Cognitive function after supplementation with B vitamins and long-chain omega-3 fatty acids: ancillary findings from the SU.FOL.OM3 randomized trial¹⁻³

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ABSTRACT

Background: Rapid aging of the population worldwide necessitates a heightened concern about preventing cognitive decline.

Objective: We investigated the effects of B vitamins and omega-3 (n−3) fatty acid supplementation on cognition in a high-risk population.

Design: This was an ancillary study of the SU.FOL.OM3 (SUPplementation with FOLate, vitamins B-6 and B-12 and/or OMEga-3 fatty acids) secondary prevention trial conducted in France between 2003 and 2009. The present sample included 1748 men and women aged 45−80 y with a history of myocardial infarction, unstable angina, or ischemic stroke and who were recruited via a network of 417 physicians. With the use of block randomization with stratification by sex, age, prior cardiovascular disease, and city of residence, participants were assigned in a 2×2 factorial design to 1 of 4 groups: 1) 5-methyltetrahydrofolate (folate, 0.56 mg) and vitamins B-6 (3 mg) and B-12 (0.02 mg), 2) eicosapentaenoic and docosahexaenoic acids (600 mg) in a 2:1 ratio, 3) B vitamins and omega-3 fatty acids, or 4) placebo. Cognitive function after 4 y of supplementation was assessed with the French version of the modified Telephone Interview for Cognitive Status.

Results: No significant main effects of group assignment on cognitive function were found; however, we found some evidence of disease history− and age-specific effects. In the subgroup with prior stroke, for example, participants assigned to receive B vitamins plus omega-3 fatty acids were significantly less likely to have a decreased score on the temporal orientation task than those assigned to receive placebo (odds ratio: 0.43; 95% CI: 0.21, 0.86).

Conclusions: If present, dietary effects on cognition are likely group-specific. These results could be useful in interventions aimed at preventing cognitive decline in high-risk individuals. This trial is registered at controlled-trials.com as ISRCTN41926726. Am J Clin Nutr 2011;94:278–86.

INTRODUCTION

Dementia prevention constitutes a serious public health challenge because of rapid population aging and steep health care and societal costs (1, 2). Alzheimer disease (AD) currently affects 1 in 8 Americans >65 y of age (3, 4), and the 2010 US health care expenditures for AD management likely surpassed $172 billion (3). Apart from age, many prevalent yet modifiable cardiovascular disease (CVD) risk factors (hypertension, obesity, elevated homocysteine concentrations, and inflammation) have been associated with dementia risk (5, 6). In the absence of curative treatments, prevention efforts targeting high-risk individuals are critical (7).

Advancing age has also been associated with the risk of vitamin B-12 deficiency (8) and with decreasing blood folate concentrations (9). In turn, low folate concentrations (≤11.8 nmol/L) have been linked to nearly a 90% greater dementia risk than have normal concentrations (10). Folate and vitamin B-12 either directly, through nucleic acid synthesis and methylation reactions, or indirectly, through a deficiency resulting in impaired DNA repair in neurons or inadequate methylation, could cause cognitive impairment (11−13). Despite promising findings from observational studies (14), a meta-analysis of randomized controlled trials (RCTs) with B vitamins did not provide support regarding dementia prevention (15). However, a recent 3-y RCT documented improvement in processing speed and memory in elderly men with somewhat elevated homocysteine concentrations who consumed 800 µg folic acid/d (16). Methodologic aspects that hinder RCT comparisons include heterogeneity of the sample studied, supplement dosage, treatment duration, and outcome measures (2, 15, 17−19).

In turn, long-chain polyunsaturated fatty acids of the omega-3 (n−3) series have established protective vascular effects, such as cerebral blood flow improvement and antithrombotic activity, and can reduce amyloid-β pathology in animals (20). Epidemiologic evidence suggests that dietary intake of omega-3 fatty acids might protect against AD (20). The few available RCTs, however, have largely yielded nonsignificant results (20), except in individuals with mild cognitive impairment (21) or age-related cognitive decline (22). The objectives of this ancillary study of the SU.FOL.

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OM3 (SUpplementation with FOLate, vitamins B-6 and B-12 and/or OMEga-3 fatty acids) RCT were to assess the effects of supplementation on cognition in individuals with a history of CVD. Consistent with evidence of poststroke dementia risk (23), we also expected increased supplementation effects in a subsample with a history of stroke.

SUBJECTS AND METHODS

Study design and participants

The SU.FOL.OM3 was a multicenter, randomized, double-blind, placebo-controlled, secondary prevention trial conducted between 2003 and 2009 (24, 25). Men and women aged 45–80 y with a recent myocardial infarction (MI), unstable angina, or ischemic stroke were eligible for participation. Primary outcomes included recurrent MI, stroke, and CVD mortality after 5 y of follow-up. Details about the trial’s design (Figure 1), implementation, and main findings are available elsewhere (24–26). Briefly, the participants were recruited via a network of 417 cardiologists, neurologists, and other physicians throughout France. Shortly after the start of enrollment, the trial’s scientific committee decided on a slight modification of the definition for acute coronary syndrome without MI to refine the eligibility criteria (24). No changes to the outcomes of interest were made after launching the trial. The sample size calculation followed an expert
literature review and was based on an estimated CVD risk of 0.087 in the placebo group (24). All participants provided written informed consent. The study protocol was approved by the Consultation Committee for the Protection of Participants in Biomedical Research of the Paris-Cochin Hospital and by the French National Information and Citizen Freedom Committee.

Randomization and intervention

We used computerized block randomization (block size = 8) with stratification by sex, age (45–54, 55–64, and 65–80 y), prior CVD, and city of residence. The statistics team at the trial’s coordinating center randomized participants in a 2-by-2 factorial design to 1 of 4 daily treatment groups: 1) B vitamins: 5-methyltetrahydrofolate (5-methyl-THF; 0.56 mg), vitamin B-6 (3 mg), and vitamin B-12 (0.02 mg); 2) long-chain omega-3 fatty acids: 600 mg eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) in a ratio of 2:1; 3) B vitamins and omega-3 fatty acids; and 4) placebo. The supplements were given as 2 capsules to be taken once daily and were provided free of charge by Merck Eprova AG (5-methyl-THF; Schaffhausen, Switzerland), Roche Laboratories (vitamins B-6 and B-12; Basel, Switzerland), and Pierre Fabre Laboratories (omega-3 fatty acids; Ramonville, France). The supplements and the placebo capsules were made of gelatin manufactured by Catalat Pharma Solutions (Beinheim, France; Swindon, United Kingdom). The participants were given sufficient supplements for 1 y and were reexamined at annual follow-up visits at 1 of the 257 participating centers.

Outcome assessment

The main outcome in the current study was cognitive function, assessed with the recently validated French version of the modified Telephone Interview for Cognitive Status (F-TICS-m) (27–30). The instrument has a maximum score of 43, takes ~10 min to complete, and includes items about temporal/spatial orientation, semantic memory, immediate and delayed recall, attention/calculation from the Mini-Mental State Examination (31), and language (repetition of 2 complex sentences and comprehension of a simple command). As part of the SU.FOL.OM3 assessment battery, the F-TICS-m was administered during 2007–2009 only to participants who had completed 4 y of follow-up. Because a validated version of the F-TICS-m had not been available at the start of the SU.FOL.OM3 trial, cognitive performance at baseline was evaluated with the Isaacs Set Test (IST) (32). This test is a 1-min verbal fluency task that has been validated in elderly French individuals (33) and in a sample of individuals who had recently had a stroke (34). The IST is considered an appropriate dementia screening tool, has easy instructions, and has objective scoring (33); however, it is not as comprehensive as the F-TICS-m regarding cognitive function assessment.

Covariates

Follow-up assessment covered a wide range of psychosocial, behavioral, and medical domains along with treatment compliance (defined as taking ≥80% of the supplements) and possible adverse effects (24). We used baseline data on serum concentrations of folate and vitamin B-12 and plasma concentrations of vitamin B-6, homocysteine, cholesterol, triglycerides, creatinine, and fasting glucose. All biomarkers were measured, treated, and stored according to strict protocol guidelines, as described elsewhere (24).

Statistical analysis

Baseline characteristics and group comparability were evaluated with likelihood-ratio chi-square tests, ANOVA, and Kruskal-Wallis tests. The demographic (age), clinical (body mass index and blood pressure), and cognitive (total scores on the IST and the F-TICS-m) continuous-scale variables were standardized (mean = 0, SD = 1) before modeling. Marital status, education, birthplace, and the responses to individual F-TICS-m components were recoded into 2- or 3-category variables (married/living with partner compared with other; less than high school diploma, high school diploma, or postsecondary education; birth in France or born abroad; a score of 0 to 4 compared with a score of 5 on temporal orientation; a score of 0 to 2 compared with a score of 3 on spatial orientation and language/repetition; and a score of 0 to 5 compared with a score of 6 on attention/calculation and semantic memory). In a preliminary step, we carried out unadjusted and adjusted analyses (with available data for slightly more than one-half of the full SU.FOL.OM3 sample), modeling the concentrations of homocysteine and omega-3 fatty acids as the respective independent variables and the total IST score as a dependent variable. This improved our understanding of the sample at baseline. Next, because of the study’s factorial design, we investigated the presence of interaction between the B vitamins and omega-3 fatty acids and found some evidence of effect modification regarding subscores on spatial orientation (P = 0.08). Because this was an important construct for this study, we proceeded with 2 sets of main effects analyses (significance level = 0.05; 2-sided). In the first set, we investigated the effect of assignment to B vitamins alone or in combination compared with all other groups and the effect of assignment to omega-3 fatty acids alone or in combination compared with all other groups. In the second set, we investigated the effect of group assignment considering all 4 groups individually. Effects on the total F-TICS-m score, the memory subscore, and the recall task subscore were evaluated with one-factor analysis of covariance; effects of group assignment on temporal and spatial orientation, attention/calculation, semantic memory, and repetition were assessed with multiple logistic regression. We performed post hoc analyses using Tukey’s Studentized range test. We carried out full-sample and subgroup analyses to assess variation by CVD history, homocysteine concentration, and age whenever significant interactions (P < 0.10) were found. The results are presented as mean (±SD) scores or as the percentage of participants scoring above the cutoff for categorical variables. All analyses were conducted with SAS version 9.1 (SAS Institute Inc, Cary, NC) according to the intent-to-treat principle.

RESULTS

Sample characteristics

The SU.FOL.OM3 response rates at 6, 12, and 24 mo and at the end of the trial were 99%, 96%, 94%, and 95%, respectively. About 86% of those who returned a questionnaire (regardless of treatment assignment) reported compliance with the supplementation regimen. In the full SU.FOL.OM3 sample, 2.1% of the
participants discontinued supplementation because of side effects (gastrointestinal disturbances, nausea, and cutaneous reactions). Baseline demographic, cognitive, clinical, and biological characteristics of the participants in the current study are summarized in Table 1. Of the 2501 individuals randomly assigned in the SU.FOL.OM3 trial, 1881 (75.2%) had completed 4 y of follow-up, of whom 1748 (69.9%) had complete cognitive assessment data and were thus eligible for the current study. Overall, 72 participants (2.9%) were not able to complete the F-TICS-m because of interfering comorbidities, language problems, or recent death, and 61 participants (2.4%) refused participation in the cognitive function assessment. The 4 treatment groups were well balanced with respect to all baseline characteristics except age, which showed marginal variability (P = 0.08; Table 1). Hence, the principal models with all 4 treatment groups were adjusted for age at baseline. Cognitive performance at baseline, assessed by the IST, was very similar across groups. Regarding assignment to B vitamins (alone or in combination), we observed statistically significant variability by age (P = 0.03) and marginal variability by EPA and vitamin B-12 concentrations (P = 0.09 and P = 0.13, respectively; Table 1), and we adjusted the principal models accordingly. Finally, all baseline characteristics of those randomly assigned to receive long-chain omega-3 fatty acids (alone or in combination) compared with the others were well balanced, and no statistical adjustment was made.

No statistically significant differences in the baseline concentrations of vitamin B-12, homocysteine, cholesterol, triglycerides, or creatinine were found between the SU.FOL.OM3 participants who were included (n = 1748) and those who were not included (n = 753) in the current study. The 2 groups were not significantly different regarding blood pressure levels, current smoking status, proportion of women, or proportion of individuals with a history of unstable angina. However, baseline concentrations of vitamin B-6, folate, EPA, and DHA were slightly elevated in the included individuals (all P < 0.05). In addition, participants included in this study had slightly higher mean IST scores (35.8 compared with 34.4, respectively; P < 0.0001), had a slightly lower mean body mass index (24.7 compared with 27.9, respectively; P < 0.05), and were somewhat younger (mean ages: 61.0 and 62.1 y, respectively; P < 0.01). Finally, the proportion of individuals with a history of ischemic stroke was slightly lower, whereas the proportion of those with MI was slightly higher in the included participants than in those not eligible for the current study (24% compared with 30% for stroke, respectively; 48% compared with 42% for MI, respectively; P < 0.01).

With the use of available baseline data, we observed strong, positive associations between plasma concentrations of the omega-3 fatty acids and cognition, assessed with the IST, in both unadjusted (P < 0.0001; n = 1341) and adjusted (P < 0.01; n = 1324; adjustment for age, education, stroke history, and group assignment) models. The unstandardized regression coefficients were somewhat larger for DHA than for EPA [1.43 (SE = 0.58) compared with 0.94 (SE =0.34), respectively, in adjusted models]. The association with homocysteine was negative and marginally significant (P < 0.08; n = 1676) in unadjusted analyses and statistically nonsignificant in the adjusted model (P > 0.79; n = 1652). Additional preliminary analyses testing for interactions between the B vitamins and the omega-3 fatty acids, with each sample characteristic (listed in Table 1) modeled as a dependent variable, showed no statistically significant interactions (data not shown). Two models showed marginal significance for serum vitamin B-12 status (P = 0.07) and plasma total cholesterol (P = 0.09). However, because of the multiple comparisons performed, we interpreted these P values as rather weak evidence of effect modification.

Assessment of cognitive function

The mean (±SD) total score on the F-TICS-m was 28.5 ± 4.8, and the scores were normally distributed (range: 7–43). In our sample, 314 participants (18%) scored at or above the cutoff of 33, which has been shown to have 86% sensitivity (30). As expected, the F-TICS-m total score was inversely correlated with age (r = −0.30, P < 0.0001). Mean values on the F-TICS-m were significantly higher in participants with a history of MI or unstable angina than in those with a history of stroke (28.9 ± 4.6 compared with 27.1 ± 5.4; P < 0.0001). The F-TICS-m total score was positively correlated with baseline IST scores (r = 0.46, P < 0.0001). The latter was also normally distributed and had a mean (±SD) value of 35.8 ± 4.5. Individual IST subscores (for naming animals, colors, fruit, and cities) did not differ significantly by treatment group (all P > 0.5).

Effects of daily dietary supplementation on cognitive function

The results of the main analyses by B vitamin assignment (with statistical adjustment for baseline EPA, vitamin B-12 status, and age), by omega-3 fatty acid assignment, and in all 4 groups (with statistical adjustment for age at baseline) are summarized in Table 2. Overall, there were no statistically significant main effects of group assignment on cognitive function (all P > 0.1). Neither the F-TICS-m total score nor the subscores on the individual F-TICS-m components showed any variability by treatment group in the full sample.

Regarding subgroup effects, there was no effect modification by homocysteine (P > 0.3); however, we observed a statistically significant effect modification by CVD history. Such effects were evident regarding the F-TICS-m total score and the memory subscore (both P < 0.05) when all 4 groups were evaluated individually and regarding the F-TICS-m total score (P < 0.09), temporal orientation (P < 0.04), and semantic memory (P < 0.03) when the role of B vitamins was evaluated (data not tabulated). These findings were explored further, and the respective results are presented in Table 3. In the subgroup with a history of ischemic stroke, participants assigned to supplementation with B vitamins plus omega-3 fatty acids were significantly less likely to score lower on temporal orientation than were those assigned to placebo [15% compared with 29% of individuals scoring <5 points, respectively; odds ratio (OR): 0.43; 95% CI: 0.21, 0.86]. When comparing those assigned to receive B vitamins (alone or in combination) with the other groups, we found borderline significant evidence that the former group was ~70% more likely to score higher on temporal orientation than was the latter group (83% compared with 72% of individuals scoring ≥4 points, respectively; OR: 1.69; 95% CI: 0.98, 2.93).

Conversely, in the subsample with a history of MI or unstable angina, subscores on the semantic memory task appeared to vary by group. Participants assigned to B vitamin supplementation...
<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Full sample(^\text{a}) (n = 1748)</th>
<th>B vitamins and omega-3 (n = 438)</th>
<th>Omega-3 (n = 439)</th>
<th>B vitamins (n = 446)</th>
<th>Placebo (n = 425)</th>
<th>(P^4)</th>
<th>(P^4)</th>
<th>(P^4)</th>
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<tr>
<td>Age (y)(^6)</td>
<td>61.0 ± 8.8</td>
<td>61.6 ± 8.8</td>
<td>60.1 ± 8.7</td>
<td>61.4 ± 8.7</td>
<td>60.9 ± 8.9</td>
<td>0.08</td>
<td>61.5 ± 8.8</td>
<td>60.5 ± 8.8</td>
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<td>Female sex [(n (%))]</td>
<td>357 (20.4)</td>
<td>80 (18.3)</td>
<td>88 (20.0)</td>
<td>93 (20.9)</td>
<td>96 (22.6)</td>
<td>NS</td>
<td>173 (19.6)</td>
<td>184 (21.3)</td>
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<td>Married [(n (%))]</td>
<td>1290 (73.8)</td>
<td>316 (72.1)</td>
<td>320 (72.9)</td>
<td>333 (74.7)</td>
<td>321 (75.5)</td>
<td>NS</td>
<td>649 (73.4)</td>
<td>641 (74.2)</td>
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<td>Less than high school diploma [(n (%))]</td>
<td>1013 (58.0)</td>
<td>256 (58.4)</td>
<td>254 (57.9)</td>
<td>253 (56.7)</td>
<td>250 (58.8)</td>
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<td>509 (57.6)</td>
<td>504 (58.3)</td>
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<td>Employed [(n (%))]</td>
<td>174 (10.0)</td>
<td>42 (9.7)</td>
<td>45 (10.3)</td>
<td>45 (10.1)</td>
<td>42 (9.9)</td>
<td>NS</td>
<td>87 (9.8)</td>
<td>87 (10.1)</td>
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<td>Current smoker</td>
<td>69 (3.5)</td>
<td>6.8 (3.4)</td>
<td>6.7 (3.4)</td>
<td>6.9 (3.5)</td>
<td>7.0 (3.7)</td>
<td>NS</td>
<td>6.8 (3.5)</td>
<td>6.9 (3.6)</td>
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<td><strong>Clinical</strong></td>
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<td>BMI (kg/m(^2))</td>
<td>27.4 ± 3.9</td>
<td>27.6 ± 4.3</td>
<td>27.4 ± 3.8</td>
<td>27.4 ± 3.8</td>
<td>27.4 ± 3.8</td>
<td>NS</td>
<td>27.5 ± 4.0</td>
<td>27.4 ± 3.8</td>
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<tr>
<td>Systolic BP (mm Hg)</td>
<td>132.8 ± 20.9</td>
<td>132.5 ± 20.6</td>
<td>133.5 ± 21.7</td>
<td>132.7 ± 20.9</td>
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<td>NS</td>
<td>132.6 ± 20.7</td>
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<td>Diastolic BP (mm Hg)</td>
<td>83.3 ± 11.9</td>
<td>83.3 ± 11.8</td>
<td>83.9 ± 11.8</td>
<td>83.2 ± 12.0</td>
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<td>NS</td>
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<td>Serum folate (ng/mL)</td>
<td>6.9 (3.5)</td>
<td>3.4 (3.4)</td>
<td>6.7 (3.4)</td>
<td>6.9 (3.5)</td>
<td>7.0 (3.7)</td>
<td>NS</td>
<td>6.8 (3.5)</td>
<td>6.9 (3.6)</td>
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<td>Plasma vitamin B-6 (nmol/L)</td>
<td>391 (24.8)</td>
<td>38.4 (23.6)</td>
<td>40.1 (25.7)</td>
<td>39.1 (23.8)</td>
<td>38.7 (27.1)</td>
<td>NS</td>
<td>38.8 (23.8)</td>
<td>39.6 (26.0)</td>
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<tr>
<td>Plasma vitamin B-12 (pg/mL)</td>
<td>3640 (164.0)</td>
<td>3595 (151.5)</td>
<td>3630 (177.0)</td>
<td>3590 (157.0)</td>
<td>3765 (171.0)</td>
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<td>3590 (155.0)</td>
<td>3710 (174.0)</td>
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<td>Plasma EPA (%)</td>
<td>2.6 (1.2)</td>
<td>3.8 (2.0)</td>
<td>3.8 (2.1)</td>
<td>3.8 (2.0)</td>
<td>3.8 (2.1)</td>
<td>NS</td>
<td>3.8 (2.0)</td>
<td>3.9 (2.1)</td>
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<tr>
<td>Plasma DHA (%)</td>
<td>2.6 (1.2)</td>
<td>3.8 (2.0)</td>
<td>3.8 (2.1)</td>
<td>3.8 (2.0)</td>
<td>3.8 (2.1)</td>
<td>NS</td>
<td>3.8 (2.0)</td>
<td>3.9 (2.1)</td>
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<td>Plasma EPA+DHA (%)</td>
<td>129 (50.0)</td>
<td>13.1 (5.0)</td>
<td>12.9 (4.8)</td>
<td>12.8 (4.6)</td>
<td>12.7 (5.1)</td>
<td>NS</td>
<td>12.9 (4.9)</td>
<td>12.8 (5.0)</td>
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<td>Plasma fasting glucose (mmol/L)</td>
<td>5.4 (1.0)</td>
<td>5.5 (1.1)</td>
<td>5.4 (1.0)</td>
<td>5.5 (0.9)</td>
<td>5.4 (1.0)</td>
<td>NS</td>
<td>5.5 (1.0)</td>
<td>5.4 (1.0)</td>
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<td>Plasma cholesterol (mmol/L)</td>
<td>4.6 (1.4)</td>
<td>4.5 (1.3)</td>
<td>4.6 (1.5)</td>
<td>4.6 (1.4)</td>
<td>4.5 (1.1)</td>
<td>NS</td>
<td>4.5 (1.4)</td>
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<tr>
<td>Plasma HDL (mmol/L)</td>
<td>1.1 (0.3)</td>
<td>1.1 (0.5)</td>
<td>1.2 (0.3)</td>
<td>1.2 (0.4)</td>
<td>1.1 (0.3)</td>
<td>NS</td>
<td>1.1 (0.4)</td>
<td>1.1 (0.3)</td>
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<tr>
<td>Plasma LDL (mmol/L)</td>
<td>2.7 (1.1)</td>
<td>2.7 (1.0)</td>
<td>2.7 (1.2)</td>
<td>2.7 (1.0)</td>
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<td>NS</td>
<td>2.7 (1.0)</td>
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<tr>
<td>Plasma creatinine ((\mu)mol/L)</td>
<td>780 (20.0)</td>
<td>78.0 (18.0)</td>
<td>76.0 (22.0)</td>
<td>78.0 (19.0)</td>
<td>78.0 (19.0)</td>
<td>NS</td>
<td>78.9 (19.0)</td>
<td>77.0 (20.0)</td>
</tr>
<tr>
<td>Plasma triglycerides (mmol/L)</td>
<td>1.2 (0.8)</td>
<td>1.2 (0.7)</td>
<td>1.2 (0.8)</td>
<td>1.3 (0.8)</td>
<td>1.2 (0.8)</td>
<td>NS</td>
<td>1.2 (0.8)</td>
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<td><strong>Cognitive</strong></td>
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<tr>
<td>Isaacs Set Test</td>
<td>35.8 ± 7.5</td>
<td>35.7 ± 7.8</td>
<td>35.9 ± 7.0</td>
<td>35.7 ± 7.9</td>
<td>36.1 ± 7.1</td>
<td>NS</td>
<td>35.7 ± 7.9</td>
<td>36.0 ± 7.1</td>
</tr>
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</table>

\(^1\) BP, blood pressure; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid.

\(^2\) Tests for interaction between the B vitamins and the omega-3 fatty acids with each sample characteristic modeled as a dependent variable resulted in \(P\) values ranging from 0.07 to 0.99.

\(^3\) Sample with available cognitive function data.

\(^4\) Based on chi-square tests for categorical variables, \(t\) tests or ANOVA for continuous variables with normal response distributions, and Wilcoxon’s or Kruskal-Wallis tests for continuous variables with nonnormal response distributions.

\(^5\) Values are means ± SDs.

\(^6\) Values are medians; interquartile ranges in parentheses.
Recall 3.3

F-TICS-m total score 28.5

6

Outcome measure

1.8 compared with 3.0

27.7

\( n \) of age.

5 points, respectively; OR: 1.70; 95% CI: 1.16, 2.51). The

examining the 4 treatment groups individually, we found that those

assigned to receive placebo were 70% more likely to score higher

on the semantic memory task than were those in the combined
treatment group (83% compared with 74% of individuals scoring

>5 points, respectively; OR: 1.70; 95% CI: 1.16, 2.51). The effect of
group assignment on the F-TICS-m total score and on the recall
score appeared to vary by age (all \( P < 0.10 \) (data not tabulated). Post hoc analyses using Tukey’s Studentized
range test indicated that, in the age group 65–80 \( y \), compared

with participants assigned to receive placebo, participants as-

signed to B vitamin supplementation had significantly lower

scores (mean 5.1 compared with 6.9; \( P < 0.05 \)) and recall
subscores (mean 4.9; \( P < 0.04 \)). No variation in cogni-
tion scores, by group assignment, was observed in participants

<65 \( y \) of age.

DISCUSSION
If present, the 4-y effects of dietary supplementation with

nutritional doses of folate, vitamin B-6, and vitamin B-12—

administered alone or in combination with long-chain omega-3

fatty acids—might be CVD history– and age-specific. No sig-
nificant differences in cognitive function between the treat-

ment groups were identified in the full sample.

Substantial research evidence has linked past stroke with

a 2-fold increased risk of dementia in the population aged >65 \( y \)

(35). The prevalence of poststroke dementia is \( \approx 30\% \), and the propor-
tion of individuals with presumed AD among those with

poststroke dementia is 19–61% in developed countries (23). We

observed that, in the subgroup with a history of ischemic stroke,
participants assigned to received B vitamins plus omega-3 fatty

acids were >50% more likely to score better on the temporal

orientation task than were those assigned to receive placebo. In

fact, temporal orientation showed a relatively low correlation

(Spearman \( r = 0.31 \)) with the total F-TICS-m score, which is

more strongly influenced by memory. No such effects on tem-

poral orientation were found in participants with a history of MI

or unstable angina. However, the latter group exhibited effects

on semantic memory that were contrary to our expectations.

Specifically, participants assigned to B vitamin supplementation

(alone or in combination) tended to score lower than their

counterparts who received placebo. Whereas these results cor-
raborate evidence for the synergistic effects of different nutrient

groups (36), it is important that future studies replicate these

models before they could be attributed to chance or before firm

conclusions could be drawn. Also contrary to our expectations,

homocysteine did not appear to moderate the effect of group

assignment on cognition. Plasma homocysteine concentra-
tions in this study were largely within the normal range; only 3% of

the sample had evidence of mild-to-moderate hyperhomocysteinemia

(homocysteine > 25 \( \mu \text{mol/L} \)). Positive effects of folic acid sup-

plementation on several aspects of cognition (memory and pro-

cessing speed) have been observed in a sample of elderly men with

elevated homocysteine concentrations (16).

Unlike the SU.FOL.OM3 trial, RCTs examining cognitive

function typically do not combine B vitamins and omega-3 fatty

acids. To an extent, this prevents drawing parallels between the

results of the current study and other available findings. Several

secondary prevention RCTs with B vitamins have shown non-
significant effects on cognition (37–39). However, a recent RCT

with some similarities to the SU.FOL.OM3 trial regarding sample

composition, duration, and outcome assessment, yet much higher

supplementation doses (2.5 mg folic acid, 50 mg vitamin B-6, and

1 mg vitamin B-12), documented positive supplementation effects

among women with low dietary B vitamins intake (39).

Available omega-3 fatty acids RCT findings were largely

derived from studies with short treatment durations (3–12 mo)

with relatively small samples of AD patients (21, 40, 41) or with

samples of elderly individuals (42, 43). For example, a recent

RCT (\( n = 295 \) AD patients completing the trial) did not show

any significant effects of 2 g algal DHA/d on cognitive decline

after 18 mo of treatment (40). However, another RCT with AD

patients showed positive effects of 1.7 g DHA/d and 0.6 g EPA/d

on delayed word recall and attention only in individuals with

very mild cognitive dysfunction (21). Daily supplementation with

DHA (900 mg) has been shown to improve episodic memory and

learning in a sample of individuals with age-related cognitive

decline, after 6 mo of treatment (22). In turn, a recent 2-y RCT

involving daily supplementation with 200 mg EPA and 500 mg

\( \text{F-TICS-m, French Telephone Interview for Cognitive Status,}

modified. No significant main effects were observed in any of the

models.

\( ^{2} \text{Mean \pm SD (all such values).} \)

\( ^{1} \) Percentages represent the proportion of participants scoring above the established cutoff. F-TICS-m, French Telephone Interview for Cognitive Status.

\( \text{TABLE 2} \)

Cognitive performance by treatment group after 4 y of follow-up

<table>
<thead>
<tr>
<th>Outcome measure</th>
<th>Treatment group</th>
<th>B vitamins</th>
<th>Omega-3 fatty acids</th>
</tr>
</thead>
<tbody>
<tr>
<td>F-TICS-m total score</td>
<td>28.5 ± 4.7</td>
<td>28.4 ± 4.8</td>
<td>28.3 ± 5.0</td>
</tr>
<tr>
<td>Temporal orientation (%)</td>
<td>90</td>
<td>90</td>
<td>90</td>
</tr>
<tr>
<td>Spatial orientation (%)</td>
<td>94</td>
<td>94</td>
<td>94</td>
</tr>
<tr>
<td>Memory</td>
<td>4.8 ± 2.5</td>
<td>4.9 ± 1.5</td>
<td>4.9 ± 1.6</td>
</tr>
<tr>
<td>Attention/calculation (%)</td>
<td>48</td>
<td>48</td>
<td>48</td>
</tr>
<tr>
<td>Semantic memory (%)</td>
<td>71</td>
<td>71</td>
<td>71</td>
</tr>
<tr>
<td>Recall (%)</td>
<td>3.3 ± 1.8</td>
<td>3.2 ± 1.7</td>
<td>3.2 ± 1.9</td>
</tr>
<tr>
<td>Repetition (%)</td>
<td>40</td>
<td>39</td>
<td>39</td>
</tr>
</tbody>
</table>

(continued on next page)
TABLE 3  
Cognitive performance by cardiovascular disease history: effects of group assignment

<table>
<thead>
<tr>
<th>Treatment groups</th>
<th>Myocardial infarction or unstable angina</th>
<th>Ischemic stroke</th>
</tr>
</thead>
<tbody>
<tr>
<td>B vitamins and omega-3</td>
<td>B vitamins</td>
<td>Placebo</td>
</tr>
<tr>
<td>Outcome measure</td>
<td>Yes</td>
<td>No</td>
</tr>
<tr>
<td>F-TICS-m total score</td>
<td>28.8 ± 4.1</td>
<td>28.9 ± 4.5</td>
</tr>
<tr>
<td>Semantic memory (%)</td>
<td>54 ± 15</td>
<td>53 ± 5 %</td>
</tr>
<tr>
<td>Visual recognition (%)</td>
<td>73 ± 19</td>
<td>73 ± 18</td>
</tr>
<tr>
<td>Attention/calculator (%)</td>
<td>81 ± 18</td>
<td>81 ± 18</td>
</tr>
<tr>
<td>Temporal orientation (%)</td>
<td>83 ± 18</td>
<td>83 ± 18</td>
</tr>
<tr>
<td>Spatial orientation (%)</td>
<td>92 ± 19</td>
<td>92 ± 19</td>
</tr>
<tr>
<td>Recall (%)</td>
<td>42 ± 15</td>
<td>42 ± 15</td>
</tr>
<tr>
<td>Repetition (%)</td>
<td>39 ± 14</td>
<td>39 ± 14</td>
</tr>
</tbody>
</table>

Percentages represent the proportion of participants scoring above the established cutoff. F-TICS-m, French Telephone Interview for Cognitive Status, modified.

Models adjusted for baseline eicosapentaenoic acid, vitamin B-12, and age.  
The F-TICS-m total score was set as the outcome in the logistic regression.  
DHA compared with olive oil did not show any significant effects in cognitively healthy elderly participants (42). Generally, methodologic aspects that prevent drawing firm conclusions include considerable heterogeneity in RCT sample selection, supplement dosage, assay techniques, and outcome measures and insufficient statistical power and/or treatment duration (2, 15, 17, 19).

Interestingly, baseline folate concentrations (assessed in plasma) in the VISP (Vitamin Intervention for Stroke Prevention) trial, involving individuals with a history of stroke (37), were about twice as high as the folate concentrations (assessed in serum) observed in our study. However, B vitamin and omega-3 fatty acid concentrations in our sample were largely comparable with those reported in other RCTs of cognitive function (16), and we believe that the baseline biomarker status of our participants did not prevent us from observing an effect of the supplementation. The low supplementation doses used in this study reflect our interest in investigating the effects of nutritional and not pharmacologic doses designed to treat cognitive function or vitamin deficiencies. This aspect was regarded as a distinctive and novel feature of the SU.FOL.OM3 trial (25). The use of nutritional doses has several advantages, including its safety and tolerance, and facilitates interpretation of the results with respect to attaining an optimal diet. In addition, a comprehensive literature review recently pointed out that, despite a wide variation in the omega-3 doses used in RCTs, there were no dose-response effects on cognition (20). That review also suggested that high DHA doses might in fact promote existing degenerative processes in the brain via lipid peroxidation, whereas low doses of DHA might be protective against oxidative stress. Nonetheless, we are aware that the optimal dose of supplementation, the optimal ratio of EPA to DHA, and the optimal duration of treatment are yet to be established (20).

In ≈54% of the 2501 individuals randomly assigned in the SU.FOL.OM3 trial, we were able to evaluate the baseline association between plasma concentrations of the omega-3 fatty acids and cognition, assessed with the IST. Whereas we noted statistically significant positive associations between cognition and both EPA and DHA, the cross-sectional nature of these models prevented the establishment of causality. Nonetheless, the presence of these relations served as further justification for our interest in the role of omega-3 fatty acid supplementation on cognitive function. The treatment duration, however, might have been insufficient to observe significant differences on the F-TICS-m between the groups. Other limitations of the current study pertain to the inability to assess baseline cognitive function with the F-TICS-m, hence the lack of data for estimating the rate of cognitive loss in our sample and the performance of multiple comparisons while retaining the significance levels set a priori. We also acknowledge that the use of low supplementation doses might be one of the reasons for the inability to show a statistically significant association with cognition in the full sample.

Important strengths of this study included the large sample size, long treatment duration, use of 5-methyl-THF (the most abundant natural folate form), and assessment of the individual and combined effects of B vitamins and long-chain omega-3 fatty acids. Treatment compliance in the trial was high, as evidenced by self-reports and by increased blood concentrations of B vitamins and omega-3 fatty acids (24). Cognitive function was assessed by telephone after 4 y of follow-up with use of the recently validated F-TICS-m, which had shown good internal consistency (Cronbach’s
z = 0.69) and a strong correlation with the Mini-Mental State Examination (29, 30). The TICS and TICS-m are considered practical and valid dementia screening tools without floor effects, which can be used in individuals with physical disabilities or visual impairment but without considerable verbal communication or hearing problems (30, 34, 44). Meanwhile, we acknowledge that the various items of the F-TICS-m tap on different cognitive domains, yet serve as simple cognition assessment tools that are certainly inferior to comprehensive neurocognitive assessment batteries.

Overall, extraction of specific dietary effects on cognitive function is challenging because of the synergy between different dietary components and because blood concentrations of vitamins can be influenced by environmental, metabolic, and genetic factors (36). Nonetheless, hyperhomocysteinemia, hypertension, and diabetes—all of which predispose to dementia—can be modified by diet (45). Even small delays in the onset or progression of dementia have been estimated to produce a sizable effect on its global burden (7, 46). In the absence of curative treatments and considering the aging worldwide population and the associated rise in the incidence and prevalence of dementia, prevention remains a critical public health objective.

The authors’ responsibilities were as follows—PG and SH: designed and conducted the research; VAA: analyzed the data and led the writing; EK-G, PB-G, and LF: assisted with the literature review and data analysis; and VAA: had primary responsibility for the final content. All authors read and edited each draft and approved the final manuscript. None of the funding organizations had any involvement in the design and conduct of the study; in the collection, management, analysis, or interpretation of the data; or in the preparation, review, and approval of the manuscript. None of the authors had any conflict of interest or all are independent of the funding bodies.

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