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Original Contribution

Intake of Long-Chain ω-3 Fatty Acids From Diet and Supplements in Relation to Mortality

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Evidence from experimental studies suggests that the long-chain ω-3 fatty acids eicosapentaenoic acid and docosahexaenoic acid have beneficial effects that may lead to reduced mortality from chronic diseases, but epidemiologic evidence is mixed. Our objective was to evaluate whether intake of long-chain ω-3 fatty acids from diet and supplements is associated with cause-specific and total mortality. Study participants ($n = 70,495$) were members of a cohort study (the Vitamins and Lifestyle Study) who were residents of Washington State aged 50–76 years at the start of the study (2000–2002). Participants were followed for mortality through 2006 ($n = 3,051$ deaths). Higher combined intake of eicosapentaenoic acid and docosahexaenoic acid from diet and supplements was associated with a decreased risk of total mortality (hazard ratio (HR) = 0.82, 95% confidence interval (CI): 0.73, 0.93) and mortality from cancer (HR = 0.77, 95% CI: 0.64, 0.92) but only a small reduction in risk of death from cardiovascular disease (HR = 0.87, 95% CI: 0.68, 1.10). These results suggest that intake of long-chain ω-3 fatty acids may reduce risk of total and cancer-specific mortality.

cancer; cohort studies; dietary supplements; fish oil; mortality; ω-3 fatty acids

Abbreviations: CI, confidence interval; CVD, cardiovascular disease; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; FFQ, food frequency questionnaire; HR, hazard ratio; ICD-10, *International Classification of Diseases, Tenth Revision*; PUFA, polyunsaturated fatty acid; RR, relative risk; VITAL, Vitamins and Lifestyle.

Omega-3 (ω-3) polyunsaturated fatty acids (PUFAs), particularly the long-chain ω-3 PUFAs eicosapentaenoic acid (EPA; 20:5) and docosahexaenoic acid (DHA; 22:6), have been a subject of scientific interest for over 40 years (1). While long-chain ω-3 PUFAs are consumed primarily through fatty fish in the diet, use of ω-3-containing fish oil supplements is increasing in the United States (2). Several lines of research suggest that long-chain ω-3 PUFAs have antiinflammatory properties, and given that inflammation has been linked to the development of cardiovascular disease (CVD) and several cancers, there is great interest in better understanding the potential health benefits of ω-3 PUFAs (3, 4). While trials of ω-3 supplements and CVD mortality have been inconsistent (5–7), a recent review of experimental animal studies, human trials, and observational studies concluded that ω-3 intake reduces coronary heart disease mortality (8). However, epidemiologic evidence of benefit for other major causes of mortality remains mixed (9, 10).

In a previous exploratory study of supplement use and total mortality in the Vitamins and Lifestyle (VITAL) cohort, we found a borderline decreased risk of total mortality associated with use of fish oil supplements (hazard ratio (HR) = 0.83, 95% confidence interval (CI): 0.70, 1.00) (11). Here, we present data on the association between long-chain ω-3 intake from diet and supplements and cause-specific mortality in the VITAL cohort. Few previous cohort studies of healthy populations have examined the effect of long-chain ω-3 PUFA consumption on total, cancer, and CVD mortality (10), and to our knowledge none have included combined intake from both dietary and supplemental sources.

METHODS

Study cohort

The VITAL Study is a prospective cohort study of men and women aged 50–76 years which has been described in detail elsewhere (12). Briefly, participants for the VITAL Study were recruited from a 13-county area in western Washington State. Between October 2000 and December 2002, a total of 364,418 potential study participants in the 13-county area were mailed a 24-page questionnaire with a special emphasis on supplement use. Of the 77,718 participants who returned questionnaires and passed general quality-control checks, 7,178 were excluded for not accurately completing the food frequency questionnaire (FFQ) portion of the questionnaire; these persons were excluded if they did not complete at least 5 items per page on the FFQ or if they reported an abnormally high (>5,000 kcal for men, >4,000 kcal for women) or low (<800 kcal for men, <600 kcal for women) daily energy intake. An additional 45 participants were excluded because they reported having a condition that would affect absorption of supplements (e.g., gastric bypass surgery). This left 70,495 participants for analysis.

Exposure measure: diet and supplement use

The exposures of interest in this study included consumption of dark fish (the main source of dietary EPA and DHA), use of fish oil supplements, and intake of EPA, DHA, and EPA + DHA from diet alone and from diet plus supplements. Diet during the year prior to baseline was assessed using a 120-item semiquantitative FFQ. The VITAL FFQ was adapted from the FFQ developed for the Women's Health Initiative, with special emphasis on measurement of micronutrient-fortified foods and dietary fats (13). The FFQ contains 12 "adjustment questions" regarding food purchasing and preparation (e.g., types of fats used in cooking and at the table, types of salad dressing used, etc.) and questions concerning frequency of food consumption, with 9 possible frequency choices. Sex-specific serving-size choices were also offered, with reference pictures provided for each serving-size option. Participants were queried about consumption of 5 different types of seafood items, including dark fish (e.g., salmon and fresh tuna), from which we calculated adjusted annual frequencies of dark fish consumption, taking portion size into account. Daily intakes of nutrients (including EPA and DHA) were calculated by multiplying the adjusted serving size of each food by the specific nutrient value for each item. Fatty acid values for serving-size-adjusted items were obtained from the University of Minnesota's Nutrition Coding Center database (14).

In addition, participants were asked to report the frequency (days per week) and duration (years) of ω-3 supplement use over the past 10 years (15). Since daily dose of nonvitamin, nonmineral supplements was not ascertained, the amount of supplemental EPA and DHA obtained from supplements was estimated on the basis of the average suggested daily dose among the most popular brands of ω-3 PUFA supplements (0.64 g/day for EPA and 0.35 g/day for DHA), multiplied by 10-year average use (days per week/7 × years/10). Total fatty acid doses were calculated by adding the estimated daily intake of the specific fatty acids from diet to the estimated 10-year average intake from supplements.

Outcome measurement

There were a total of 3,051 deaths in the study on or before December 31, 2006. Information on deaths was gathered primarily from Washington State death records ($n = 3,021$) through linkage based on participant identifiers. Deaths were also ascertained through the Social Security Death Index ($n = 26$), next of kin ($n = 2$), or the Surveillance, Epidemiology, and End Results cancer registry for western Washington ($n = 2$).

Cause of death was obtained through the Washington State death records using *International Classification of Diseases, Tenth Revision* (ICD-10), codes but was not available for deaths reported by other sources. We categorized causes of death as CVD (ICD-10 codes I00–I99), cancer (ICD-10 codes C00–D48), or other causes (all other codes). CVD deaths were further classified as being due to ischemic heart disease (ICD-10 codes I20–I25) or not. Cancer deaths were further categorized by cancer type: colorectal (ICD-10 codes C18–C20 and C26.0), pancreatic (ICD-10 code C25), bronchus and lung (ICD-10 code C34), breast (ICD-10 code C50), lymphoid, hematopoietic, and related tissues (ICD-10 codes C81–C96), and all other types (ICD-10 codes C00–D48, excluding those already listed). Disease-specific analyses were performed for categories with more than 100 deaths.

Covariates

Covariates of interest were assessed by means of the baseline questionnaire, and data are summarized in Table 1. Different sets of covariates were selected a priori to be included in the total mortality model and each cause-specific model based on established and probable risk factors for each outcome (listed fully in the footnotes of Tables 2 and 3). All models included adjustment for demographic factors, body mass index (weight (kg)/height $(m)^2$), physical activity, alcohol drinking, fruit and vegetable intake, smoking, use of nonsteroidal antiinflammatory drugs, screening, self-reported health, and energy intake. We used the residual method of adjustment for total energy in a sensitivity analysis. We also adjusted for ω-6 PUFAs because they competitively inhibit the less inflammatory properties of ω-3 PUFAs (16). We adjusted for arachidonic acid (20:4)—the ω-6 PUFA most associated with inflammation—in all analyses and for another ω-6 PUFA, linoleic acid (18:2), in a sensitivity analysis.

To adjust for health history in the total mortality models, we used a summary morbidity score (similar to a propensity score) to reduce the number of covariates in these models. This score was calculated from age-adjusted, sex-specific proportional hazards models which included 23 serious medical conditions for men and 27 for women (listed in footnote c of Table 2), with death as the outcome. We then assigned each individual a morbidity score using the β coefficients for death based on the participant's own set of health conditions. Forty-six percent of participants had a score of zero (reference group; no adverse health conditions), and 3% had a risk score above 1.5 (mortality HR > 10) (17).

Table 1. Distribution (%) of Baseline Risk Factors for Mortality According to Total EPA + DHA Intake (Diet + Supplements), VITAL Study, 2000–2006

Variable	Quartile of EPA + DHA Intake, g/day			
	First (0–0.082) (*n* = 17,703)	Second (0.083–0.174) (*n* = 17,485)	Third (0.175–0.322) (*n* = 17,601)	Fourth (>0.322) (*n* = 17,498)
Demographic Factors				
Female sex	63	57	47	36
Age at baseline, years				
50–<55	24	24	25	23
55–<60	22	23	23	24
60–<65	18	18	19	19
65–<70	16	16	17	17
70–<77	20	20	17	18
Race				
White	94	94	93	92
Hispanic	1	1	1	1
Black	1	1	1	1
American Indian or Alaska Native	2	1	1	1
Asian or Pacific Islander	1	2	2	3
Other/missing	1	1	1	2
Education				
High school or less	27	21	16	13
Some college/technical school	42	40	36	35
College or advanced degree	31	39	48	53
Lifestyle Factors				
Body mass index[a] at age 45 years				
<18.5	2	1	1	1
18.5–<25	53	53	53	50
25–<30	29	31	33	35
≥30	11	11	10	11
Missing	5	4	3	3
Average physical activity in previous 10 years, MET-hours/week				
None	20	16	13	10
Tertile 1 (0–4.38)	33	31	27	23
Tertile 2 (4.39–13.59)	26	28	30	29
Tertile 3 (>13.59)	20	25	30	38
Smoking				
Never smoker	47	48	48	47
Ever smoker, pack-years				
1–12.5	15	16	17	18
12.6–35.0	17	18	18	19
>35.0	21	18	17	17

Table continues

However, in the cause-specific models, we instead adjusted for personal history of that specific condition as reported at baseline. Similarly, in analyses of total mortality, we adjusted for family history by adjusting for the ages at death of the mother and father, and in cause-specific mortality analyses, we instead adjusted for family history of the specific condition under study.

Statistical analysis

We used Cox proportional hazards regression with age as the time scale to estimate risk of mortality across levels of the ω-3 PUFA exposures of interest. We checked proportional hazards assumptions using the Schoenfeld residual method, finding no evidence of violation of these assumptions.

Table 1. Continued

Variable	Quartile of EPA + DHA Intake, g/day			
	First (0–0.082) (*n* = 17,703)	Second (0.083–0.174) (*n* = 17,485)	Third (0.175–0.322) (*n* = 17,601)	Fourth (>0.322) (*n* = 17,498)
Alcohol use at age 45 years,[b] drinks/day				
None	25	19	16	15
<1	54	57	56	53
1–2	9	11	14	16
>2	9	10	12	14
Missing	2	2	2	2
Vegetable consumption, servings/day				
Quartile 1 (0.0–1.05)	40	27	19	14
Quartile 2 (1.06–1.66)	26	26	26	22
Quartile 3 (1.67–2.53)	19	25	27	28
Quartile 4 (>2.53)	15	21	28	36
Medical History				
Aspirin use in past 10 years[c]				
None	74	73	71	69
Low (<4 days/week or <4 years)	12	12	13	13
High (≥4/days/week and ≥4 years)	10	11	11	13
Missing	5	5	5	5
Sigmoidoscopy in past 10 years (yes)	53	56	59	62
Mammogram in past 2 years (yes)	90	91	93	91
Prostate-specific antigen test in past 2 years (yes)	70	72	73	77
Self-rated health				
Excellent	12	13	16	18
Very good	36	38	39	40
Good	38	36	34	32
Fair	12	11	9	9
Poor	2	2	1	2

Abbreviations: DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; MET, metabolic equivalent of task; VITAL, Vitamins and Lifestyle.
[a] Weight (kg)/height (m)^2.
[b] Former users were omitted.
[c] Aspirin use (baby aspirin omitted) over the 10 years before baseline.

Participants were censored when they moved out of Washington State (*n* = 2,920; 4.1%) or withdrew from the study (*n* = 17; 0.02%). We used the National Change of Address system, followed by phone calls and mailings, to identify persons who moved out of the state. We divided participants into quartiles of intake and calculated hazard ratios comparing categories of ω-3 PUFA intake, using the lowest quartile as the reference group. Tests for trend were performed by modeling quartiles of intake as a single "trend" or "grouped linear" variable in a separate Cox model. Cause-specific mortality was evaluated with adjustment for different sets of covariates appropriate for each cause (as indicated in the footnotes of Table 3) and was further stratified according to whether the participant had reported a history of that condition at baseline.

We created a "missing" category for 8 covariates with more than 5% missing data in order to reduce the number of participants who would be dropped from the analysis. In a sensitivity analysis, we also reanalyzed the association between total EPA + DHA and total mortality, using 3 other approaches to missing data: inclusion of only those persons with complete data; inverse probability weighting; and use of a more parsimonious model which included more participants with complete data. In another sensitivity analysis, we removed the first 2 years of follow-up for each participant. Finally, we explored the possibility that factors associated with inflammation (sex, smoking, body mass index, and use of nonsteroidal antiinflammatory drugs) could modify the association between long-chain ω-3 PUFA intake and total mortality. Analyses were conducted using Stata 12.1 (StataCorp LP, College Station, Texas).

RESULTS

Table 1 presents participant characteristics by quartile of long-chain ω-3 PUFA intake (EPA + DHA from diet plus

Table 2. Hazard Ratios for Total Mortality Associated With Total EPA + DHA Intake (Diet + Supplements), VITAL Study, 2000–2006[a]

Supplement Use or Intake, by Quartile	No. of Participants	% of Participants	No. of Deaths	Crude Mortality Rate per 1,000 Person-Years	Sex- and Age-Adjusted		Multivariate-Adjusted[b]		
					HR	95% CI	HR	95% CI	*P* for Trend
Dark fish, adjusted annual frequency of consumption									
1 (0–0)	24,917	35.4	1,395	11.22	1.00	Reference	1.00	Reference	0.003
2 (6–12)	16,508	23.4	694	8.37	0.74	0.68, 0.81	0.95	0.86, 1.04	
3 (14–28)	16,732	23.7	594	7.09	0.63	0.57, 0.69	0.88	0.79, 0.97	
4 (>42)	12,338	17.5	368	5.97	0.56	0.50, 0.62	0.86	0.76, 0.98	
Use of fish oil supplements									
None	63,313	90.0	2,760	90.9	1.00	Reference	1.00	Reference	0.369
Low (0–4 days/week or <3 years)	3,929	5.6	147	4.8	0.97	0.84, 1.14	0.98	0.83, 1.18	
High (≥4 days/week for ≥3 years)	3,045	4.3	130	4.3	0.84	0.71, 1.00	0.92	0.76, 1.10	
Dietary EPA + DHA, g/day									
1 (0–0.076)	17,768	25.2	935	10.53	1.00	Reference	1.00	Reference	0.008
2 (0.077–0.160)	17,512	24.8	792	9.02	0.83	0.76, 0.91	0.96	0.87, 1.06	
3 (0.161–0.289)	17,606	25.0	702	7.97	0.72	0.65, 0.80	0.93	0.84, 1.04	
4 (>0.289)	17,609	25.0	622	7.07	0.61	0.55, 0.68	0.85	0.76, 0.96	
EPA + DHA from diet and supplements, g/day									
1 (0–0.082)	17,703	25.2	935	10.57	1.00	Reference	1.00	Reference	0.004
2 (>0.082–0.174)	17,485	24.9	785	8.95	0.83	0.75, 0.91	0.97	0.87, 1.07	
3 (>0.174–0.322)	17,601	25.0	667	7.75	0.69	0.62, 0.76	0.89	0.80, 1.00	
4 (>0.322)	17,498	24.9	650	7.45	0.64	0.58, 0.71	0.86	0.77, 0.96	
EPA + DHA from diet and supplements, adjusted for dietary AA, g/day									
1 (0–0.082)	17,703	25.2	935	10.57	1.00	Reference	1.00	Reference	0.001
2 (>0.082–0.174)	17,485	24.9	785	8.95	0.83	0.75, 0.91	0.95	0.86, 1.06	
3 (>0.174–0.322)	17,601	25.0	667	7.75	0.69	0.62, 0.76	0.87	0.78, 0.97	
4 (>0.322)	17,498	24.9	650	7.45	0.64	0.58, 0.71	0.82	0.73, 0.93	
Dietary DHA, g/day									
1 (0–0.049)	17,624	25.0	931	10.58	1.00	Reference	1.00	Reference	0.008
2 (>0.049–0.106)	17,624	25.0	815	9.23	0.85	0.77, 0.93	0.98	0.89, 1.08	
3 (>0.106–0.194)	17,624	25.0	690	7.82	0.71	0.64, 0.79	0.92	0.82, 1.02	
4 (>0.194)	17,623	25.0	615	6.99	0.60	0.54, 0.67	0.86	0.76, 0.97	

Table continues

supplements) as reported at baseline. Compared with participants reporting lower levels of long-chain ω-3 consumption, participants in the highest quartile were more likely to be male, to be more highly educated, to have higher intakes of fruits and vegetables, to drink more alcohol, and to be more physically active. Participants with higher long-chain ω-3 consumption were less likely to have ever used estrogen therapy (among women) but had higher energy intakes and a lower percentage of energy derived from saturated fat and *trans* fat (data not shown).

A total of 3,051 (4.33%) participants died during a mean of 5.0 years of follow-up (8.65 deaths per 1,000 person-years), including 769 deaths from CVD, 1,485 from cancer, and 679 from other causes. Table 2 gives the hazard ratios for total mortality associated with the two major sources of EPA and DHA: dark fish and fish oil supplements. After multivariable adjustment, persons in the highest quartile of dark fish consumption had a significantly lower risk of death than those in the lowest quartile (HR = 0.86, 95% CI: 0.76, 0.98; *P* for trend = 0.003). The adjusted hazard ratio for the

Table 2. Continued

Supplement Use or Intake, by Quartile	No. of Participants	% of Participants	No. of Deaths	Crude Mortality Rate per 1,000 Person-Years	Sex- and Age-Adjusted		Multivariate-Adjusted[b]		
					HR	95% CI	HR	95% CI	*P* for Trend
DHA from diet and supplements, g/day									
1 (0–0.054)	17,572	25.0	933	10.62	1.00	Reference	1.00	Reference	0.004
2 (>0.054–0.113)	17,572	25.0	806	9.15	0.84	0.76, 0.92	0.99	0.89, 1.09	
3 (>0.113–0.207)	17,572	25.0	662	7.52	0.69	0.62, 0.76	0.89	0.80, 1.00	
4 (>0.207)	17,572	25.0	636	7.26	0.63	0.57, 0.69	0.86	0.77, 0.97	
Dietary EPA, g/day									
1 (0–0.024)	17,624	25.0	909	10.32	1.00	Reference	1.00	Reference	0.055
2 (>0.024–0.053)	17,624	25.0	780	8.82	0.84	0.76, 0.92	0.97	0.88, 1.07	
3 (>0.053–0.096)	17,624	25.0	694	7.87	0.73	0.66, 0.80	0.95	0.85, 1.06	
4 (>0.096)	17,623	25.0	668	7.59	0.66	0.59, 0.73	0.89	0.80, 1.00	
EPA from diet and supplements, g/day									
1 (0–0.027)	17,573	25.0	922	10.49	1.00	Reference	1.00	Reference	0.014
2 (>0.027–0.058)	17,571	25.0	762	8.65	0.81	0.73, 0.89	0.95	0.86, 1.05	
3 (>0.058–0.112)	17,572	25.0	664	7.54	0.69	0.62, 0.76	0.89	0.80, 0.99	
4 (>0.112)	17,571	25.0	689	7.87	0.68	0.62, 0.76	0.88	0.79, 0.99	

Abbreviations: AA, arachidonic acid; CI, confidence interval; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; HR, hazard ratio; MET, metabolic equivalent of task; VITAL, Vitamins and Lifestyle.

[a] Numbers may not sum to totals because of missing data.

[b] Adjusted for age (as the time scale), sex, race/ethnicity, marital status (married/living together, never married, separated/divorced, widowed, or missing), education (high school graduate or less, some college, or college/advanced degree), total energy intake (kcal/day; continuous), body mass index (weight (kg)/height $(m)^2$) at age 45 years (<18.5, 18.5–<25.0, 25.0–29.9, ≥30.0, or missing), average alcohol intake at age 45 years (none, <1 drink/day, 1–2 drinks/day, >2 drinks/day, or missing), average physical activity in the 10 years before baseline (MET-hours/week; tertiles), self-rated health (excellent, very good, good, fair, or poor), mammogram in the last 2 years (yes/no), prostate-specific antigen test in the last 2 years (yes/no), sigmoidoscopy in the last 10 years (yes/no), current use of cholesterol-lowering medication (yes/no), aspirin use in the past 10 years (none, low, high, or missing), use of nonaspirin nonsteroidal antiinflammatory drugs in the past 10 years (none, low, high, or missing), smoking (never, 1–12.5 pack-years, 12.6–35.0 pack-years, or >35.0 pack-years), morbidity score,[c] percentage of calories derived from *trans* fat (quartiles), percentage of calories derived from saturated fat (quartiles), number of servings per day of fruits (quartiles), number of servings per day of vegetables (quartiles), years of estrogen therapy (none, <5, 5–9, ≥10, or missing), years of estrogen + progestin therapy (none, <5, 5–9, ≥10, or missing), age at menopause (≤39 years, 40–44 years, 45–49 years, 50–54 years, ≥55 years, or missing), age at death of father (≤59 years, 60–69 years, 70–79 years, 80–89 years, or ≥90 years), and age at death of mother (≤59 years, 60–69 years, 70–79 years, 80–89 years, or ≥90 years).

[c] By using Cox regression, the following conditions, categorized as yes or no, were modeled simultaneously in sex-specific and age-adjusted models to obtain the morbidity score: depression or anxiety; hypertension; a history of lung cancer, colon cancer, bladder cancer, leukemia, non-Hodgkin's lymphoma, pancreatic cancer, melanoma, prostate cancer, breast cancer, cervical cancer, uterine cancer, or ovarian cancer (as separate variables), and all other cancers except nonmelanoma skin cancer combined; ischemic heart disease (defined as history of heart attack, coronary bypass surgery, angioplasty, or diagnosis of angina); stroke; congestive heart failure; rheumatoid arthritis; diabetes; viral hepatitis; cirrhosis of the liver; other chronic liver disease; emphysema, chronic bronchitis, or chronic obstructive pulmonary disease; kidney disease; ulcerative colitis or Crohn's disease; Parkinson's disease; and osteoporosis in women. Depression or anxiety, hypertension, and diabetes were defined as use of medications for these conditions at baseline.

highest quartile of combined EPA + DHA intake from diet was 0.85 (95% CI: 0.76, 0.96; *P* for trend = 0.008). When supplements were included, the adjusted hazard ratio for the highest quartile of total EPA + DHA (from diet plus supplements) was similar, but the trend was slightly stronger (HR = 0.86, 95% CI: 0.77, 0.96; *P* for trend = 0.004). Upon adjustment for arachidonic acid intake, the association between total long-chain ω-3 intake and mortality became stronger (HR = 0.82, 95% CI: 0.73, 0.93; *P* for trend = 0.001). We also examined the association between total EPA + DHA intake and total mortality additionally controlling for arachidonic + linoleic acid; in this alternative model, no difference in the association was observed (data not shown).

We further examined the associations between total EPA + DHA intake (from diet plus supplements) and specific causes of death in the model which included adjustment for arachidonic acid (Table 3). As compared with persons in the lowest quartile of total EPA + DHA intake, those in the highest quartile had reduced risk of mortality from total CVD (HR = 0.87, 95% CI: 0.68, 1.10) and from ischemic heart disease (HR = 0.80, 95% CI: 0.58, 1.10), though both confidence intervals included 1. High intake was associated with a decrease in risk

Table 3. Hazard Ratios for Mortality Associated With Total EPA + DHA Intake (Diet + Supplements), by History of Disease, VITAL Study, 2000–2006[a]

Cause of Death	Quartile of EPA + DHA Intake, g/day										*P* for Trend
	First[b] (0–0.082), No. of Deaths	Second (0.083–0.174)			Third (0.175–0.322)			Fourth (>0.322)			
		No. of Deaths	HR	95% CI	No. of Deaths	HR	95% CI	No. of Deaths	HR	95% CI	
All causes[c]	935	785	0.95	0.86, 1.06	667	0.87	0.78, 0.97	650	0.82	0.73, 0.93	0.001
No history of CVD or cancer at baseline	404	352	0.96	0.83, 1.12	280	0.83	0.70, 0.98	283	0.81	0.67, 0.98	0.011
History of CVD or cancer at baseline	529	433	0.92	0.80, 1.05	383	0.84	0.72, 0.98	362	0.81	0.69, 0.95	0.006
CVD[d]	228	183	0.88	0.71, 1.08	165	0.79	0.63, 0.99	189	0.87	0.68, 1.10	0.158
No history at baseline	129	92	0.77	0.58, 1.02	84	0.78	0.57, 1.05	95	0.91	0.66, 1.27	0.799
History at baseline	98	91	1.01	0.74, 1.37	80	0.81	0.58, 1.14	93	0.82	0.57, 1.17	0.167
Ischemic heart disease[e]	131	105	0.90	0.68, 1.18	96	0.80	0.59, 1.07	110	0.80	0.58, 1.10	0.120
No history at baseline	76	60	0.87	0.60, 1.26	49	0.71	0.47, 1.08	48	0.62	0.39, 0.99	0.029
History at baseline	54	45	0.92	0.61, 1.40	47	0.86	0.55, 1.35	62	0.96	0.60, 1.52	0.812
Cancer[f]	453	390	0.94	0.82, 1.09	332	0.82	0.70, 0.96	304	0.77	0.64, 0.92	0.001
No history at baseline	218	202	0.98	0.80, 1.20	162	0.81	0.65, 1.02	166	0.79	0.62, 1.02	0.027
History at baseline	234	188	0.89	0.72, 1.09	168	0.83	0.66, 1.03	138	0.72	0.56, 0.93	0.010
Lung cancer[g]	142	97	0.81	0.62, 1.06	82	0.72	0.53, 0.97	90	0.77	0.56, 1.07	0.069
No history at baseline	105	76	0.84	0.62, 1.15	63	0.69	0.49, 0.98	79	0.89	0.62, 1.28	0.310
History at baseline	37	21	0.35	0.16, 0.77	18	0.75	0.34, 1.65	11	0.12	0.04, 0.40	0.176
Hematopoietic cancer[h]	48	52	1.11	0.72, 1.72	29	0.79	0.48, 1.32	35	0.84	0.49, 1.45	0.396
No history at baseline	28	35	1.26	0.73, 2.18	20	0.97	0.52, 1.83	20	0.95	0.52, 1.90	0.736
History at baseline	20	17	0.72	0.32, 1.62	9	0.64	0.12, 0.82	14	0.55	0.21, 1.45	0.100
Colorectal cancer[i]	47	30	0.72	0.44, 1.17	24	0.61	0.35, 1.04	30	0.71	0.40, 1.25	0.194
No history at baseline	23	18	0.79	0.41, 1.54	13	0.71	0.34, 1.50	11	0.60	0.25, 1.40	0.220
History at baseline	24	12	0.49	0.21, 1.12	11	0.47	0.20, 1.10	19	0.73	0.31, 1.74	0.414
Breast cancer[j]	38	35	1.12	0.68, 1.85	29	1.07	0.61, 1.86	13	0.66	0.32, 1.36	0.391
No history at baseline	7	6	1.05	0.34, 3.29	6	1.27	0.37, 4.31	2	0.51	0.08, 3.13	0.713
History at baseline	30	29	1.11	0.63, 1.94	23	0.96	0.51, 1.79	11	0.61	0.27, 1.39	0.297
Pancreatic cancer[k]	29	32	1.58	0.91, 2.75	23	1.09	0.58, 2.06	16	0.78	0.36, 1.70	0.505
No history at baseline	26	26	1.17	0.66, 2.08	19	0.95	0.49, 1.86	13	0.65	0.28, 1.51	0.349
History at baseline	3	6	—[l]	—	4	—	—	3	—	—	—

Table continues

of death from cancer (HR = 0.77, 95% CI: 0.64, 0.92; *P* for trend = 0.001), with each of the 5 most common causes of cancer death contributing to the reduced risk. All associations remained in the same direction when risk of mortality was stratified by baseline history of the cause of mortality.

After removing the first 2 years of follow-up, results were very similar to those presented in Tables 2 and 3. The hazard ratios comparing the highest quartiles of total EPA + DHA intake with the lowest were 0.84 (95% CI: 0.72, 0.98) for total mortality, 0.86 (95% CI: 0.64, 1.15) for CVD mortality, 0.79 (95% CI: 0.63, 0.99) for cancer mortality, and 0.86 (95% CI: 0.64, 1.17) for other causes of mortality. Using 3 other approaches to missing data, the results were similar to those for total EPA + DHA in Table 2, and the *P* values for

Table 3. Continued

Cause of Death	Quartile of EPA + DHA Intake, g/day										*P* for Trend
	First[b] (0–0.082), No. of Deaths	Second (0.083–0.174)			Third (0.175–0.322)			Fourth (>0.322)			
		No. of Deaths	HR	95% CI	No. of Deaths	HR	95% CI	No. of Deaths	HR	95% CI	
Other cancers[f]	133	130	1.08	0.84, 1.40	124	1.01	0.77, 1.33	109	0.90	0.67, 1.23	0.485
Other causes[c]	244	205	1.02	0.84, 1.25	161	0.92	0.73, 1.14	150	0.84	0.64, 1.08	0.145

Abbreviations: CI, confidence interval; CVD, cardiovascular disease; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; HR, hazard ratio; MET, metabolic equivalent of task; VITAL, Vitamins and Lifestyle.

[a] Numbers may not sum to totals because of missing data. All models adjusted for age (as the time scale), sex, race/ethnicity, marital status (married/living together, never married, separated/divorced, widowed, or missing), education (high school graduate or less, some college, or college/advanced degree), body mass index (weight (kg)/height $(m)^2$) at age 45 years (<18.5, 18.5–<25.0, 25.0–29.9, ≥30.0, or missing), average physical activity in the 10 years before baseline (MET-hours/week; tertiles), smoking (never, 1–12.5 pack-years, 12.6–35.0 pack-years, >35.0 pack-years, or missing), average alcohol intake at age 45 years (none, <1 drink/day, 1–2 drinks/day, >2 drinks/day, or missing), total energy intake (kcal/day; continuous), number of servings per day of fruits (quartiles), number of servings per day of vegetables (quartiles), dietary intake of arachidonic acid (g/day; continuous), aspirin use in the past 10 years (none, low, high, or missing), use of nonaspirin nonsteroidal antiinflammatory drugs in the past 10 years (none, low, high, or missing), self-rated health (excellent, very good, good, fair, or poor), sigmoidoscopy in the last 10 years (yes/no), mammogram in the last 2 years (yes/no), prostate-specific antigen test in the last 2 years (yes/no), and current use of cholesterol-lowering medication (yes/no).

[b] Reference category (HR = 1).

[c] Results were additionally adjusted for morbidity score (see footnote c of Table 2), percentage of calories derived from *trans* fat (quartiles), percentage of calories derived from saturated fat (quartiles), years of estrogen therapy (none, <5, 5–9, ≥10, or missing), years of estrogen + progestin therapy (none, <5, 5–9, ≥10, or missing), age at menopause (≤39 years, 40–44 years, 45–49 years, 50–54 years, ≥55 years, or missing), age at death of father (≤59 years, 60–69 years, 70–79 years, 80–89 years, or ≥90 years), and age at death of mother (≤59 years, 60–69 years, 70–79 years, 80–89 years, or ≥90 years).

[d] Results were additionally adjusted for history of cardiovascular disease (yes/no; defined as history of heart attack, coronary bypass surgery, angioplasty, stroke, congestive heart failure, or diagnosis of angina) family history of heart attack (number of relatives: 0, 1, or ≥2), current use of blood pressure medication (yes/no), percentage of calories derived from *trans* fat (quartiles), percentage of calories derived from saturated fat (quartiles), years of estrogen therapy (none, <5, 5–9, or ≥10), and years of estrogen + progestin therapy (none, <5, 5–9, or ≥10).

[e] Results were additionally adjusted for history of ischemic heart disease (yes/no; defined as history of heart attack, coronary bypass surgery, angioplasty, or diagnosis of angina), family history of heart attack (number of relatives: 0, 1, or ≥2), current use of blood pressure medication (yes/no), percentage of calories derived from *trans* fat (quartiles), percentage of calories derived from saturated fat (quartiles), years of estrogen therapy (none, <5, 5–9, or ≥10), and years of estrogen + progestin therapy (none, <5, 5–9, or ≥10).

[f] Results were additionally adjusted for history of cancer other than nonmelanoma skin cancer (yes/no), family history of cancer (number of relatives: 0, 1, or ≥2), years of estrogen therapy (none, <5, 5–9, or ≥10), years of estrogen + progestin therapy (none, <5, 5–9, or ≥10), age at menopause (≤39, 40–44, 45–49, 50–54, or ≥55 years), age at menarche (≤11, 12, 13, or ≥14 years), and number of servings of red/processed meat per week (quartiles).

[g] Results were additionally adjusted for history of lung cancer (yes/no), family history of lung cancer (number of relatives: 0, 1, or ≥2), history of emphysema, chronic bronchitis, or chronic obstructive pulmonary disease (yes/no), years of smoking, and pack-years squared.

[h] Results were additionally adjusted for history of leukemia/lymphoma (yes/no), family history of leukemia/lymphoma (number of relatives: 0, 1, or ≥2), history of any cancer (yes/no), history of rheumatoid arthritis (yes/no), and history of fatigue/lack of energy (yes/no).

[i] Results were additionally adjusted for history of colorectal cancer (yes/no), family history of colorectal cancer (number of relatives: 0, 1, or ≥2), history of diabetes (yes/no), years of estrogen therapy (none, <5, 5–9, or ≥10), years of estrogen + progestin therapy (none, <5, 5–9, or ≥10), calcium intake from diet, calcium intake from supplements, and number of servings of red/processed meat per week (quartiles).

[j] Results were additionally adjusted for history of breast cancer (yes/no), family history of breast cancer (number of relatives: 0, 1, or ≥2), years of estrogen therapy (none, <5, 5–9, or ≥10), years of estrogen + progestin therapy (none, <5, 5–9, or ≥10), age at first birth, history of hysterectomy (none, simple, total), age at menopause (≤39, 40–44, 45–49, 50–54, or ≥55 years), and age at menarche (≤11, 12, 13, or ≥14 years).

[k] Results were additionally adjusted for history of pancreatic cancer (yes/no), family history of pancreatic cancer (number of relatives: 0, 1, or ≥2), number of servings of red/processed meat per week (quartiles), and history of diabetes (yes/no).

[l] Hazard ratios and 95% confidence intervals were not calculated because of small numbers of deaths.

trend were all 0.05 or less). Using the residual method of adjustment for total energy did not affect results (data not shown). There was no evidence of interaction between long-chain ω-3 PUFAs and sex, smoking, body mass index, or nonsteroidal antiinflammatory drug use (data not shown).

DISCUSSION

In this prospective cohort study of US men and women, we found that greater consumption of the long-chain ω-3 PUFAs EPA and DHA was associated with an 18% reduced risk of mortality from all causes and a 23% decrease in mortality from cancer. There was a 13% nonsignificant decrease in mortality from CVD, which contributed to the total mortality risk reduction. There was no clear trend, however, so our results add little to the substantial literature on this topic (8). There was no effect modification of the association between total EPA + DHA and total mortality by sex, body mass index, smoking, or nonsteroidal antiinflammatory drug use.

Findings from randomized trials of ω-3 supplements and total mortality have been inconsistent (18). Two recent meta-analyses examining ω-3 supplements in secondary prevention of CVD (7, 19) found no evidence of a reduction in total mortality risk (Kwak et al. (7): relative risk (RR) = 0.99, 95% CI: 0.89, 1.09; Rizos et al. (19): RR = 0.96, 95% CI: 0.91, 1.02). However, the analysis by Kwak et al. (7) excluded 2 large open-label studies, the Japan Eicosapentaenoic Acid Lipid Intervention Study (20) and the Gruppo Italiano per lo Studio della Sopravvivenza nell'Infarto Miocardico trial (21), which both showed a protective association of supplement use with mortality. Several recent trials of ω-3 PUFA supplementation have failed to show an effect on mortality (22–24). However, Mozaffarian and Wu (8) have argued that these studies were of limited duration (2–4.2 years) and may have lacked the statistical power needed to detect a modest association. A 2009 Cochrane review of randomized controlled trials of ω-3 supplements found a protective effect on risk of total mortality (RR = 0.87, 95% CI: 0.73, 1.03; *P* for heterogeneity = 0.04) (6). The protective association with total mortality was seen in trials of patients at high risk of CVD (RR = 0.84, 95% CI: 0.70, 1.02) but not in those at low risk (RR = 1.07, 95% CI: 0.70, 1.64) (6). In the SU.FOL.OM3 (Supplementation With Folate, Vitamins B_6 and B_{12}, and/or Omega-3 Fatty Acids) trial, Andreeva et al. (5) reported a nonsignificantly increased risk of cancer mortality associated with ω-3 supplement use (with or without B vitamin supplements) (HR = 1.47, 95% CI: 0.87, 2.48), but this was based on a small number of deaths.

Few observational studies have examined the association between long-chain ω-3 PUFAs and mortality, and findings have been inconsistent. The Iowa Women's Health Study examined associations for reported long-chain ω-3 intake from fish in a cohort of CVD- and cancer-free older women, with mortality outcomes similar to those of our study—cancer, CVD, and total. The investigators found little evidence of association with total mortality (RR = 0.93, 95% CI: 0.83, 1.05), cancer mortality (RR = 0.91, 95% CI: 0.75, 1.11), or CVD mortality (RR = 0.95, 95% CI: 0.78, 1.15) (10). That study did not include measurement of ω-3 supplementation, and dietary changes over the long period of follow-up may have attenuated a modest association. In contrast to the Iowa Women's Health Study results but similar to our results, Mozaffarian et al. (25) recently reported a 27% reduction in total mortality among persons in the highest quintile of plasma ω-3 PUFAs versus the lowest (*P* for trend < 0.001).

Most studies of long-chain ω-3 PUFA intake and cancer have examined cancer incidence rather than mortality. In the Cochrane review, which examined trials and observational studies, no association was reported between ω-3 supplements and total cancer outcomes (RR = 1.07, 95% CI: 0.88, 1.30) (6). Similarly, a study of male US physicians found no association between dietary long-chain ω-3 intake and total cancer incidence (RR = 1.03, 95% CI: 0.86, 1.23) (26). In a 2007 meta-analysis, Geelen et al. (27) reported an inverse association between fish intake and incidence of colorectal cancer (RR = 0.88, 95% CI: 0.78, 1.00) but no association with colorectal cancer mortality. Studies of fish or ω-3 intake and incidence of breast, lung, pancreatic, and blood cancers have been inconsistent (9, 28–30).

If long-chain ω-3 PUFAs indeed reduce mortality from cancer, it is likely to be through antiinflammatory pathways. Evidence from in vitro studies indicates that long-chain ω-3 PUFAs act to reduce inflammation in several ways, including inhibition of nuclear factor κ B, a transcription factor central to the inflammatory cascade (4, 31). Long-chain ω-3 PUFAs have also been shown to competitively inhibit proinflammatory ω-6 PUFAs (32). By competing with ω-6 PUFAs such as arachidonic acid for cyclooxygenase activity and storage in the cell membrane, ω-3 PUFAs act to block the production of prostaglandin E_2, a proinflammatory ω-6-derived eicosanoid (32). Animal research has further demonstrated that diets rich in long-chain ω-3 PUFAs reduce prostaglandin E_2 production in rodents (9, 31). In support of this growing body of experimental evidence, epidemiologic studies have observed inverse associations between ω-3 PUFA intake and concentrations of several inflammatory markers, including C-reactive protein, tumor necrosis factor α, and interleukin-6 (8, 33). Despite inconsistent early trials (6), 2 recent large trials have also demonstrated that supplementation with long-chain ω-3 PUFAs reduces circulating levels of C-reactive protein and tumor necrosis factor α (34, 35). In addition to reducing inflammation, long-chain ω-3 PUFAs may reduce cancer mortality through other anticancer mechanisms. In vitro and animal studies have demonstrated that EPA and DHA may not only have antiproliferative, proapoptotic, and antiangiogenic properties but may also act to inhibit metastasis (31).

Our study had a number of strengths. To our knowledge, it was the first to prospectively examine the combined association of dietary ω-3 intake and fish oil supplements with cause-specific mortality. The questionnaire was specifically designed to ascertain supplement use and included detailed assessment of frequency and years of supplement use. Special efforts were made in the design of the FFQ to capture dietary intake of specific fatty acids. Our study also included adjustment for numerous health behaviors and existing health conditions, including conditions that are common indications for fish-oil supplement use or that may lead people to increase their dietary consumption of ω-3-rich fish. Nonetheless, estimates of risk may still be residually confounded by unmeasured factors. Other limitations include the possibility of selection bias if participants who consumed high levels of ω-3 PUFAs and were more (or less) healthy elected to join the study, as well as the possibility of reverse causality if poor health influenced participants to change their diet or supplement use. We addressed these issues by adjusting for numerous factors associated with ω-3 intake and mortality, addressing potential confounding by indication for supplement use as well as healthy user bias. We also addressed selection bias and reverse causality by presenting results stratified according to whether participants had the condition at baseline in the cause-specific mortality analyses. To further address reverse causality, we analyzed the data with the first 2 years of follow-up omitted, finding no meaningful differences in the associations.

Measurement error might have also affected our results. An earlier version of our FFQ was validated against 8 days of diet records, with a Pearson correlation coefficient (deattenuated) for correlation between the two methods of 0.59 for EPA, suggesting good validity of self-reported ω-3 PUFAs

in our study; however, DHA was not assessed (15). In a review of studies comparing ω-3 intake as measured by other FFQs with biomarker measurements, Overby et al. (36) found summary correlations of 0.42 for EPA and 0.44 for DHA. A validity study conducted to compare supplement use as ascertained from the VITAL baseline questionnaire with an in-home supplement inventory and with nutrient biomarkers found excellent validity for a range of supplements (15). Use of ω-3 supplements was not specifically evaluated, and for these the effect of imputing dose is not known. These errors in ascertainment of ω-3 intake from diet and supplements would probably be nondifferential, which would usually (but not always) bias the estimates towards the null (37, 38). Another limitation is that diet and supplement use were ascertained at only 1 point in time. Increased awareness of positive health benefits of ω-3 consumption may have led to changes in ω-3 intake over time, introducing additional error into the measurement.

In summary, we found in our prospective cohort study that higher intake of long-chain ω-3 PUFAs from diet and supplements was associated with decreased risks of total mortality and cancer mortality. Our results for total mortality are supported by some trials that have demonstrated benefits from ω-3 supplementation, but not by most trials, while there is less information about the effects of ω-3 intake on mortality from cancer. The upcoming Vitamin D and Omega-3 Trial, which is currently enrolling participants, should provide useful data concerning the health effects of ω-3 supplementation for primary prevention (39).

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