



EFFECT OF HIGH DOSE FISH OIL SUPPLEMENTATION ON CEREBRAL BLOOD FLOW AND COGNITIVE PERFORMANCE IN PATIENTS WITH MILD COGNITIVE IMPAIRMENT: A PROOF OF CONCEPT STUDY

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Abstract: *Objective:* The current study aimed to examine the effect of n-3 polyunsaturated fatty acids (PUFAs) on cerebral blood flow and age-related loss of cognitive functioning in subjects diagnosed with mild cognitive impairment (MCI). *Design, setting and participants:* A total of 20 patients with single or multiple domain MCI took part in this randomized, double-blind, placebo-controlled trial, wherein we investigated the effects of four weeks of daily supplementation with either a high pharmacological dose of 3 g eicosapentaenoic acid (EPA) plus docosahexaenoic acid (DHA) (n=10) or placebo (n=10) on cerebral blood flow and cognitive performance. *Measurements:* Cerebral blood flow and cortical oxygenation were measured by Transcranial Doppler and Near Infrared Spectroscopy together with blood pressure measurements using Finapres. Cognitive function was assessed by sensitive neuropsychological tests measuring reaction times, episodic memory and attention. *Results:* Daily supplementation with EPA+DHA for 4 weeks had no effect on cerebral hemodynamics or blood pressure. All subjects improved on most of the neuropsychological tests, but there was no difference between the fish oil and the placebo groups. *Conclusions:* In this randomized controlled trial in MCI patients, daily supplementation with EPA+DHA for four weeks had no effect on cerebral hemodynamics, blood pressure, or cognitive performance.

Key words: EPA, DHA, cerebral blood flow, cognitive functioning, MCI patients.

Introduction

Considering the large and continuously growing number of people with dementia and the lack of effective medication, prevention of cognitive impairment is of utmost importance. Even modest delays in the onset and progression of Alzheimer's Disease (AD) can significantly reduce the global burden of this disease (1). Prevention trials should preferably focus on people at increased risk of developing AD, such as patients diagnosed with mild cognitive impairment (MCI) (2), an intermediate stage between normal cognitive aging and dementia with a high conversion rate to AD (3).

The intake of n-3 polyunsaturated fatty acids (PUFA),

eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), which are found abundantly in fish oils in particular, is considered a potent modifiable risk factor for cognitive decline. Several observational studies examining cognitive performance in relation to dietary intake of n-3 PUFA or n-3 PUFA concentrations in the blood have been performed of which the results have been summarized in different reviews (4-6). The most recent review by van de Rest et al reports on nine cross-sectional and 28 prospective, observational studies, of which the majority showed protective effects of higher n-3 PUFA in diet or blood against dementia, AD, or cognitive decline (7). Trial evidence is based on 13 RCTs with 6 observing a small beneficial effect of n-3 PUFA treatment on cognitive performance (7, 8). Based on these studies it has been indicated that there is at best a modest clinical benefit of n-3 PUFA and it is suggested that supplementation may be particularly effective in more vulnerable subjects who have not yet been diagnosed with AD, such as elderly with memory complaints or MCI patients.

Several mechanisms could explain the relationship of n-3 PUFA with cognitive performance and dementia.

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DHA is the predominant n-3 fatty acid in the brain and an integral component of neural membrane phospholipids (9). Both EPA and DHA may reduce oxidative stress, are anti-inflammatory and have been linked with neurotransmission, membrane fluidity, ion channel and enzyme regulation and gene expression (10). N-3 PUFA have also been linked to vascular risk factors, which also contribute to the development and progression of AD (11). The effect of cerebrovascular disease might also be caused by impairments in cerebral hemodynamics, such as a decline in cerebral blood flow (CBF) with advancing age (12). Several studies have described the influence of n-3 PUFA on cerebral circulation and this includes effects on reductions in platelet agonist thromboxane A₂ synthesis (13), blood pressure (14), epinephrine and norepinephrine concentrations (15), and blood viscosity (16). Two studies observed that EPA might increase the oxygenation level (17) and that DHA supplementation increased oxygenated hemoglobin (Hb) and total levels of Hb, indicative of increased CBF in healthy adults (18).

In the present study, we assessed the effect of supplementation with a high dose challenge of EPA+DHA on cerebral blood flow, cortical oxygenation, and cognitive performance in subjects with MCI.

Methods

Participants

Patients with amnesic MCI were recruited by the Radboud Alzheimer Centre Nijmegen and hospital Gelderse Vallei at Ede, the Netherlands. Men and women diagnosed as having amnesic (single or multiple) MCI were included. The diagnosis of MCI was based on a multidisciplinary approach, including extensive cognitive testing, neuroradiological findings and medical examination. Cognitive impairments had to be present in one or more cognitive domains (cut-off of 1 standard deviation below age and education adjusted normative mean, including episodic memory (single or multiple-domain amnesic MCI, according to Busse A et al (19)). Furthermore, patients were excluded if 1] no principal caregiver willing to assist for a successful participation was present; 2] they used fish oil supplements; 3] they consumed fish >2 times/week; 4] they used dementia (Alzheimer) medication, Acenocoumarol or other anti-thrombotic drugs; 5] they had a serious liver disease; or 6] they consumed >4 glasses of alcohol per day. After screening, eligible subjects started with a run-in period of one week to get used to swallowing six capsules/day. This study was conducted according to the guidelines laid down in the Declaration of Helsinki and all procedures involving human patients were approved by the Medical Ethical Committee region Arnhem-Nijmegen. Written informed consent was obtained from all patients.

Sample size calculation was based on the Digit Span test. A difference in score of 2 points on the Digit Span was considered clinically relevant. The mean and SD for non-cognitively impaired elderly are 8.5 and 2.0 respectively (20). With these assumptions we calculated that a minimum sample size of 17 participants per group would be required to detect an effect with a power of 80% at a significance level of 0.05. Taking into account an anticipated dropout rate of 15%, 20 participants per treatment group would be needed. However, after two years of extensive screening only 20 eligible participants could be recruited. According to a post-hoc power calculation this provided adequate power to detect an effect size of $d = 1.33$ with all cases included in the analyses.

Intervention

For the intervention period of four weeks, participants were randomly allocated to either treatment with capsules with a high dose challenge of 3 g EPA/DHA or placebo capsules. Randomization was performed by an independent person and taking into account stratification by gender and APOE4 status, i.e. carriers and non-carriers of the APOE4 allele. All subjects and researchers were blinded towards the type of treatment assigned to subjects until the end of the study, after blind review of the data had been completed. The oils were administered in six white-colored soft gelatin capsules daily, each containing 900 mg fish oil (EPA:DHA =3:2) and 29 mg of a natural tocopherol mix as antioxidant, or a placebo oil (sunflower oil) (Orthica, Almere, the Netherlands).

Compliance was checked by counting returned capsules and by inspecting the diaries that subjects kept throughout the study to report the intake of the capsules. A participant was considered to be compliant if >80% of the capsules were taken. An objective measure of compliance was obtained by measuring the n-3 fatty acid composition in the serum cholesteryl esters, which directly reflects intake over the past weeks (21).

Cerebral blood flow (CBF), cortical oxygenation, and blood pressure

We used Transcranial Doppler (TCD) to assess cerebral blood flow at baseline and after four weeks of intervention. TCD applies ultrasound with high temporal resolution to measure changes in cerebral blood flow velocity (CBFV) in the middle cerebral artery accessed through the temporal bone ('the temporal window'). Under the assumption that the vessel diameter is constant, changes in CBFV (Cerebral blood flow velocity) represent changes in CBF. In addition, Near-Infrared Spectroscopy (NIRS) (Oxymon, Artinis Medical Systems, The Netherlands) was used to measure cerebral cortical





oxygenation over the frontal cortices. NIRS uses light at near infrared wavelengths to monitor changes in oxygenated and deoxygenated cerebral Hb concentrations. For this purpose, optodes were placed on the skin of the skull overlying the cortical region of interest. Finally, we assessed arterial blood pressure using Finapres (Finapres Medical Systems, Amsterdam).

We used these measures to look at changes between baseline and after four weeks of intervention in blood pressure, CBF, and cortical oxygenation. In addition to these steady-state hemodynamics, we used spectral analysis of the beat-to-beat time series of blood pressure, CBFV and oxygenated Hb to investigate possible changes in spontaneous oscillations (very low frequency, low frequency and high frequency) in these parameters. Oscillations in these frequency bands reflect cardiovascular autonomic control (baroreflex function, parasympathetic and sympathetic action) (22, 23). The transfer of these oscillations in blood pressure into oscillations in cerebral blood flow reflects cerebral autoregulation that in turn is influenced by cerebrovascular smooth muscle function, endothelial function, and vessel stiffness. Transfer function analysis was thus used to investigate cerebral autoregulation by comparing changes in cerebral blood flow oscillations relative to changes in blood pressure oscillations (22, 24-26). In addition, transfer function analysis was used to calculate the phase difference between blood pressure oscillations and oscillations in oxygenated Hb. This phase shift is an important marker for effective autoregulation, and is sensitive to changes in cerebrovascular properties (26, 27).

Neuropsychological tests

Cognitive performance was assessed at baseline and after four weeks of intervention by means of several sensitive neuropsychological tests focusing on memory, reaction times and attention/working memory. As part of the screening the Mini-Mental State Examination (28) and Clock Drawing test (29) were performed.

The forward test of Wechsler's Digit Span task was included as a measure of attention and the backward test as an index of working memory (30). Two subtests of the computerized Test of Attentional Performance (TAP) were administered: Alertness and Flexibility (31). The Alertness test measures simple and cued reaction time and the Flexibility test measures selective attention. The Paired Associate Learning (PAL) subtest of the Wechsler Memory Scale-Revised (WMS-R) was included to assess episodic (associative) memory (32). The total battery of tests required on average 30 minutes.

Blood

During the screening a blood sample was collected into a 10 ml EDTA-vacutainer and stored at -20°C for APOE genotype determination by the polymerase chain reaction-based restriction fragment length polymorphism method and restriction enzyme digestion with Hha I (33) to determine APOE4 status. At baseline and after four weeks of intervention 10 ml blood was collected to determine omega-3 fatty acid composition in cholesteryl esters. Fatty acid composition in cholesteryl esters was analyzed by gas chromatography as described previously (34) at the laboratory of the Division of Human Nutrition, Wageningen University.

Other measurements

Weight was measured to the nearest 0.5 kg with the person dressed in light clothing and without shoes. Height was measured to the nearest 0.1 cm with the person in standing position and wearing no shoes. Information on educational level, smoking behavior, alcohol and fish consumption, medical history and current use of medication was obtained by a structured questionnaire during a personal interview. During the study participants had to report any adverse events in a diary.

Statistical analyses

Differences in baseline values (for all parameters) between the two treatment groups were analyzed with an independent t-test (continuous variables) or chi-square analysis (categorical variables). Changes from baseline to the end of the study within each treatment group, for hemodynamic and cognitive tests, were analyzed using paired samples t-tests. Changes between the fish oil and the placebo group were compared with the independent samples t-test. Alpha was set at 0.05 (two-tailed testing). Statistical analyses were performed using PASW Statistics 18.0.3.

Results

Eligible participants were screened between October 2008 and October 2010 and intervention took place between December 2008 and December 2010. Figure 1 shows the participant flow through the study. Of all subjects only one subject (who was in the fish oil group) dropped out, because of not feeling well. Apart from the individual who stopped treatment prematurely, the average adherence to treatments based on counts of returned capsules was 94%. Compliance was confirmed by a change in the proportion of EPA+DHA in plasma cholesteryl esters of 321% in the fish oil group (from 1.66





± 0.61 to 6.98 ± 1.02 g/100g fatty acids) and -5.6% (from $1.75 \pm .074$ to 1.66 ± 0.61 g/100g fatty acids) in the placebo group. The supplements were well tolerated; adverse events were reported by two participants, one in the fish oil group (feeling warm at night) and one in the placebo group (itch and joint pain). Mean age of the participants was 73.1 ± 8.8 years and 45% were male. The two treatment groups were similar with regard to baseline characteristics as presented in Table 1.

Figure 1
Flow of participants through study

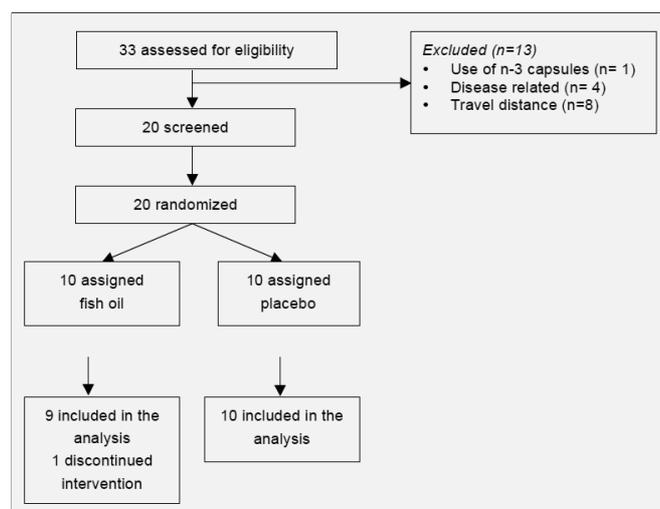


Table 1

Characteristics of 20 subjects with MCI participating in a randomized, placebo-controlled trial, by treatment group*

	Fish oil (n=10)	Placebo (n=10)
Age (years)	72.6 \pm 10.8†	73.6 \pm 6.8
Sex, Male (%)	40	50
Education Low/ Middle/ High (%)	10/ 50/ 40	20/ 40/ 40
BMI (kg/m ²)	25.9 \pm 4.1	26.3 \pm 4.2
Smoking behavior (%)		
Smoker	0	20
Ex-smoker	40	20
Never smokers	60	60
Alcohol consumers (%)	70	70
Alcohol consumption (glasses/ week) ‡ §	7 (2-7)	10 (8-14)‡
Fish consumers (%)	90	90
Fish consumption (times/ month)	3 (3-4)	4 (2-6)
Plasma EPA+DHA (mass%)	1.7 \pm 0.6	1.8 \pm 0.7
APOE ϵ 4 allele 0/ 1/ 2 (%)	50/ 50/ 0	30/ 70/ 0
MMSE (Mini-Mental State Examination)	26.0 (24.8-27.5)	26.5 (24.8-27.0)
Range	24-30	19-30
Clock drawing test (% abnormal)	20	40

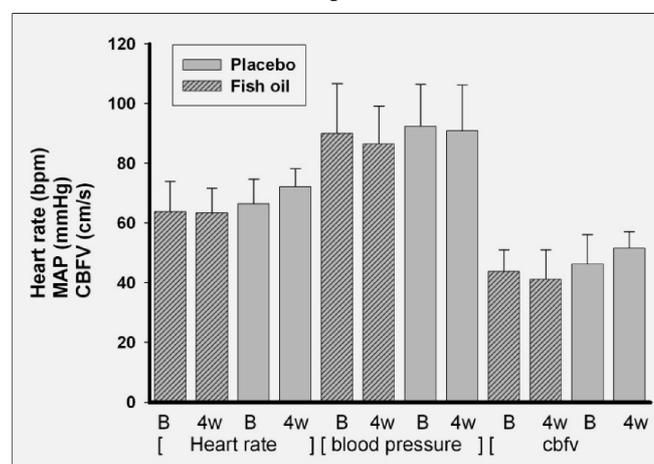
Abbreviations: BMI, Body Mass Index; MMSE, Mini-Mental State Examination; EPA, eicosapentaenoic acid; DHA, docosahexaenoic acid; APOE, apolipoprotein E * There were no significant differences between the two treatment groups (independent samples t-test for continuous variables and chi-square analysis for categorical variables); † Mean \pm SD (all such values); ‡ Mean consumption in consumers only; § Median (Q1-Q3) (all such values); I Missing value for 1 participant

Cerebral measures

Figures 2-5 show that both groups were comparable at baseline for all steady-state systemic and cerebral measures (blood pressure, heart rate, cerebral blood flow velocity) as well as for the dynamic measures obtained through spectral and transfer function analysis (spectral power of blood pressure oscillations and cerebral blood flow oscillations, and phase shift between blood pressure and cortical oxygenation). The intervention with four weeks of fish oil versus placebo did not result in changes in any steady-state or dynamic measures of blood pressure, cerebral perfusion or cerebrovascular autoregulation.

Figure 2

Effects of fish-oil on heart rate, blood pressure and cerebral perfusion



Legend: mean heart rate, mean arterial blood pressure (MAP) and cerebral blood flow-velocity (CBFV) in subjects with mild cognitive impairment, at baseline (B) and after 4 weeks (4w) of fish oil (shaded bars) or placebo (gray bars). Values are mean, error bars represent standard deviation. There were no significant differences between groups or between baseline and follow-up.

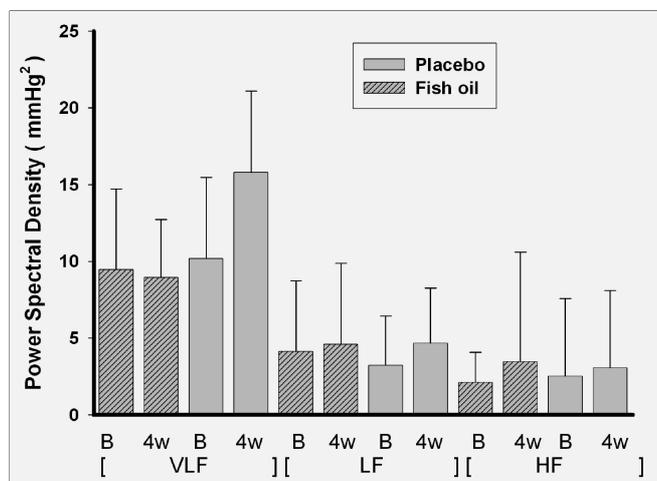
Neuropsychological tests

Baseline scores on the cognitive tests were comparable between the two groups, except for the TAP flexibility tests letters ($P=0.03$ [95% CI -214 - 943]) and letter-digit alternating ($P=0.05$ [95% CI -589 - 1622]). After four weeks of intervention participants in both treatment groups improved significantly on the Digit Span forward test and almost all TAP tests, except non-cued Alertness in the fish oil group. Contrarily, on the Digit Span Backward and especially on the Paired Associate Learning test they performed worse, but only in the fish oil group these declines were significant (Table 2). However, compared to the placebo group, participants in the fish oil group did not change significantly, except for TAP Flexibility Letters where the placebo group performed faster after 4 weeks of intervention, $P=0.01$; -334 [95% CI -805-136] (Independent-samples t-test).



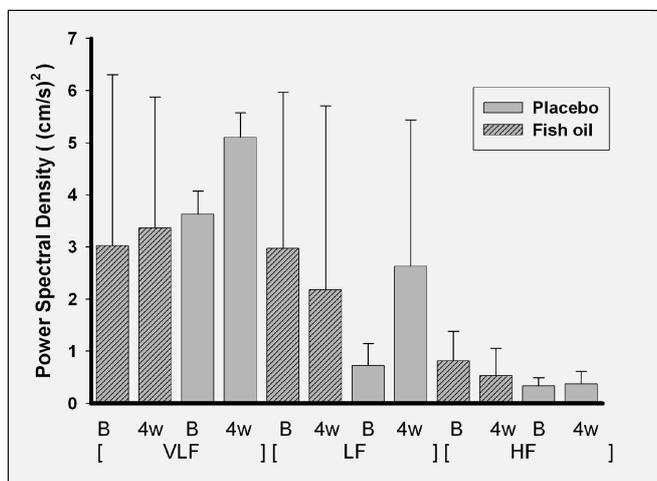


Figure 3
Effects of fish-oil on spontaneous blood pressure oscillations



Legend: results of spectral analysis of 5 min of continuous (beat-to-beat) blood pressure recordings, showing the power spectral density of spontaneous oscillations in blood pressure. Three commonly used frequency bands are shown: very low frequency (VLF), low frequency (LF) and high frequency (HF). Subjects with mild cognitive impairment, at baseline (B) and after 4 weeks (4w) of fish oil (shaded bars) or placebo (gray bars). Values are mean, error bars represent standard deviation. There were no significant differences between groups or between baseline and follow-up.

Figure 4
Effects of fish-oil on spontaneous cerebral blood flow oscillations



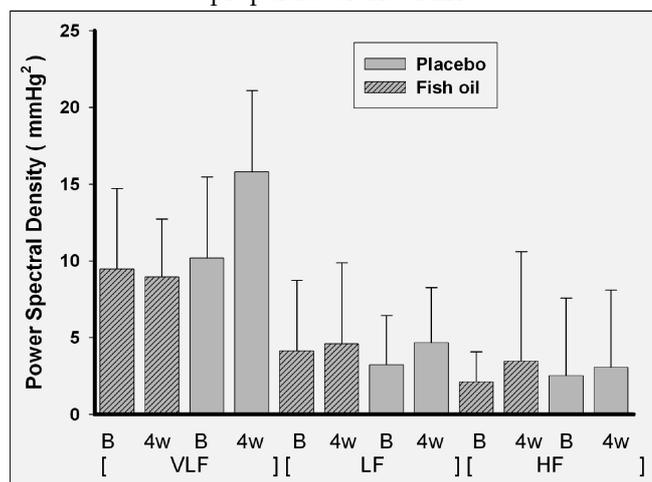
Legend: results of spectral analysis of 5 min of continuous (beat-to-beat) cerebral blood flow-velocity recordings, showing the power spectral density of spontaneous oscillations in cerebral blood flow-velocity. Three commonly used frequency bands are shown: very low frequency (VLF), low frequency (LF) and high frequency (HF). Subjects with mild cognitive impairment, at baseline (B) and after 4 weeks (4w) of fish oil (shaded bars) or placebo (gray bars). Values are mean, error bars represent standard deviation. There were no significant differences between groups or between baseline and follow-up.

Discussion

In this randomized controlled trial in MCI patients, daily supplementation with EPA+DHA for four weeks

had no effects on systemic and cerebral hemodynamics. The intervention also had no effect on cognitive performance. All participants improved significantly on most of the neuropsychological tests, but no differences were found between the fish oil and placebo groups.

Figure 5
Effects of fish oil on microvascular autoregulatory properties of the brain



Legend: results of spectral analysis of 5 min of continuous (beat-to-beat) recordings of spontaneous oscillations in blood pressure and cortical oxygenation. The graph shows the phase shift between oscillations in blood pressure and cortical oxygenation, derived from transfer function analysis. Three commonly used frequency bands are shown: very low frequency (VLF), low frequency (LF) and high frequency (HF). Subjects: mild cognitive impairment, at baseline (B) and after 4 weeks (4w) of fish oil or placebo. Black squares are mean values, gray triangles indicate 95 % confidence interval. The negative phase shift in LF and HF reflects a combination of transit time for oxygenated blood from conduit vessels to cortical brain tissue, and a progressive reduction in autoregulatory capacity with increasing frequency (from VLF to LF to HF). The much smaller phase shift in VLF (around zero) is caused by optimal autoregulatory properties of the brain microvasculature at this frequency. Hence, any beneficial cerebrovascular effects of fish oil were expected to lead to a positive phase shift (i.e. a phase shift becoming less negative or more positive). There were no significant differences between groups or between baseline and follow-up.

This study has been designed as a challenge study: to examine rapid changes in response to a high pharmacological treatment dose in a sensitive group of patients. Therefore, the duration of the study is limited, but previous studies showed that near-maximal incorporation of EPA and DHA in serum cholesteryl esters (21), plasma phospholipids and blood mononuclear cells (35) is already reached after four weeks of supplementation. In contrast, we intervened with a rather high dose of fish oil (3 g EPA+DHA daily) in patients with MCI, a transition stage between normal cognitive aging and dementia, characterized specifically by memory impairment beyond that expected for age and education (36). From previous (epidemiological) studies it appears that subjects sensitive to progression, such as MCI subjects, are the preferred group to investigate (37). In comparison to several other trials investigating the effect of fish oil supplementation on cognitive performance our trial is a relatively small study. Our



**Table 2**

Scores on cognitive tests of 20 subjects with MCI at baseline and after 4 weeks of EPA+DHA supplementation, by treatment group

	Fish oil (n=10)			Placebo (n=10)			Fish oil vs placebo Mean difference (95% CI)
	Baseline	4 weeks†	Change	Baseline	4 weeks	Change	
Digit Span Forward, # words	7.9 ± 1.8‡	8.6 ± 1.7	0.6 ± 0.7*	7.8 ± 1.2	8.0 ± 1.2	0.2 ± 0.9*	-0.36 (-1.16 – 0.45)
Digit Span Backward, # words	6.0 ± 2.4	5.7 ± 1.8	-0.4 ± 1.2*	5.2 ± 1.7	4.8 ± 1.8	-0.4 ± 1.5	0.04 (-1.30 – 1.39)
Paired Associate Learning, total	10.8 ± 4.8	8.9 ± 3.2	-2.2 ± 2.7*	11.2 ± 3.9	8.0 ± 2.6	-3.2 ± 3.6	-0.98 (-4.06 – 2.10)
Paired Associate Learning, easy	15.4 ± 3.0	13.6 ± 2.0	-1.7 ± 1.8*	14.9 ± 2.2	12.8 ± 3.1	-2.1 ± 3.1	-0.43 (-2.95 – 2.09)
Paired Associate Learning, difficult	3.1 ± 3.8	2.1 ± 2.8	-1.3 ± 2.0*	3.7 ± 3.0	1.6 ± 1.6	-2.1 ± 2.5	-0.77 (-2.96 – 1.43)
TAP Alertness, simple RT (ms) §	361 ± 120	330 ± 73	-27 ± 95	328 ± 114	323 ± 84	-5 ± 72*	22 (-59 – 103)
TAP Alertness, cued RT (ms)	330 ± 74	311 ± 77	-13 ± 41*	323 ± 98	333 ± 93	11 ± 55*	23 (-24 – 71)
TAP Flexibility, letters (ms)	656 ± 308	556 ± 82	-6 ± 42*	1021 ± 814**	680 ± 217	-340 ± 666**/**	-334 (-805 – 136)
TAP Flexibility, digits (ms)	633 ± 192	555 ± 75	-31 ± 63*	814 ± 496	665 ± 224	-148 ± 324*	-117 (-349.8 – 115)
TAP Flexibility, letter-digit (ms)	1531 ± 897	1003 ± 327	-305 ± 369*	2048 ± 1401**	1667 ± 1034	-382 ± 763*	-77 (-668 – 515)

Abbreviations: TAP, Attention Test Battery; APOE, apolipoprotein E; † N=9; ‡ Mean ± SD (all such values); § For all TAP tests: higher scores indicate more time needed to complete the task, i.e. poorer performance; * Significant change from baseline after 4 weeks of intervention; ** Significant difference or change between the fish oil and the placebo group (Independent samples t-test, P<0.05).

power analysis was based on the Digit Span cognitive tests and at least 17 subjects per group would be required. Unfortunately, despite a long recruitment period of two years, only 10 subjects per group that fulfilled our study criteria and were willing to participate were found. This would, however, still be sufficient to identify an effect size of 1.33. Moreover, the results do not show a trend towards a significant difference. Also, besides studying cognitive performance, we also investigated possible underlying mechanisms by using TCD, NIRS, and Finapres. To study the detailed information that these methods provide requires extensive and time-consuming data analysis and therefore such detailed studies are limited to small numbers of participants, where sufficient power is achieved with sample sizes as small as n= 8 (24-26). Neuropsychological tests were included to see whether changes in cerebral blood flow would also result in measurable changes on these tests, which are more sensitive than cognitive screens such as the MMSE.

Both groups improved significantly on the neuropsychological tests, which could be due to nonspecific learning effects, although parallel versions were used to take material-specific learning effects into account. The fact that all improved could also be the reason that no additional effect of fish oil compared to placebo was found. Unexpectedly, both groups performed worse on the episodic memory test after 4 weeks of intervention.

In this study stratified randomization was used based on gender and APOE4 status. It is known that carriers of the APOE-ε4 allele have a higher risk of developing dementia and moreover several studies suggest that APOE genotype affects responsiveness to EPA+DHA intake and its subsequent effect on cognitive functioning. However, the role of APOE4 status is contradictory and

merits further research as studies show that EPA+DHA intake may either protect against cognitive impairment in non-carriers of the APOE-ε4 allele (38-42) or in carriers (20, 43, 44). In our study there was no significant interaction between APOE4 status and treatment.

To the best of our knowledge there are no other trials studying the effects of fish oil supplementation that included TCD measurements and only two other trials (17, 18) that used NIRS to gain more insight in one of the possible mechanisms explaining the potential association between n-3 fatty acids and cognitive performance. They both observed an increase in oxygenation level after supplementation with either EPA or DHA, which is indicative of an increased CBF. Gaining more insight in vascular disease related mechanisms is of clinical relevance, considering the importance of vascular disease in the etiology of AD, frequent occurrence of cerebrovascular comorbidity in AD, and in the growing interest in the use of fish oil on (vascular) cognitive impairment. However, in this study we did not confirm the earlier observation that supplementation with either EPA or DHA increased CBF. There were no changes in cerebral blood flow-velocity, as measured with TCD in either placebo or intervention group. There was also no indication of more subtle positive effects on the cerebral vasculature, as we observed no changes in spontaneous oscillations in cerebral blood flow, and no effect on cerebral autoregulation as measured with TCD (cerebral blood flow-velocity) and NIRS (oxygenated Hb).

Our method is based on the assumption that an increase in CBF improves cognition. Direct evidence for this assumption is lacking, although the available circumstantial evidence is highly suggestive. In the Rotterdam Study individuals with cognitive decline were found to have lower CBF than the individuals who had stable cognitive function in the previous years (45). The





mechanism for this relationship is likely to be the increasing sensitivity of neurons to ischemia or hypoperfusion with age (46). The evidence for the causal relationship between impairment in CBF, neuronal injury, and cognitive decline has recently been reviewed elsewhere (47, 48). Hooijmans et al studied the effects of dietary lipids (cholesterol and DHA) on A β accumulation and brain circulation in mice (49). They observed that the DHA-enriched diet increased relative cerebral blood volume without changing blood flow indicating a larger circulation in the brain probably due to vasodilation, and decreased the amount of vascular β -amyloid deposition.

We and others have shown earlier that AD is associated with reduced cerebral perfusion and increased cerebrovascular resistance (50-52). More recently, it was shown that MCI patients show changes in cerebral hemodynamics that lie in between normal controls and AD. In addition, we found that AD patients exhibit impairments in cardiovascular autonomic control (baroreflex) and that, again, MCI patients show changes that are between normal controls and AD. Therefore, we argue that cardiovascular and cerebral hemodynamics in our MCI patients were plausible targets that had a theoretical potential for improvement, e.g. in the form of reduction in cerebrovascular resistance, increase in cerebral blood flow, or changes in cardiovascular autonomic control.

In summary, in this randomized controlled trial performed in a small group of MCI patients, we observed no effect of daily supplementation with EPA+DHA for four weeks on cognitive performance, cerebral hemodynamics or blood pressure.

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