oils was raised by the observation of extremely low cardiovascular mortality in Greenland Eskimos, a population with an average estimated intake >350 g/day of whale and seal meat (1–3). High intakes of fish and very low rates of coronary heart disease were also found in Japan (4,5), where areas of higher fish intake were specifically shown to have lower rates of mortality for coronary heart disease compared with the rest of the country (6).

Basic research identified omega n-3 (ω-3) fatty acids in fish, represented primarily by eicosapentaenoic and docosahexaenoic acids, as agents affecting processes involved in coronary atherosclerosis (7–10). Their mechanisms of action seem to be related, at least in part, to eicosanoid metabolism (1,11). Eicosapentaenoic acid competes with arachidonic acid as substrate for cyclooxygenase and lipoxygenase. In particular, eicosapentaenoic acid is transformed into thromboxane A_3 (a weak platelet aggregator and vasoconstrictor) and prostacyclin I_3 (an active inhibitor of platelet aggregation and vasodilator) and decreases the production of thromboxane A_2 (a potent platelet aggregator and vasoconstrictor). This resulting decrease in thrombogenicity and vascular reactivity is a plausible mechanism for a possible protective effect of fish oils in cardiovascular disease (11–13). Fish oils may also reduce the risk of thrombosis by increasing concentrations of plasminogen activator and decreasing concentrations of plasminogen inhibitor and fibrinogen (14). Other potentially antiatherogenic actions of fish oils are reduction of plasma triglyceride levels (9) and blood pressure (15,16).

A randomized trial (17) of diet in patients with a previous myocardial infarction found a significant reduction in all-cause mortality and ischemic heart disease deaths in the patients assigned to the fatty fish intervention group. However, it is not known whether the oil in the fish or some other nutrients were responsible for the observed benefit. Observational studies of fish intake and cardiovascular disease in populations free of coronary disease have presented conflicting results (18–24).

The present study was carried out to investigate the association between fish oil levels in plasma cholesterol esters and phospholipids and the incidence of myocardial infarction in a cohort of apparently healthy men without a previous myocardial infarction. Because plasma levels of fatty acids are not available from previous cohort studies, the present study can

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address the specific role of fish oils on the risk of cardiovascular disease.

Methods

Study population. The study was conducted among participants in the Physicians' Health Study, a randomized, double-blind, placebo-controlled clinical trial designed to test the effect of aspirin on cardiovascular disease and of beta-carotene on cancer and cardiovascular disease. Detailed descriptions of the methods of the trial have been published elsewhere (25,26). Briefly, 21,071 U.S. physicians who were 40 to 84 years of age in 1982 and had no previous history of myocardial infarction, stroke, cancer, severe debilitating disease, current use of aspirin or vitamin A supplements or any contraindication to the use of aspirin were randomized to take either aspirin (325 mg every other day, supplied as Bufferin by Bristol-Meyers Products) and beta-carotene (50 mg every other day, supplied as Lurotin by BASF), aspirin and beta-carotene placebo, beta-carotene and aspirin placebo or both placebos. The randomized aspirin arm of the trial was terminated early after 5 years of follow-up, principally because of a significant beneficial effect of aspirin on the incidence of myocardial infarction (25,26), but the beta-carotene component is still ongoing.

Plasma collection. Before randomization, between August 1982 and December 1984, all participants were sent kits for blood sampling. Participants were instructed to have their blood drawn into the provided Vacutainer tubes containing EDTA, to centrifuge the blood and to return the plasma in polypropylene cryopreservation vials and preserved in cold packs (without freezing) by overnight shipment. From August 1982 through December 1984, a total of 14,916 blood samples were received, corresponding to 68% of the trial participants.

On their arrival at the laboratory, plasma samples were refrigerated and realiquoted into six 1.2-ml tubes. Computer-printed labels with the identification number only were attached to the tubes, and they were frozen at −82°C. Throughout the period of storage, the samples did not thaw.

Study design. The present study was designed as a nested case-control study based on the cohort of participants in the Physician's Health Study who sent a plasma sample before randomization (26,27). An incidence-density sampling plan within the cohort was implemented (28). At 6-month intervals and for each case of myocardial infarction occurring during the interval, the blood samples of the case participant and of one randomly selected control participant from the set at risk at the time the case developed the end point, matched to the case participant for age (±1 year) and smoking status at baseline (current smoker, past smoker or never-smoker), were retrieved and analyzed. Additional information on blood pressure, diabetes and other cardiovascular risk factors was obtained from the prerandomization self-administered questionnaire returned by each participant. The study was approved by the Human Research Committee of the Brigham and Women's Hospital.

End point ascertainment. The case definition was that of myocardial infarction (fatal plus nonfatal) adopted in the Physicians' Health Study (25). Specifically, every 6 months for the first year and annually thereafter, participants were sent a questionnaire to elicit reports of outcomes. Questionnaires of deceased participants were usually returned by their families or by the postal authority. Those who did not return the questionnaires were contacted by telephone. After 5 years, morbidity follow-up was 99.7% complete, and mortality follow-up was 100% complete.

For each participant reporting a myocardial infarction or for each death, relevant medical records were sought and reviewed by an end points committee. Complete medical records were obtained for >95% of the reported cases. The end points committee, comprising two internists, one cardiologist and one neurologist, used the World Health Organization criteria to confirm a myocardial infarction (29). According to these criteria, chest pain and either enzyme elevations or characteristic electrocardiographic changes have to be present for a positive diagnosis. In particular, silent myocardial infarctions and sudden unexpected deaths of unknown cause are not included in this definition.

Of 254 cases of myocardial infarction that occurred during the initial 5 years of follow-up among participants with a sample of plasma, 32 pairs for cholesterol esters and 41 for phospholipids could not be included because there was insufficient blood for biochemical analysis in a member of the case-control pair. Thus, we included in the analysis 222 case-control pairs for cholesterol esters and 213 for phospholipids.

Biochemical analyses. The samples of the case participants and their corresponding control participants were retrieved and thawed in a sonicator. Fatty acid levels were assayed in the Lipid Research Laboratory at the Brigham and Women's Hospital.

Fatty acids in plasma lipoprotein cholesterol esters and phospholipids were quantified after plasma extraction with hexane/isopropanol (3:2) (30), separation of the cholesterol esters and phospholipids by thin layer chromatography (31) and transmethylation in methanolic hydrochloride (32). The resulting fatty acid methyl esters were quantitated by capillary gas chromatography on a 100-m cyanosilicone column (SP2330, Supelco) using a temperature program from 140 to 240°C. A total of 56 peaks are reliably identified by pure standards and published retention times, representing >98% of total peak area. Retention times used to identify the eicosapentaenoic and docosahexaenoic peaks were 44.83 and 48.94 min, respectively. Additionally, both peaks were added to form a variable representing the total level of marine omega-3 fatty acids in cholesterol esters and in phospholipids.

For external quality control, plasma samples from study participants were pooled, aliquoted and stored under the same conditions as the individual plasma samples. At least one pair of pooled plasma specimens was included for biochemical analysis in each batch of 40 samples. The blind-replicate coefficients of variation for total levels of fish oils, calculated

Table 1. Comparison of Cardiovascular Risk Factors Between Case and Control Participants at Baseline

	Mean Value for Case Participants	Mean Value for Control Participants	p Value
BMI (kg/m^2)	25.7	24.8	<0.01
Systolic BP (mm Hg)	130.4	128.2	0.04
Diastolic BP (mm Hg)	81.3	79.5	0.04
Total cholesterol (mg/dl)	221.1	213.9	0.05
HDL cholesterol (mg/dl)	43.6	47.9	<0.01

p values correspond to the paired differences. BMI = body mass index; BP = blood pressure; HDL = high density lipoprotein.

Table 2. Correlation Coefficients Between Plasma Levels of Fish Oils and Continuous Cardiovascular Risk Factors at Baseline for Case and Control Participants Combined

	Eicosapentaenoic Acid	Docosahexaenoic Acid	Both Fish Oils
Cholesterol esters			
Age	0.04	0.04	0.05
BMI	0.09	0.02	0.06
Systolic BP	0.05	−0.03	0.00
Diastolic BP	0.08	−0.01	0.04
Total cholesterol	0.04	−0.02	0.00
HDL cholesterol	0.01	0.08	0.06
Phospholipids			
Age	0.04	0.07	0.07
BMI	0.08	0.04	0.06
Systolic BP	0.12*	0.07	0.10*
Diastolic BP	0.13*	0.09	0.11*
Total cholesterol	0.05	0.02	0.03
HDL cholesterol	0.06	0.02	0.02

*$p < 0.05$. Abbreviations as in Table 1.

on the basis of the analyses of these samples, were 49.5% and 21.0% for cholesterol esters and phospholipids, respectively.

All laboratory procedures were performed in a blinded manner with regard to case-control status. Samples for each matched pair were treated together and identically and included in the same analytic run from the outset, with the order within each pair assigned at random.

Statistical methods. The relation of fish oil levels with baseline risk factors for cardiovascular disease was assessed by product-moment correlation coefficients for continuous variables or by paired *t* tests for binary variables. Initially, the association between fish oil levels and myocardial infarction was assessed by paired *t* tests comparing mean differences between case and control samples (33).

Modeling of the probability of developing a myocardial infarction as a function of fish oil levels and of other covariates was performed by conditional logistic regression, which provides an adequate parametric model for the matched design and the incidence density sampling plan used. Under these circumstances, the analysis is equivalent to proportional hazards regression, with the difference that only a randomly chosen member of the risk set for a given failure, rather than the entire risk set, is used (34). The coefficients in the model are interpreted as natural logarithms of the relative risks for the corresponding variables, adjusted for other covariates in the model. Conditional logistic regression models were fitted with Egret, version 0.25.01 (35).

Fish oils were included in the conditional logistic models as continuous covariates as well as grouped by quintiles according to the distribution of levels in the controls. All p values reported are two-tailed.

Results

As expected from the matching, case and control participants had a mean age of 58.7 years (SD 8.46), with 17% of both reporting smoking at baseline. The levels of other cardiovascular risk factors for cases and controls at baseline and the 95% confidence intervals and corresponding p values for the paired comparisons are shown in Table 1. As expected, case participants had significantly higher levels of body mass index, systolic and diastolic blood pressures and total cholesterol and lower levels of high density lipoprotein (HDL) cholesterol.

The product-moment correlation coefficients between eicosapentaenoic acid and docosaheaenoic acid, and their combination, with body mass index, systolic and diastolic blood pressures and total and HDL cholesterol levels at baseline for the combined sample of case and control participants are shown in Table 2 for both cholesterol esters and phospholipids. The magnitude of each of these coefficients was small, and a positive correlation is significant for blood pressure and eicosapentaenoic acid only ($r = 0.12$ for systolic blood pressure and $r = 0.13$ for diastolic blood pressure) and for blood pressure and the sum of both fish oil levels ($r = 0.10$ for systolic blood pressure and $r = 0.11$ for diastolic blood pressure) in phospholipids. Fish oil levels were similar between diabetic and nondiabetic participants and between those with and without angina (data not shown).

Eicosapentaenoic and docosahexaenoic acids were moderately correlated with one another ($r = 0.33$ [$p < 0.001$] in cholesterol esters; $r = 0.45$ [$p < 0.001$] in phospholipids). The correlation between omega-3 fatty acids in cholesterol esters and phospholipids was $r = 0.35$ ($p < 0.001$) for eicosapentaenoic acid, $r = 0.06$ ($p = 0.21$) for docosanexaenoic acid, and $r = 0.19$ ($p < 0.001$) for the combination of both fish oils.

Mean levels of eicosapentaenoic and docosahexaenoic acids and their combination in cholesterol esters and phospholipids at baseline for case and control participants are shown in Table 3. Case participants had levels very similar to those of their matched control participants, and none of the differences were significant.

The conditional logistic regression coefficients for the levels of fish oils considered as continuous variables, adjusted for body mass index, history of diabetes, angina, blood pressure,

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Table 3. Comparison of Fish Oil Peak Values at Baseline Between Case and Control Participants

	Mean Value for Case Participants (SD)	Mean Value for Control Participants (SD)	95% CI	p Value
Cholesterol esters				
Eicosapentaenoic acid	0.26 (0.22)	0.25 (0.22)	−0.03–0.05	0.70
Docosahexaenoic acid	0.23 (0.28)	0.24 (0.34)	−0.07–0.04	0.64
Both fish oils	0.49 (0.40)	0.49 (0.47)	−0.08–0.07	0.88
Phospholipids				
Eicosapentaenoic acid	0.56 (0.35)	0.54 (0.36)	−0.04–0.09	0.44
Docosahexaenoic acid	2.22 (1.05)	2.14 (1.02)	−0.10–0.27	0.36
Both fish oils	2.79 (1.23)	2.58 (1.25)	...–0.33	0.32

Fish oils are given as percent of total fatty acid peak area. The 95% confidence intervals (CI) and p values reported correspond to the paired differences.

Table 4. Adjusted Results of Conditional Logistic Regression Models for Effect of Fish Oil Levels on Incidence of Myocardial Infarction*

	Coefficient†	Standard Error of Coefficient	p Value	Relative Risk‡ (95% CI)
Cholesterol esters				
Eicosapentaenoic acid	0.363	0.587	0.54	1.05 (0.91–1.21)
Docosahexaenoic acid	0.192	0.354	0.59	1.02 (0.94–1.11)
Both fish oils	0.180	0.263	0.50	1.05 (0.92–1.19)
Phospholipids				
Eicosapentaenoic acid	0.033	0.350	0.92	1.01 (0.84–1.21)
Docosahexaenoic acid	0.057	0.130	0.66	1.06 (0.81–1.39)
Both fish oils	0.042	0.107	0.70	1.06 (0.80–1.40)

In these models, fish oil levels are considered continuous variables measured in percentage points of total fatty acid peak area. *Results adjusted for body mass index, history of diabetes, angina, blood pressure, randomized treatment assignment and total and HDL cholesterol levels. †To calculate the relative risk associated with an increase in x percentage point units of fatty acid peak area for a particular fish oil, use the formula RR = $e^{x \cdot coefficient}$ (e.g., the relative risk associated with an increase of 0.2 percentage point units in docosahexaenoic acid in cholesterol esters is $e^{0.2 \cdot (-0.143)} = 0.97$). ‡Relative risk and 95% confidence interval (CI) associated with a 50% increase in the corresponding fish oil peak level.

randomized treatment assignment and levels of total and HDL cholesterol, are shown in Table 4. Further adjustment for levels of palmitic, stearic, oleic, linoleic, linolenic and arachidonic acids did not materially affect the conclusions (data not shown). The magnitude of these coefficients must be interpreted with respect to the magnitude of the individual fatty acid peak areas. For instance, an average increase of 0.5% U in peak area would represent an 89% increase in eicosapentaenoic acid peak area in phospholipids but a 22% increase in docosahexaenoic acid in phospholipids. To obtain estimates of relative risk that could be compared in terms of proportional increase in each fatty acid peak area, we used the logistic regression model to calculate the corresponding estimated risk ratios associated with a 50% increase in fatty acid peak area and their 95% confidence intervals. For eicosapentaenoic acid and docosahexaenoic acid, and their combination, they were 1.05 (95% CI 0.91 to 1.21), 1.02 (95% CI 0.94 to 1.11) and 1.05 (95% CI 0.92 to 1.19) in cholesterol esters and 1.01 (95% CI 0.84 to 1.21), 1.06 (95% CI 0.81 to 1.39) and 1.06 (95% CI 0.80 to 1.40) in phospholipids. For a 50% increase in total omega-3 fatty acid area, the power of the study to detect a 25% reduction in the incidence of myocardial infarction was >99% in cholesterol esters and 67% in phospholipids, and the power to detect a 50% reduction in risk was >99% in both classes of lipids.

Table 5 presents the risk ratios for the conditional logistic regression of fish oil levels categorized by quintiles, adjusted for other cardiovascular risk factors. None of the reported p values for linear trend across the quintiles was significant, indicating no evidence for a linear component in a dose-response model. As in the analysis with fish oils considered as continuous variables, further adjustment for levels of major fatty acids did not alter the estimates appreciably.

Despite a lack of association between fish oil levels and myocardial infarction, we evaluated whether there was effect modification by other cardiovascular risk factors. To do so, we evaluated the interaction term (the product of the cardiovascular risk factor and the specific fish oil) in conditional logistic regression models (with omega-3 fatty acids as continuous variables). None of the interactions between the fish oil levels and the cardiovascular risk factors studied, including those for age, smoking, systolic and diastolic blood pressures and total and HDL cholesterol levels, was statistically significant. Because aspirin reduced the risk of myocardial infarction in the trial by 44%, the possibility of interaction between fish oils and random assignment to aspirin was of particular relevance. The p values for the interaction terms between assignment to aspirin in the trial and eicosapentaenoic and docosahexaenoic acids and their sum considered as continuous variables were 0.22, 0.90 and 0.32 for cholesterol esters and 0.52, 0.38 and 0.35 for phospholipids, respectively. To explore further the joint effect of aspirin and fish oils, we categorized fish oil levels in two groups, below and above the median of the corresponding fish oil in the control participants. Unadjusted relative risk estimates for each combination of assignment to aspirin (yes or no) and fish oil levels (above or below median) are shown in

Table 5. Adjusted Relative Risk Estimates (and 95% confidence intervals) From Conditional Logistic Regression Model for Effect of Fish Oil Levels on Incidence of Myocardial Infarction, With Fish Oil Levels Categorized by Quintiles*

	Quintile 1	Quintile 2	Quintile 3	Quintile 4	Quintile 5	p Value Trend
Cholesterol esters						
Eicosapentaenoic acid						
Median level†	0.05	0.12	0.20	0.30	0.52	
Relative risk	1.00	1.37	1.68	0.96	1.79	0.26
95% CI	Reference	0.68–3.04	0.75–3.77	0.43–2.14	0.83–3.90	
Docosahexaenoic acid						
Median level†	0.00	0.11	0.18	0.25	0.43	
Relative risk	1.00	1.14	1.48	1.09	1.68	0.19
95% CI	Reference	0.50–2.61	0.67–3.28	0.45–2.66	0.75–3.74	
Both fish oils						
Median level†	0.12	0.23	0.39	0.55	0.94	
Relative risk	1.00	1.16	1.22	1.05	1.96	0.11
95% CI	Reference	0.51–2.60	0.50–2.94	0.47–2.34	0.87–4.41	
Phospholipids						
Eicosapentaenoic acid						
Median level†	0.24	0.38	0.49	0.61	0.90	
Relative risk	1.00	0.93	0.55	0.67	1.11	0.75
95% CI	Reference	0.44–1.98	0.25–1.23	0.31–1.45	0.52–2.35	
Docosahexaenoic acid						
Median level†	1.02	1.69	2.11	2.49	3.43	
Relative risk	1.00	1.23	0.81	1.06	1.26	0.64
95% CI	Reference	0.52–2.90	0.35–1.87	0.50–2.28	0.54–2.91	
Both fish oils						
Median level†	1.37	2.12	2.58	3.00	4.33	
Relative risk	1.00	1.62	0.61	1.52	1.55	0.30
95% CI	Reference	0.65–4.02	0.25–1.52	0.69–3.31	0.67–3.54	

*Results adjusted for body mass index, history of diabetes, angina, blood pressure, randomized treatment assignment and total and HDL cholesterol levels. †Median level for the quintile of the corresponding fish oil, expressed as percent of total fatty acid peak area. CI = confidence interval.

Table 6 (results were similar after adjustment for cardiovascular risk factors). Fish oil levels did not modify the benefit of aspirin on the incidence of myocardial infarction. Furthermore, there is no evidence for a protective effect of increased fish oil levels among those assigned to the no-aspirin arm of the trial.

Discussion

Strengths and limitations of present study. After 5 years of follow-up, increased levels of fish oils in plasma lipids, either analyzed as continuous variables or categorized in quintiles, conferred no significant reduction of the risk of myocardial infarction. Furthermore, for most of the estimates, the 95% confidence intervals were reasonably narrow, suggesting that even a substantial increase in plasma fish oil levels could have at most only a small benefit. Similarly, analysis of fish intake based on reported dietary data in the same cohort showed no association between fish consumption and risk of cardiovascular disease (36).

It is possible that the physicians at higher risk of myocardial infarction were more likely to be aware of the possible benefits of fish oils and may have modified their fish or fish oil intake differently than those who considered themselves at lower risk. If fish oils had a beneficial effect, selective increased intake of fish or use of fish oil supplements among those at higher risk would bias estimates of relative risk associated with fish oil intake toward the null. In our study, we used two strategies to control for other risk factors for cardiovascular disease. First, we used a matched design in which the control participant for each case participant was restricted to the same age (±1 year) and the same smoking category (current, past or never) as the corresponding case participant. Second, we used conditional logistic regression to adjust for other levels of important cardiovascular risk factors, including blood pressure, total and HDL cholesterol and presence of diabetes. The correlation between plasma levels of fish oils in cholesterol esters and phospholipids and these risk factors at baseline was very small, suggesting that such self-selection by risk factor status did not occur.

An additional limitation of this study is the precision of the chromatographic determinations of the marine omega-3 fatty acids, as reflected in the relatively high coefficients of variation for blind-replicate analysis of pooled plasma sample from the

Table 6. Relative Risk Estimates for the Effect of Aspirin and Fish Oil Levels on Incidence of Myocardial Infarction

	Fish Oil Levels Below Median		Fish Oil Levels Above Median		
	No Aspirin	Aspirin	No Aspirin	Aspirin	p Value Interaction
Cholesterol esters					
Eicosapentaenoic acid	1.00	0.62	0.95	0.65	0.79
Docosahexaenoic acid	1.00	0.65	1.12	0.73	0.99
Both fish oils	1.00	0.54	0.99	0.74	0.41
Phospholipids					
Eicosapentaenoic acid	1.00	0.67	1.81	1.23	0.98
Docosahexaenoic acid	1.00	0.62	1.25	0.84	0.95
Both fish oils	1.00	0.82	2.15	1.44	0.90

The groups with fish oil levels below and above the median are defined with respect to the corresponding fish oil in the control group. Estimates are from conditional logistic regression models.

study participants. These are small peaks on the chromatogram, typically constituting <0.5% of total fatty acid peak area in cholesterol esters and <3% in phospholipids because of the higher affinity of polyunsaturated omega-3 fatty acids for phospholipids (9). Nonetheless, there is no suggestion of a trend toward lower risk among those with higher levels of fish oils; in fact, the trend (nonsignificant) is in the opposite direction.

Comparison with results from other studies. Compared with results from dose–response trials of fish or fish oil supplements (9,37–40), the levels of eicosapentaenoic acid in cholesterol esters and phospholipids in our study were even lower than those typically resulting from the lowest intervention doses in these trials (usually 1.5 g of fish oil/day, corresponding roughly to two servings of 300 g of fatty fish/week). Although we cannot derive a precise estimate, the average intake of fish or fish oils in the present study was probably comparable to the estimated intake of ~150 mg of omega-3 fatty acids/day for the U.S. population based on foods available for consumption in the national food supply (10). Similar intakes have been found in other large-scale studies in the United States (24). Therefore, although confounding effects cannot be excluded, it seems unlikely that preventive intake of fish oils was not widespread enough to materially affect the results.

In the Zutphen study (18), 872 men who had completed a dietary history in 1960 were followed for an average of 20 years (78 died of coronary causes). In that study, a 40% reduction in the risk of death from coronary heart disease was found even in those participants who ate only 1 to 14 g of fish/day compared with those who did not eat fish. However, in the Zutphen cohort, intake of lean fish was also inversely related to coronary heart disease mortality, and the marked decrease in cardiovascular risk was associated with levels of fish intake lower than those required to show a physiologic effect of fish oils on blood pressure or lipid levels.

Fish intake also was associated with reduced coronary heart disease mortality in two additional cohort studies. In the Western Electric Study (19) 1,931 men were followed for an average of 25 years. In this study, the estimated risk reduction was less than that in the Zutphen study (men eating 1 to 17 g of fish had a 19% lower risk than those who ate no fish), although a dose–response relation between fish intake and coronary mortality was present. Similar results were found among 6,258 participants in the usual-care group of the Multiple Risk Factor Intervention Trial (24). After 7.5 years of follow-up, the relative risks for quintiles of omega-3 fatty acid intake two through five compared with the quintile one were 1.01, 0.87, 0.87 and 0.59.

Other prospective observational studies of fish intake have shown no beneficial effects. In a cohort in Norway, 11,000 men were followed for an average of 14 years (20). The estimated risk ratios of coronary heart disease mortality for the categories of 5 to 9, 10 to 14, 15 to 19, 20 to 24 and ≥25 servings of fish eaten/month, compared to 0 to 4 servings eaten/month, were 1.24, 1.10, 1.10, 1.23 and 1.06, respectively (p = 0.93 for the test of trend). Similarly, in a cohort of 10,966 Swedish men and women followed for 14 years (22), the relative risk estimates for the categories of moderate and high fish intake, compared with low fish intake, were 0.94 and 0.85 (neither achieved statistical significance). The 12-year follow-up study of 7,616 Japanese men in the Honolulu Heart Program (21) showed no consistent effect of fish intake. The relative risks for those consuming 1 to 2, 3 to 4, 5 to 6 and >6 oz (28 to 56, 84 to 112, 140 to 168 and >168 g) of fish/day compared to those with no intake of fish were 0.69, 1.11, 0.84 and 0.92, respectively, with a nonsignificant test for trend. Finally, in the report (23) of a study of dietary habits in relation to incidence of cardiovascular disease in 1,462 women in Sweden, there was no relation between fish intake and cardiovascular disease.

In contrast to previous studies, we used plasma levels of omega-3 fatty acids as biologic markers for fish intake. This is justified by the almost linear relation between intake of fish or fish oil supplements and plasma levels of eicosapentaenoic and docosahexaenoic acids over a range of doses (37–40). However, omega-3 fatty acids are not the only components of fish.

In fact, the possible effect of other constituents of fish has been proposed as an explanation of the inverse association between intake of lean fish and coronary mortality among the participants in the Zutphen study (18). Although the number of published studies is too small to permit a systematic study, differences in design, including end point definition, length of follow-up and level of fish intake in the participants, do not seem to explain the different results.

In a randomized controlled trial, 2,033 men with a previous myocardial infarction were randomized using a factorial design to receive advice or no advice on each one of three interventions: reduction in fat intake and increase in the polyunsaturated/saturated fat ratio, increase in fatty fish intake and increase in intake of cereal fiber (17). The intervention for the fish advice group consisted of recommending to the participants the intake of at least two weekly portions (200 to 400 g) of fatty fish. After 2 years of follow-up, with 224 deaths from all causes, the fish advice group showed a 29% reduction in total mortality and a 16% reduction in ischemic heart disease events. The differences in mortality between the two groups appeared very early in the trial and were maintained throughout the follow-up period.

Conclusions. The only clinical trial of fish intake in patients with preexisting coronary heart disease supports the hypothesis of a protective effect of fish intake on cardiovascular mortality. However, the conflicting results of observational cohort studies require further clarification. Because of the large number of cohort studies with dietary information already collected and processed, further insight into this issue could be gained from existing data. However, our results, based on plasma levels of fish oils rather than on nutrient intake data in a population with a relatively low fish intake, do not support the hypothesis of a beneficial effect of increased fish oil levels in plasma on the incidence of a first myocardial infarction in men.

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