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Effects of seafood consumption and toenail mercury and selenium levels on cognitive function among American adults: 25 y of follow up

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ABSTRACT

Objectives: The aim of this study was to examine the longitudinal association between seafood and intake of long-chain ω-3 polyunsaturated fatty acids (LCω-3 PUFA) and cognitive function and to explore the possible effect modifications owing to mercury (Hg) and selenium (Se) levels.
Methods: Participants (N = 3231) from the CARDIA (Coronary Artery Risk Development in Young Adults) study underwent baseline examination and were reexamined in eight follow-up visits. Diet was assessed at baseline and in exam years 7 and 20. Toenail Hg and Se were measured at exam year 2. Cognitive function was measured at exam year 25 using three tests: Rey Auditory Verbal Learning Test (RAVLT), Digit Symbol Substitution Test (DSST), and the Stroop test. The general linear regression model was used to examine cumulative average intakes of LCω-3 PUFA and seafood in relation to the cognitive test scores; and to explore the possible effect modifications caused by Hg and Se.
Results: LCω-3 PUFA intake was significantly associated with better performance in the DSST test (quintile 5 versus quintile 1; mean difference = 1.74; 95% confidence interval, 0.19–3.29; P_{trend}, 0.048]), but not in the RAVLT and Stroop tests. Similar results were observed for intakes of eicosapentaenoic acid, docosahexaenoic acid, and non-fried seafood. The observed associations were more pronounced in participants with body mass index ≥25 kg/m^2, but not significantly modified by toenail Hg or Se.
Conclusion: This longitudinal study supported the hypothesis that LCω-3 PUFA or non-fried seafood intake is associated with better cognitive performance in psychomotor speed among US adults, especially those who are overweight or obese.

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Introduction

Cognitive dysfunction is a common outcome of aging and may lead to dementia, which is an irreversible disorder. As cognitive decline begins in middle age and progresses slowly during the decades before a diagnosis of clinical dementia [1], its prevention at early stages is of great medical and economic importance.

Long-chain ω-3 polyunsaturated fatty acids (LCω-3 PUFA) from seafood may play an important role in maintaining cognitive function by reducing inflammation [2]. However, epidemiologic data are inconsistent and non-conclusive. Two systematic reviews among randomized clinical trials (RCTs) concluded that no sufficient evidence supports the beneficial effects of LCω-3 PUFA supplementation on cognitive decline, but most of the RCTs included in the systematic reviews had small sample sizes (<100 participants) with short intervention durations (<5 mo) [3,4]. On the other hand, observational studies supported benefits of long-term intakes of LCω-3 PUFA and seafood on age-related cognitive decline [5,6]. Of note, most of the previous studies focused on older adults and those of young adults are limited. As cognition declines with aging and young adults are under the risk development stage, following up a cohort of young adults to midlife or later will provide insight on the natural history or etiology of cognitive decline.

When examining LCω-3 PUFA intake and cognitive decline, mercury (Hg) and selenium (Se) are two elements that should be considered as they often coexist with LCω-3 PUFA in seafood. Hg has potential neurotoxicity [7], while Se may be neuroprotective due to its antioxidative and anti-inflammatory properties [8] or by modulating the neurotoxicity effect of Hg [9]. It has been hypothesized that the benefit of LCω-3 PUFA or seafood on cognitive function may be more pronounced in the setting of high Se and low Hg concentrations. However, to our knowledge, no study has examined the potential three-way interaction.

Therefore, we analyzed the data from a cohort of young adults with 25 y of follow up to prospectively examine the long-term associations between LCω-3 PUFA intake and seafood consumption with cognitive performance. We hypothesized that higher intakes of LCω-3 PUFA and seafood are associated with better cognitive function, especially in the setting of high Se and low Hg levels.

Method

Study population

The CARDIA (Coronary Artery Risk Development in Young Adults) study is a multicenter, ongoing longitudinal cohort study that recruited 5115 biracial male and female young adults initially 18 to 30 y of age in 1985–1986 from four study centers. The participants underwent baseline examination and were re-examined in eight follow-up visits at exam years 2, 5, 7, 10, 15, 20, 25, and 30. The detailed design and methods of the CARDIA study are published elsewhere [10].

Among the 3499 participants who remained at exam year 25, information was available on cognitive function measurements for 3316. Of these participants, 85 were sequentially excluded because of missing data on baseline LCω-3 PUFA or seafood (n = 2), report of an extreme energy intake (<600 or >6000 kcal/d for women; <800 or >8000 kcal/d for men; n = 13), and experience of a stroke event during the follow-up period (n = 70). Thus, the final database included 3231 participants. Written informed consent was provided by all of the participants. The study design, data collection, and analyses were approved by the institutional review boards of the CARDIA participating institutions.

Seafood consumption and LCω-3 PUFA intake

Dietary intake information was collected using the interviewer-administered CARDIA Diet History questionnaire at baseline and in exam years 7 and 20. The CARDIA questionnaire has been evaluated and discussed elsewhere [11,12]. In brief, participants were asked about their diet habits in the previous 30 d, including frequency, amount of food, and food preparation methods. Because preparation method, particularly frying, may substantially alter the fatty acid content of a seafood meal [13], seafood consumption was divided into fried and non-fried seafood intake groups. Fried seafood consumption included intakes of fried fish and fried shellfish from commercial and fast food. Non-fried seafood consumption was the sum of fresh, smoked, lean, and shellfish intakes. Because the distribution of fried seafood consumption was extremely skewed and relatively narrow, it was considered as a covariate when examining non-fried seafood, but not as an exposure of interest. Nutrient intake was estimated using an adaptively updated nutrient database from the Nutrition Coordinating Center at the University of Minnesota. Information on supplementation use also was collected. In this study, LCω-3 PUFA intake was defined as the sum of eicosapentaenoic acid (EPA), docosapentaenoic acid (DPA), and docosahexaenoic acid (DHA) intakes from diet and supplementation. Because of the relatively small amount and the narrow distribution, DPA was not analyzed as a separate exposure in the analysis. To reduce the measurement errors caused by within-person variation and to best represent the long-term dietary intakes, cumulative average daily intakes of LCω-3 PUFA and seafood, which were calculated by averaging the corresponding measurements at baseline and in exam years 7 and 20, were used in the analyses. A modified a priori diet quality score was calculated to measure the quality of diet based on other food groups at baseline and in exam years 7 and 20 [14]. The average of the three measurements was used in the analyses.

Cognitive function assessments

Cognitive function was measured in exam year 25 using three tests: the Rey Auditory Verbal Learning Test (RAVLT), the Digit Symbol Substitution Test (DSST), and the Stroop test. All tests were administered by trained and certified CARDIA research technicians following a standardized protocol. RAVLT examined verbal learning and memory by assessing the ability to correctly memorize and recall 15 words after a 10-min delay. More words recalled (corresponds to a higher score; possible range: 0–15) indicated better cognitive performance. DSST from the Wechsler Adult Intelligence Scale II measured psychomotor speed, sustained attention, and working memory [15]. The possible scores ranged from 0 to 133, with a higher score indicating better cognitive performance. The Stroop test evaluated executive function by assessing the ability to view a complex visual stimulus and to respond while suppressing the responses to another dimension [16,17]. The test was scored by the time it took to complete the trials plus the number of errors, thus a higher score indicated worse cognitive performance.

Other variables

Toenail clippings from all 10 toes were collected at the exam year 2 follow-up visit and shipped to a central laboratory at the University of Missouri Research Reactor [18]. Toenail Hg and Se were measured using instrumental neutron-activation analysis [19]. Plasma high-density lipoprotein cholesterol (HDL-C) and triacylglycerol (TG) concentrations were measured from baseline to exam year 30 by enzymatic methods at Northwest Lipids Research Laboratory (Seattle, WA, USA). Plasma low-density lipoprotein cholesterol (LDL-C) concentration was estimated by the Friedewald equation. Fasting glucose was measured at baseline and in exam years 7, 10, 15, 20, 25, and 30 using hexokinase coupled to glucose-6-phosphate dehydrogenase (Millipore, Inc, Bellerica, MA, USA; later at the University of Minnesota) [10]. The cumulative average of repeated measurements across all available examinations of HDL-C, LDL-C, TG, and fasting glucose by exam year 25 when cognitive function was examined were used in the analyses. The cumulative average daily intakes of total energy, B_6, B_{12}, and folate were calculated by using the repeated measurements at baseline and exam years 7 and 20 through CARDIA questionnaire. In addition, some other important covariates were collected at each follow-up visit. Demographic and lifestyle information, including age, sex, race (black or white), study center, education levels, smoking status, alcohol consumption, and physical activity were collected through a self-administered questionnaire and were verified in clinic examinations. Education attained through exam year 25 has three levels (<12, 12–15.9, or ≥16 y). Smoking status at exam year 25 was classified into three groups: never, former, or current smokers. Alcohol consumption was presented as mL/d of alcohol. Physical activity was assessed using the CARDIA Physical Activity History Questionnaire. Body weight and height were measured to calculate body mass index (BMI). Blood pressure was measured by using a random zero sphygmanometer from baseline to exam year 15 and the Omron HEM907 XL sphygmomanometer (Omron Corporation, Kyota, Japan) at exam years 20, 25, and 30 [20]. A calibration study was performed at exam year 20 in a subgroup of participants. Cumulative average alcohol intake, physical activity, BMI, and blood pressure by exam year 25 were calculated by averaging the repeated measurements.

Statistical analysis

Characteristics of participants were summarized using mean values with standard deviations (SDs) or medians with interquartile ranges for continuous variables and proportions for categorical variables. Analysis of variance (ANOVA), the Kruskal–Wallis test, or the χ^2 test, as appropriate, were used to test for the differences

Table 1
Characteristics of the study population by quintiles (Q) of cumulative average daily intake of LCω-3 PUFA: The CARDIA study (N = 3231), 1985 to 2010*,†

Characteristics	Quintiles of LCω-3 PUFA intake					Total (N = 3231)	*P*-value
	Q1 (n = 630)	Q2 (n = 650)	Q3 (n = 663)	Q4 (n = 637)	Q5 (n = 651)		
LCω-3 PUFA (g/d)	0.04 ± 0.02	0.08 ± 0.01	0.13 ± 0.02	0.19 ± 0.03	0.41 ± 0.22	0.17 ± 0.17	NA
DHA (g/d)	0.02 ± 0.01	0.04 ± 0.01	0.06 ± 0.02	0.09 ± 0.02	0.20 ± 0.11	0.08 ± 0.08	NA
EPA (g/d)	0.01 ± 0.01	0.03 ± 0.01	0.05 ± 0.01	0.07 ± 0.02	0.16 ± 0.11	0.06 ± 0.07	NA
Non-fried seafood (servings/d)	0.24 ± 0.43	0.50 ± 0.29	0.80 ± 0.50	1.19 ± 0.62	2.04 ± 1.25	0.96 ± 0.94	NA
Fried seafood (servings/d)	0.04 ± 0.18	0.05 ± 0.17	0.06 ± 0.19	0.10 ± 0.35	0.08 ± 0.32	0.07 ± 0.26	NA
Total energy (kcal/d)	2348.7 ± 1074.6	2501.1 ± 1126.7	2683.6 ± 1237.2	2768.7 ± 1098.8	3227.4 ± 1280.8	2707.9 ± 1204.4	<0.01
Age at exam Y25 (y)	49.6 ± 3.8	50 ± 3.6	50.1 ± 3.7	50.6 ± 3.5	50.5 ± 3.6	50.1 ± 3.6	<0.01
Female (%)	64	60.2	56.6	54.6	46.7	56.4	<0.01
Black (%)	38.9	42.2	47.4	44.3	52.4	45.1	<0.01
Education attained through exam Y25 (y)	14.8 ± 2.7	15.1 ± 2.7	15.1 ± 2.7	15.4 ± 2.5	15.3 ± 2.6	15.1 ± 2.7	<0.01
Smoking status (%)							0.02
Never smoker	63.7	63.6	62.6	61.5	57.8	61.8	
Former smoker	18.6	18.8	22.5	23.8	26.4	22.1	
Current smoker	17.7	17.6	14.9	14.7	15.8	16.1	
Alcohol consumption (mL/d)							<0.01
Median	3.7	4.1	4.8	7.5	7.8	5.5	
IQR	0.3–11.5	0.4–12.3	0.9–13.8	1.8–16.3	2.1–21.3	0.9–15	
Physical activity (exercise unit)							<0.01
Median	265.1	305.8	321.3	334.8	386.3	323.8	
IQR	161.1–414.2	191.9–458.5	198.8–476.9	213.1–481.1	254–573.9	200.4–479.1	
BMI (%)							0.12
<18.5 kg/m^2	0.5	1.1	0.9	0.5	0.3	0.7	
18.5–24.9 kg/m^2	40.2	43.1	36.1	44	39.9	40.6	
25–29.9 kg/m^2	33.7	28.8	39.4	35.6	36.6	34.8	
≥30 kg/m^2	25.7	27.1	23.7	20.0	23.2	23.9	
HDL cholesterol (mg/dL)	53.1 ± 12.5	53 ± 13	53.2 ± 11.9	53.9 ± 12.7	54.6 ± 13.6	53.6 ± 12.8	0.08
LDL cholesterol (mg/dL)	110.7 ± 25.8	109.2 ± 26.3	110.3 ± 26.1	110.5 ± 25.7	113 ± 26.3	110.7 ± 26	0.12
Triacylglycerols (mg/dL)	89 ± 40.7	93.3 ± 62.1	89.7 ± 51.2	89.7 ± 51.1	94.6 ± 63.2	91.3 ± 54.4	0.24
Glucose (mg/dL)	89.3 ± 12.8	90.5 ± 14.4	89.7 ± 12.9	89.7 ± 10.5	91.3 ± 15.2	90.1 ± 13.3	0.048
SBP (mm Hg)	110.5 ± 9.9	111.2 ± 9.5	111.6 ± 9.3	111 ± 9.9	111.8 ± 10.1	111.2 ± 9.7	0.14
DBP (mm Hg)	70.3 ± 7.3	71 ± 7.3	71.1 ± 7	70.7 ± 7.6	71.1 ± 7.7	70.8 ± 7.4	0.22
Toenail selenium at exam Y2 (ppm)	0.88 ± 0.15	0.87 ± 0.16	0.86 ± 0.16	0.85 ± 0.13	0.84 ± 0.15	0.86 ± 0.15	<0.01
Toenail mercury at exam Y2 (ppm)	0.22 ± 0.25	0.26 ± 0.26	0.30 ± 0.32	0.37 ± 0.41	0.43 ± 0.50	0.32 ± 0.37	<0.01

BMI, body mass index; CARDIA, Coronary Artery Risk Development in Young Adults; DBP, diastolic blood pressure; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid; HDL, high-density lipoprotein; IQR, interquartile range; LCω-3 PUFA, long-chain ω-3 polyunsaturated fatty acid; LDL, low-density lipoprotein; NA, not applicable; SBP, systolic blood pressure

*All variables are cumulative averages of all available observations of the variable in question, except where noted. Results are presented by means ± SD, medians (IQRs), or proportions.

†*P*-values are for any differences across quintiles of LCω-3 PUFA intake (analysis of variance, Kruskal–Wallis test, or χ^2 test, as appropriate).

across quintiles of LCω-3 PUFA intake. Covariates related to cognitive function and associated with LCω-3 PUFA intake were considered in the main analyses.

The general linear regression model was used to examine the associations between intake of LCω-3 PUFA, DHA, EPA, or non-fried seafood and cognitive test scores. The mean differences of each cognitive test score with the corresponding 95% confidence intervals (CIs) using the lowest quintile of each exposure as the referent were estimated with adjustment for potential confounders (models 1 and 2). Linear trends were tested by using the continuous variable of the exposure with values >99th percentile excluded.

Several sensitivity analyses were conducted to test the robustness of the findings. First, only participants with all repeated measurements of LCω-3 PUFA, DHA, EPA, and non-fried seafood were included in the analysis. Second, shellfish was excluded from seafood consumption owing to its relatively low levels of LCω-3 PUFA. Third, model 2 was further adjusted for toenail Hg and Se levels and the dietary intakes of total energy, B_6, B_{12}, and folate to explore possible confounding effects from other nutrients and contaminants in seafood. Fourth, model 2 was additionally adjusted for the modified a priori diet quality score to reduce the influence of the diet quality based on other food groups.

In stratified analyses, the associations between intakes of LCω-3 PUFA and cognitive test scores using tertiles or the continuous variable for LCω-3 PUFA were estimated by stratifying age at exam year 25 (<median 51 y versus ≥median 51 y), sex (female versus male), race (black versus white), BMI (<25 versus ≥25 kg/m^2), smoking status (never- versus ever-smokers), toenail Se levels (<median 0.85 versus ≥median 0.85 ppm), and toenail Hg levels (<median 0.22 versus ≥median 0.22 ppm) with adjustment for all covariates in model 2 except the potential effect modifier. To further explore the possible joint modification by Hg and Se, we examined the association of interest in four subgroups defined by the median levels of Hg (0.22 ppm) and Se (0.85 ppm). Interaction was tested using the continuous variable of LCω-3 PUFA with values >99th percentile excluded.

All analyses were performed by using SAS version 9.4 (SAS Institute, Cary, NC, USA). Two- sided $P \leq 0.05$ (main effect) and $P \leq 0.10$ (interaction) were considered statistically significant.

Results

In the study population (N = 3231), 56% of participants were women and 45% were black, with an average age of 50 y at exam year 25. Table 1 shows the characteristics of the study population across quintiles of LCω-3 PUFA intake. Participants with higher LCω-3 PUFA intake were more likely to be older, male, and black; have higher total energy intake and education level; have higher alcohol consumption,; be physically active; and have higher levels of fasting glucose and toenail Hg but lower levels of toenail Se. They were less likely to be current smokers. Higher LCω-3 PUFA intake was significantly associated with a better cognitive performance measured by DSST (Table 2). Compared with participants in the lowest quintile (Q1) of LCω-3 PUFA intake, those in the highest quintile (Q5) had 2.79 more points in DSST (95% CI, 1.19– 4.40; $P_{trend} < 0.01$; model 1), and 1.42 fewer points in the Stroop test (95% CI, −2.53 to −0.31; $P_{trend} = 0.013$; model 1). The observed associations were attenuated to some extent after further adjustment for other confounding variables in model 2, but remained statistically significant for DSST (Q5 versus Q1; mean difference: 1.74; 95% CI, 0.19–3.29; $P_{trend} = 0.048$).

Table 2
Multivariable-adjusted mean differences (95% CI) in cognitive test scores according to quintiles (Q) of cumulative average daily intakes of LCω3 PUFA, DHA, EPA, and non-fried seafood: The CARDIA study (N = 3231), 1985 to 2010[*,†,‡,§]

	RAVLT words Mean 8.7 (SD 2.6)		DSST symbols Mean 70.4 (SD 16)		Stroop test points Mean 22.7 (SD 10.6)	
	Model 1	Model 2	Model 1	Model 2	Model 1	Model 2
LCω-3 PUFA (g/d)						
Q1 (≤0.06)	0 (Ref.)	0 (Ref.)	0 (Ref.)	0 (Ref.)	0 (Ref.)	0 (Ref.)
Q2 (0.07 to 0.10)	0.15 (−0.10 to 0.40)	0.12 (−0.13 to 0.36)	0.21 (−1.35 to 1.78)	−0.12 (−1.60 to 1.36)	−0.92 (−2.00 to 0.16)	−0.77 (−1.84 to 0.30)
Q3 (0.11 to 0.15)	0.09 (−0.16 to 0.34)	0.02 (−0.23 to 0.27)	1.06 (−0.51 to 2.62)	0.28 (−1.21 to 1.77)	0.04 (−1.04 to 1.12)	0.46 (−0.61 to 1.53)
Q4 (0.16 to 0.25)	0.09 (−0.17 to 0.35)	−0.03 (−0.28 to 0.22)	1.35 (−0.24 to 2.94)	0.02 (-−1.50 to 1.54)	−1.10 (−2.20 to −0.003)	−0.39 (−1.49 to 0.70)
Q5 (>0.25)	0.17 (−0.09 to 0.43)	0.08 (−0.17 to 0.34)	2.79 (1.19 to 4.40)	1.74 (0.19 to 3.29)	−1.42 (−2.53 to −0.31)	−0.76 (−1.88 to 0.36)
P_{trend}	0.38	0.84	<0.01	0.048	0.013	0.21
DHA (g/d)						
Q1 (≤0.03)	0 (Ref.)	0 (Ref.)	0 (Ref.)	0 (Ref.)	0 (Ref.)	0 (Ref.)
Q2 (0.04−0.05)	0.26 (0.005 to 0.51)	0.26 (0.02 to 0.50)	−0.54 (−2.10 to 1.02)	−0.55 (−2.02 to 0.93)	−0.39 (−1.47 to 0.70)	−0.29 (−1.35 to 0.78)
Q3 (0.06−0.07)	0.13 (−0.12 to 0.38)	0.07 (−0.18 to 0.31)	1.26 (−0.30 to 2.81)	0.59 (−0.89 to 2.06)	−0.25 (−1.33 to 0.82)	0.10 (−0.97 to 1.17)
Q4 (0.08−0.12)	0.18 (−0.08 to 0.43)	0.04 (−0.21 to 0.29)	1.55 (−0.02 to 3.13)	0.21 (−1.29 to 1.71)	−1.00 (−2.09 to 0.09)	−0.26 (−1.35 to 0.82)
Q5 (>0.12)	0.24 (−0.01 to 0.50)	0.15 (−0.11 to 0.40)	2.80 (1.21 to 4.38)	1.75 (0.22 to 3.28)	−1.50 (−2.60 to −0.40)	−0.82 (−1.93 to 0.28)
P_{trend}	0.09	0.41	<0.01	0.051	<0.01	0.16
EPA (g/d)						
Q1 (≤0.02)	0 (Ref.)	0 (Ref.)	0 (Ref.)	0 (Ref.)	0 (Ref.)	0 (Ref.)
Q2 (0.03−0.04)	0.04 (−0.21 to 0.29)	−0.06 (−0.30 to 0.19)	1.23 (−0.33 to 2.78)	0.43 (−1.05 to 1.91)	−1.49 (−2.56 to −0.41)	−1.12 (−2.19 to −0.06)
Q3 (0.05−0.06)	0.16 (−0.10 to 0.41)	0.09 (−0.16 to 0.33)	0.99 (−0.59 to 2.57)	0.18 (−1.32 to 1.68)	−0.72 (−1.81 to 0.37)	−0.29 (−1.37 to 0.79)
Q4 (0.07−0.09)	0.07 (−0.19 to 0.32)	−0.07 (−0.32 to 0.18)	2.12 (0.53 to 3.71)	0.69 (−0.83 to 2.22)	−1.58 (−2.69 to −0.48)	−0.84 (−1.94 to 0.26)
Q5 (>0.09)	0.07 (−0.19 to 0.33)	−0.01 (−0.27 to 0.24)	2.51 (0.90 to 4.11)	1.50 (−0.06 to 3.05)	−1.63 (−2.74 to −0.52)	−0.99 (−2.11 to 0.13)
P_{trend}	0.35	0.68	<0.01	0.02	0.015	0.15
Non-fried seafood (servings/d)						
Q1 (≤0.29)	0 (Ref.)	0 (Ref.)	0 (Ref.)	0 (Ref.)	0 (Ref.)	0 (Ref.)
Q2 (0.30−0.55)	0.34 (0.09 to 0.59)	0.21 (−0.03 to 0.46)	1.68 (0.12 to 3.24)	0.65 (−0.83 to 2.13)	−1.44 (−2.52 to −0.36)	−0.96 (−2.02 to 0.11)
Q3 (0.56−0.88)	0.21 (−0.04 to 0.47)	0.09 (−0.16 to 0.33)	−0.04 (−1.61 to 1.52)	−1.45 (−2.94 to 0.04)	−0.38 (−1.47 to 0.70)	0.24 (−0.83 to 1.32)
Q4 (0.89−1.43)	0.28 (0.03 to 0.54)	0.13 (−0.12 to 0.38)	1.28 (−0.30 to 2.85)	−0.25 (−1.75 to 1.26)	−1.36 (−2.45 to −0.27)	−0.56 (−1.65 to 0.52)
Q5 (>1.43)	0.11 (−0.14 to 0.37)	−0.01 (−0.27 to 0.24)	2.87 (1.29 to 4.45)	1.48 (−0.05 to 3.01)	−1.72 (−2.81 to −0.62)	−0.97 (−2.08 to 0.14)
P_{trend}	0.58	0.96	<0.01	0.04	<0.01	0.13

BMI, body mass index; CARDIA, Coronary Artery Risk Development in Young Adults; DHA, docosahexaenoic acid; DSST, Digit Symbol Substitution Test; EPA, eicosapentaenoic acid; LCω-3 PUFA, long-chain ω-3 polyunsaturated fatty acids; RAVLT, Rey Auditory Verbal Learning Test

*All models were constructed using general linear model analysis. P_{trend} was examined by using the continuous variable of exposure with values >99th percentile excluded.

†Model 1 was adjusted for age, sex, race (white or black), and study center.

‡Model 2 was additionally adjusted for educational attainment through exam year 25 (<12, 12−15.9, or ≥16 y), cumulative average BMI (<18.5, 18.5−24.9, 25−29.9, or ≥30 kg/m^2), smoking status at exam year 25 (never, former, or current smokers), cumulative average alcohol consumption (0, 0.1−11.9, 12−23.9 or ≥24 mL/d), cumulative average physical activity (quintiles), and cumulative average glucose level (continuous).

§Fried seafood intake (yes or no) was adjusted for in both models 1 and 2 when studying non-fried seafood consumption.

No statistically significant association was found between LCω-3 PUFA intake and the RAVLT score. The findings persisted when examining DHA and EPA separately (Table 2).

Similar results were observed for non-fried seafood consumption (Table 2). Higher non-fried seafood consumption was associated with better performances in DSST (Q5 versus Q1; mean difference: 2.87; 95% CI, 1.29−4.45; $P_{trend} < 0.01$; model 1) and the Stroop test (Q5 versus Q1; mean difference: −1.72; 95% CI, -−2.81 to −0.62; $P_{trend} < 0.01$; model 1). After adjusting for other confounding variables in model 2, the significant findings remained for DSST (Q5 versus Q1; mean difference: 1.48; 95% CI, −0.05 to 3.01; $P_{trend} = 0.04$). Non-fried seafood consumption was not associated with performance measured by the RAVLT.

In a sensitivity analysis, when only including participants who have data in all repeated measurements of LCω-3 PUFA, DHA, EPA, and non-fried seafood, the results were not significantly changed. In addition, the results were generally consistent when excluding shellfish from seafood consumption or further adjusting for toenail Hg and Se levels. The observed associations were not significantly changed when additionally adjusting for total energy intake and other nutrients in seafood. The associations were attenuated when adjusting for the modified a priori diet quality score (data not shown).

Because intakes of LCω-3 PUFA and non-fried seafood were consistently associated with cognitive performance measured by DSST, we conducted stratified analyses based on DSST by a few prespecified factors (Table 3). A significant interaction was observed between LCω-3 PUFA intake and BMI ($P_{interaction} < 0.01$); the association of interest was more pronounced in participants with BMI ≥25 kg/m^2 (tertile 3 versus tertile 1; mean difference: 1.50; 95% CI, −0.08 to 3.09; $P_{trend} < 0.01$).

We also examined the main effects of toenail Se and Hg on cognitive function tests, but no significant associations were found after adjustment for potential confounders (Supplementary Table 1). Although the interaction was not statistically significant ($P_{interaction} = 0.25$), the association between LCω-3 PUFA intake and DSST score was more profound in participants with higher toenail Se levels (tertile 3 versus tertile 1; Se ≥median 0.85 ppm: mean difference: 2.06; 95% CI, 0.30−3.82; $P_{trend} = 0.04$; Se <median 0.85 ppm: mean difference: 0.25; 95% CI, −1.59 to 2.09; $P_{trend} = 0.43$). However, Hg levels did not appreciably modify the association ($P_{interaction} = 0.54$). We further explored the joint modification of Se and Hg on LCω-3 PUFA intake, a three-way interaction. When participants were jointly classified according to median levels of Se (0.85 ppm) and Hg (0.22 ppm), the association between LCω-3 PUFA intake and DSST score was not materially modified ($P_{interaction} = 0.60$).

Discussion

Over 25 y of follow-up, we found that higher LCω-3 PUFA intake and non-fried seafood consumption were associated with better

Table 3
Associations (adjusted mean differences [95% CI]) between cumulative average daily intake of LCω-3 PUFA and DSST score by prespecified factors, the CARDIA study (N = 3231), 1985 to 2010*

	Levels of LCω-3 PUFA mean (SD)	Tertiles of LCω-3 PUFA intake			P_{trend}
		T1 (≤0.09 g/d)	T2 (0.09–0.18 g/d)	T3 (≥0.18 g/d)	
All participants	0.17 (0.17)	0 (Ref.)	0.37 (−0.78 to 1.52)	1.12 (−0.08 to 2.32)	0.048
Age at exam Y25 (y)					
<Median 51	0.16 (0.16)	0 (Ref.)	0.13 (−1.40 to 1.66)	1.32 (−0.30 to 2.94)	0.11
≥Median 51	0.18 (0.17)	0 (Ref.)	0.51 (−1.25 to 2.27)	0.19 (−1.61 to 2.00)	0.59
$P_{interaction}$	—	0.68			
Sex					
Female	0.15 (0.14)	0 (Ref.)	0.35 (−1.17 to 1.86)	0.66 (−0.97 to 2.29)	0.26
Male	0.19 (0.19)	0 (Ref.)	0.34 (−1.43 to 2.11)	1.57 (−0.22 to 3.36)	0.12
$P_{interaction}$	—	0.65			
Race					
Blacks	0.19 (0.19)	0 (Ref.)	1.24 (−0.53 to 3.00)	2.57 (0.77 to 4.37)	0.03
Whites	0.16 (0.14)	0 (Ref.)	−0.20 (−1.72 to 1.32)	−0.19 (−1.81 to 1.42)	0.66
P for interaction	—	0.50			
BMI (kg/m^2)					
<25	0.17 (0.18)	0 (Ref.)	1.38 (−0.41 to 3.16)	0.73 (−1.11 to 2.57)	0.97
≥25	0.17 (0.16)	0 (Ref.)	−0.27 (−1.78 to 1.24)	1.50 (−0.08 to 3.09)	<0.01
$P_{interaction}$	—	<0.01			
Smoking status					
Never	0.16 (0.15)	0 (Ref.)	−0.11 (−1.53 to 1.32)	0.68 (−0.83 to 2.19)	0.17
Former	0.19 (0.19)	0 (Ref.)	0.39 (−2.12 to 2.90)	1.45 (−1.10 to 4.01)	0.15
Current	0.17 (0.17)	0 (Ref.)	2.84 (−0.36 to 6.03)	2.32 (−1.00 to 5.65)	0.63
$P_{interaction}$	—	0.82			
Toenail selenium levels (ppm)					
<Median 0.85	0.18 (0.16)	0 (Ref.)	−0.39 (−2.18 to 1.41)	0.25 (−1.59 to 2.09)	0.43
≥Median 0.85	0.16 (0.17)	0 (Ref.)	1.18 (−0.47 to 2.83)	2.06 (0.30 to 3.82)	0.04
$P_{interaction}$	—	0.25			
Toenail mercury levels (ppm)					
<Median 0.22	0.14 (0.14)	0 (Ref.)	0.27 (−1.40 to 1.94)	1.45 (−0.46 to 3.35)	0.12
≥Median 0.22	0.20 (0.19)	0 (Ref.)	0.79 (−1.00 to 2.58)	0.88 (−0.90 to 2.67)	0.21
$P_{interaction}$	—	0.54			
Joint classification of selenium–mercury levels					
Low Se/High Hg	0.21 (0.18)	0 (Ref.)	−0.12 (−2.76 to 2.52)	−0.30 (−2.90 to 2.30)	0.76
High Se/High Hg	0.19 (0.20)	0 (Ref.)	1.46 (−1.03 to 3.95)	2.01 (−0.52 to 4.53)	0.16
Low Se/Low Hg	0.15 (0.15)	0 (Ref.)	−0.72 (−3.26 to 1.81)	0.75 (−2.04 to 3.54)	0.42
High Se/Low Hg	0.13 (0.13)	0 (Ref.)	1.13 (−1.15 to 3.41)	1.91 (−0.76 to 4.58)	0.19
$P_{interaction}$	—	0.60			

BMI, body mass index; CARDIA, Coronary Artery Risk Development in Young Adults; DSST, Digit Symbol Substitution Test; Hg, mercury; LCω-3 PUFA, long-chain ω-3 polyunsaturated fatty acid; Se, selenium

*All models were constructed using general linear model analysis with adjustment for covariates in model 2, Table 2, except the potential modifier. P_{trend} and $P_{interaction}$ were examined by using the continuous variable of LCω-3 PUFA intake with values >99th percentile excluded.

cognitive performance particularly measured by DSST, but not RAVLT nor the Stroop test. The observed associations were more pronounced in overweight or obese individuals. Although DSST score mainly reflects psychomotor speed indicating the overall efficiency of brain operations [21], RAVLT and the Stroop test measure verbal memory and executive functions, respectively. Because slow psychomotor speed appears far before the onset of mild cognitive impairment [22,23], the DSST score is considered a more sensitive index reflecting cognitive decline [21,24].

Although the beneficial effects of LCω-3 PUFA on cognitive function are supported by evidence from laboratory studies [25–27], epidemiologic studies yielded inconsistent findings. Two recent systematic reviews of RCTs did not find a significant improvement of cognitive function with respect to LCω-3 PUFA supplementation. However, most of the RCTs included in the reviews were conducted with <100 participants with an intervention period <5 mo [3,4]. Of note, some RCTs suggested a potential beneficial effect with a relatively long-term supplementation of LCω-3 PUFA [28,29]. Observational studies also suggested that long-term intakes of LCω-3 PUFA and seafood were associated with a slower age-related cognitive decline [5,6].

One important contribution of this study is that it recruited young adults and followed them for >25 y, whereas most of the previous studies focused on older adults. Middle-aged or older men and women are more likely to already have onset of diseases, so their lifestyle choice and health conditions may be affected by perceived ill health or treatment for existing disease. Follow-up of a cohort of young adults for 25 y provides insight on the natural history or early etiology of cognitive decline. Because long-term RCTs may not be feasible, a longitudinal study such as the present one certainly provides important data to the literature. In addition, we used cumulative average intake to reflect the usual diet as well as took cooking methods into account, which, to our knowledge, has not been considered in previous studies due to lack of data.

When examining LCω-3 PUFA and seafood, Se and Hg are two elements that warrant consideration because they often coexist with LCω-3 PUFA in seafood. Se is an element with antioxidant capacity and may protect against cognitive decline alone or by interacting with LCω-3 PUFA [30]. Se also can modulate the neurotoxicity effect of Hg [9,31,32]. The present findings are consistent with a recent report on US adults that failed to observe a significant interaction between LCω-3 PUFA and Hg with neurobehavioral

outcomes [33]. The association appeared to be more pronounced at higher toenail Se levels, although the interaction was not statistically significant.

In the present study, overweight or obese individuals seemed to have a better response to intake of LCω-3 PUFA in terms of cognitive decline. Because obesity is characterized by chronic inflammation [34], it is possible that the influence of LCω-3 PUFA on inflammation, and consequently cognitive function, is more evident in overweight and obese individuals who have a higher level of inflammation [35]. This hypothesis is supported by previous studies that found associations between LCω-3 PUFA and inflammatory biomarkers [36,37]. It is also possible that having more adipose tissues protects against fat-soluble pollutants, which are suggested to be associated with the risk for cognitive impairment [38].

Some limitations of the present study should be acknowledged. First, because cognitive function was assessed only once and not at baseline, we were unaable to evaluate the association with changes in cognitive function. The inclusion of cognitively impaired individuals at baseline is unlikely given the young age of the participants at study enrollment and the fact that they remained in the study for 25 y. Second, other nutrients in seafood may confound the association between LCω-3 PUFA and cognitive function. We adjusted for some nutrients in a sensitivity analysis and found the results were not significantly changed, but the concern still remains. The fact that the associations were attenuated after adjustment for the diet quality based on other food groups suggests that seafood intake may be a part of healthy diet [39]. Third, objective measurements of LCω-3 PUFA were not available. However, the diet history used in the present study has been validated [11,12]. The use of a food frequency questionnaire to reflect the levels of LCω-3 PUFA exposure is also supported in other studies [40,41]. Fourth, similar to other observational studies, the possibility of residual confounding from dietary and non-dietary factors cannot be completely ruled out. But the consistent results from the main and sensitivity analyses provide reassurance about the validity of our findings.

Conclusions

Findings from the present longitudinal study support that intakes of LCω-3 PUFA and non-fried seafood are associated with better cognitive performance in psychomotor speed. This study adds additional scientific evidence supporting the recommendation of seafood consumption for preventing or slowing down the process of cognitive decline.

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Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.nut.2018.11.002.

References

[1] Singh-Manoux A, Kivimaki M, Glymour MM, Elbaz A, Berr C, Ebmeier KP, et al. Timing of onset of cognitive decline: results from Whitehall II prospective cohort study. BMJ 2012;344:d7622.
[2] Weiser MJ, Butt CM, Mohajeri MH. Docosahexaenoic acid and cognition throughout the lifespan. Nutrients 2016;8:99.
[3] Rangel-Huerta OD, Gil A. Effect of omega-3 fatty acids on cognition: an updated systematic review of randomized clinical trials. Nutr Rev 2018;76:1–20.
[4] Teo L, Crawford C, Yehuda R, Jaghab D, Bingham JJ, Chittum HK, et al. Omega-3 polyunsaturated fatty acids to optimize cognitive function for military mission-readiness: a systematic review and recommendations for the field. Nutr Rev 2017;75(suppl_2):36–48.
[5] Zhang Y, Chen J, Qiu J, Li Y, Wang J, Jiao J. Intakes of fish and polyunsaturated fatty acids and mild-to-severe cognitive impairment risks: a dose-response meta-analysis of 21 cohort studies. Am J Clin Nutr 2016;103:330–40.
[6] Samieri C, Morris MC, Bennett DA, Berr C, Amouyel P, Dartigues JF, et al. Fish Intake, genetic predisposition to Alzheimer disease, and decline in global cognition and memory in 5 cohorts of older persons. Am J Epidemiol 2018;187:933–40.
[7] Aschner M, Aschner JL. Mercury neurotoxicity: mechanisms of blood–brain barrier transport. Neurosci Biobehav Rev 1990;14:169–76.
[8] Zhang ZH, Wen L, Wu QY, Chen C, Zheng R, Liu Q, et al. Long-term dietary supplementation with selenium-enriched yeast improves cognitive impairment, reverses synaptic deficits, and mitigates tau pathology in a triple transgenic mouse model of Alzheimer's disease. J Agric Food Chem 2017;65:4970–9.
[9] Sumino K, Yamamoto R, Kitamura S. A role of selenium against methylmercury toxicity. Nature 1977;268:73–4.
[10] Friedman GD, Cutter GR, Donahue RP, Hughes GH, Hulley SB, Jacobs Jr DR, et al. CARDIA: study design, recruitment, and some characteristics of the examined subjects. J Clin Epidemiol 1988;41:1105–16.
[11] McDonald A, Van Horn L, Slattery M, Hilner J, Bragg C, Caan B, et al. The CARDIA dietary history: development, implementation, and evaluation. J Am Diet Assoc 1991;91:1104–12.
[12] Liu K, Slattery M, Jacobs Jr D, Cutter G, McDonald A, Van Horn L, et al. A study of the reliability and comparative validity of the CARDIA dietary history. Ethn Dis 1994;4:15–27.
[13] Candela MAI, Bello J. Deep-fat frying modifies high-fat fish lipid fraction. J Agric Food Chem 1998;46:2783–6.
[14] Zhu N, Jacobs DR, Meyer KA, He K, Launer L, Reis JP, et al. Cognitive function in a middle aged cohort is related to higher quality dietary pattern 5 and 25 y earlier: the CARDIA study. J Nutr Health Aging 2015;19:33–8.
[15] Wechsler D. WAIS-III: administration and scoring manual: Wechsler adult intelligence scale. San Antonio, TX: Psychological Corporation; 1997.
[16] MacLeod CM. Half a century of research on the Stroop effect: an integrative review. Psychol Bull 1991;109:163–203.
[17] Stroop JR. Studies of interference in serial verbal reactions. J Exp Psychol 1935;18:643–62.
[18] Xun P, Liu K, Morris JS, Daviglus ML, Stevens J, Jacobs Jr DR, et al. Associations of toenail selenium levels with inflammatory biomarkers of fibrinogen, high-sensitivity c-reactive protein, and interleukin-6: the CARDIA trace element study. Am J Epidemiol 2010;171:793–800.
[19] Steven Morris J, Stampfer MJ, Willett W. Dietary selenium in humans toenails as an indicator. Biol Trace Elem Res 1983;5:529–37.
[20] Kramer H, Colangelo L, Lewis CE, Jacobs Jr. DR, Pletcher M, Bibbins-Domingo K, et al. Cumulative exposure to systolic blood pressure during young adulthood through midlife and the urine albumin-to-creatinine ratio at midlife. Am J Hypertens 2017;30:502–9.
[21] Salthouse TA. The processing-speed theory of adult age differences in cognition. Psychol Rev 1996;103:403–28.
[22] Backman L, Jones S, Berger AK, Laukka EJ, Small BJ. Cognitive impairment in preclinical Alzheimer's disease: a meta-analysis. Neuropsychology 2005;19:520–31.
[23] Saxton J, Lopez OL, Ratcliff G, Dulberg C, Fried LP, Carlson MC, et al. Preclinical Alzheimer disease: neuropsychological test performance 1.5 to 8 years prior to onset. Neurology 2004;63:2341–7.
[24] Proust-Lima C, Amieva H, Dartigues JF, Jacqmin-Gadda H. Sensitivity of four psychometric tests to measure cognitive changes in brain aging-population-based studies. Am J Epidemiol 2007;165:344–50.
[25] Dyall SC, Michael-Titus AT. Neurological benefits of omega-3 fatty acids. Neuromolecular Med 2008;10:219–35.
[26] Serhan CN, Chiang N, Van Dyke TE. Resolving inflammation: dual antiinflammatory and pro-resolution lipid mediators. Nat Rev Immunol 2008;8:349–61.
[27] Fraga VG, Carvalho MDG, Caramelli P, de Sousa LP, Gomes KB. Resolution of inflammation, n-3 fatty acid supplementation and Alzheimer disease: a narrative review. J Neuroimmunol 2017;310:111–9.
[28] Witte AV, Kerti L, Hermannstadter HM, Fiebach JB, Schreiber SJ, Schuchardt JP, et al. Long-chain omega-3 fatty acids improve brain function and structure in older adults. Cereb Cortex 2014;24:3059–68.
[29] Stonehouse W, Conlon CA, Podd J, Hill SR, Minihane AM, Haskell C, et al. DHA supplementation improved both memory and reaction time in healthy young adults: a randomized controlled trial. Am J Clin Nutr 2013;97:1134–43.
[30] Gao S, Jin Y, Hall KS, Liang C, Unverzagt FW, Ji R, et al. Selenium level and cognitive function in rural elderly Chinese. Am J Epidemiol 2007;165:955–65.
[31] Ganther HE, Goudie C, Sunde ML, Kopecky MJ, Wagner P. Selenium: relation to decreased toxicity of methylmercury added to diets containing tuna. Science 1972;175:1122–4.
[32] Seppanen K, Kantola M, Laatikainen R, Nyyssonen K, Valkonen VP, Kaarlopp V, et al. Effect of supplementation with organic selenium on mercury status as measured by mercury in pubic hair. J Trace Elem Med Biol 2000;14:84–7.
[33] Vacchi-Suzzi C, Karimi R, Kruse D, Silbernagel SM, Levine KE, Rohlman DS, et al. Low-level mercury, omega-3 index and neurobehavioral outcomes in an adult US coastal population. Eur J Nutr 2016;55:699–711.

[34] Ridker PM, Buring JE, Cook NR, Rifai N. C-reactive protein, the metabolic syndrome, and risk of incident cardiovascular events: an 8-year follow-up of 14 719 initially healthy American women. Circulation 2003;107:391–7.
[35] Donath MY, Dalmas E, Sauter NS, Boni-Schnetzler M. Inflammation in obesity and diabetes: islet dysfunction and therapeutic opportunity. Cell Metab 2013;17:860–72.
[36] Rodriguez-Cruz M, Cruz-Guzman ODR, Almeida-Becerril T, Solis-Serna AD, Atilano-Miguel S, Sanchez-Gonzalez JR, et al. Potential therapeutic impact of omega-3 long chain-polyunsaturated fatty acids on inflammation markers in Duchenne muscular dystrophy: a double-blind, controlled randomized trial. Clin Nutr 2018.
[37] Yan Y, Jiang W, Spinetti T, Tardivel A, Castillo R, Bourquin C, et al. Omega-3 fatty acids prevent inflammation and metabolic disorder through inhibition of NLRP3 inflammasome activation. Immunity 2013;38(6):1154–63.
[38] Lee DH, Lind PM, Jacobs Jr. DR, Salihovic S, van Bavel B, Lind L. Association between background exposure to organochlorine pesticides and the risk of cognitive impairment: A prospective study that accounts for weight change. Environ Int 2016;89–90:179–84.
[39] Pistollato F, Iglesias RC, Ruiz R, Aparicio S, Crespo J, Lopez LD, et al. Nutritional patterns associated with the maintenance of neurocognitive functions and the risk of dementia and Alzheimer's disease: A focus on human studies. Pharmacol Res 2018;131:32–43.
[40] Lucas M, Asselin G, Merette C, Poulin MJ, Dodin S. Validation of an FFQ for evaluation of EPA and DHA intake. Public health nutrition 2009;12(10):1783–90.
[41] Tjonneland A, Overvad K, Thorling E, Ewertz M. Adipose tissue fatty acids as biomarkers of dietary exposure in Danish men and women. The American journal of clinical nutrition 1993;57(5):629–33.