ALINEA

NUTRITION

JUNE 2023

www.alineanutrition.com

TABLE OF

CONTENTS

He Y, Huang SY, Wang HF, Zhang W, Deng YT, Zhang YR, Dong Q, Feng JF, Cheng W, Yu JT. Circulating polyunsaturated fatty acids, fish oil supplementation, and risk of incident dementia: a prospective cohort study of 440,750 participants. Geroscience. 2023 Apr 12. doi: 10.1007/s11357-023-00778-6.

What We Know, Think We Know, or Are Starting to Know

Up to ~60% of the human brain is composed of lipids [fat], of which around half consists of essential polyunsaturated fatty acids [PUFA], specifically the omega-3 docosahexaenoic acid [DHA] and the omega-6 arachidonic acid [AA] [(1)]. These long-chain fatty acids are rapidly incorporated into central nervous system cells, and play a critical role in development of the human brain and central nervous system through gestation and into infancy [(2)].

In relation to the early life-stage, there is little debate about the role, and need, for long-chain PUFA. Where the evidence gets slightly more ambiguous, however, is at the other end of the lifecycle; are PUFA still beneficial for the brain at that stage in life? This question is pertinent given the predicted dramatic rise in Alzheimer's Disease incidence by 2060 [(3)], and absence of effective available pharmacological interventions [(4)].

This question has been difficult to tease out due to some common challenges that face nutrition research. The first is the temporal relationship between diet, nutrients, and disease outcomes; neurodegenerative disease may take years to develop, and studying healthy adults over a short period in a controlled trial may not produce any detectable effects [(5)]. The second is that, conversely, studying adults that have already developed neurodegenerative disease may be too late for any intervention to be effective [(6)].

Practically, this means that to study the effects of PUFA on neurodegenerative disease risk requires nutritional epidemiology to utilise prospective studies. Increasingly, cohorts that have been recently established have conducted more extensive baseline assessments of participants, including clinical evaluations of participants' health status. The present study utilised the UK Biobank cohort to investigate effects of PUFA on dementia risk.

The Study

The UK Biobank cohort recruited over half-a-million participants across England, Wales, and Scotland between 2006–2010. All participants completed a range of in-person assessments at baseline, including biological samples for biomarkers of nutritional status* [see ***Geek Box**, below, for further details], immune markers, and brain imaging. The present study investigated the associations between:

- **Exposure**: There were two distinct exposures of interest, including:
    - **Plasma PUFA**: Plasma levels of total PUFA, omega-3 PUFA, DHA, omega-6 PUFA, and linoleic acid [LA].
    - **Fish Oil [FO] Supplementation**: FO supplementation was assessed at baseline as a dichotomous "Yes/No" question regarding FO supplement use.
- **Outcome:** Overall dementia incidence was the primary outcome. A subgroup analysis also investigated the two most prevalent dementia subtypes, Alzheimer's Disease [AD] and vascular dementia [VD].

The study also analysed associations between plasma PUFA, FO supplementation, and brain structure assessed with neuroimaging scans. Finally, a mediation analysis was conducted to determine whether plasma PUFA levels mediated the association between FO and dementia risk.

*Geek Box: Biomarkers of Dietary Assessment

The term "biomarker" means use of a specific biochemical measure that provides an indication of nutrient intakes. This isn't always as straightforward as "nutrient in = nutrient measured" because nutritional status is influenced by variations in the digestion, absorption, metabolism, distribution, and excretion of a nutrient, which differs from nutrients to nutrient. For example, when measuring fatty acids, whether it is red blood cells, phospholipid content of cell membranes, lipoproteins, or adipose tissue measured, each will provide different indications of dietary intake.

Biomarkers may be classified according to what measurement they allow for. A biomarker for which there is a quantitative relationship between dietary intake and the value of the biomarker, such that absolute intake over a 24 h period can be measured accurately, is known as a "recovery biomarker". "Recovery" reflects the fact that all intake over a 24 h period is excreted, usually through urine, with minimal losses through other excretory pathways. These are very rare in nutrition science: only 24 h urinary sodium, 24 h urinary potassium, 24 h urinary nitrogen, and total energy measured by doubly-labelled water, are considered recovery biomarkers.

The most commonly used biomarkers, which measure the concentration of a nutrient in plasma, red blood cells, adipose tissue, etc., are known as "concentration biomarkers", as they are measuring the concentration of that specific nutrient in the circulation or tissue. The use of biomarkers is very attractive for nutritional epidemiology, as it allows for an objective assessment of the validity of dietary questionnaires, and quantification of intake that is independent of measurement error.

However, there remain limitations to their application. First, there is not a reliable biomarker for every nutrient of interest to nutrition science. Secondly, many non-dietary factors may influence the status of a biochemical indicator, thus introducing a potential measurement error that is unrelated to actual dietary intake. Nonetheless, for exposures of interest like sodium, potassium, fatty acids, or total energy expenditure, biomarkers are reliable, and provide a means of quantifying accurate dietary intake.

Results: For the final analysis, 102,722 participants were included in the plasma PUFA analysis, and 425,374 participants were included in the FO supplement analysis. The average age of participants at baseline was 56yrs, and ~54% were female. The average follow-up duration was 9.2yrs, during which there were 7,768 incident cases of dementia.

***Associations Between Plasma PUFA and Dementia Risk*:** Higher plasma concentrations of all five PUFA biomarkers measured were associated with lower dementia risk:

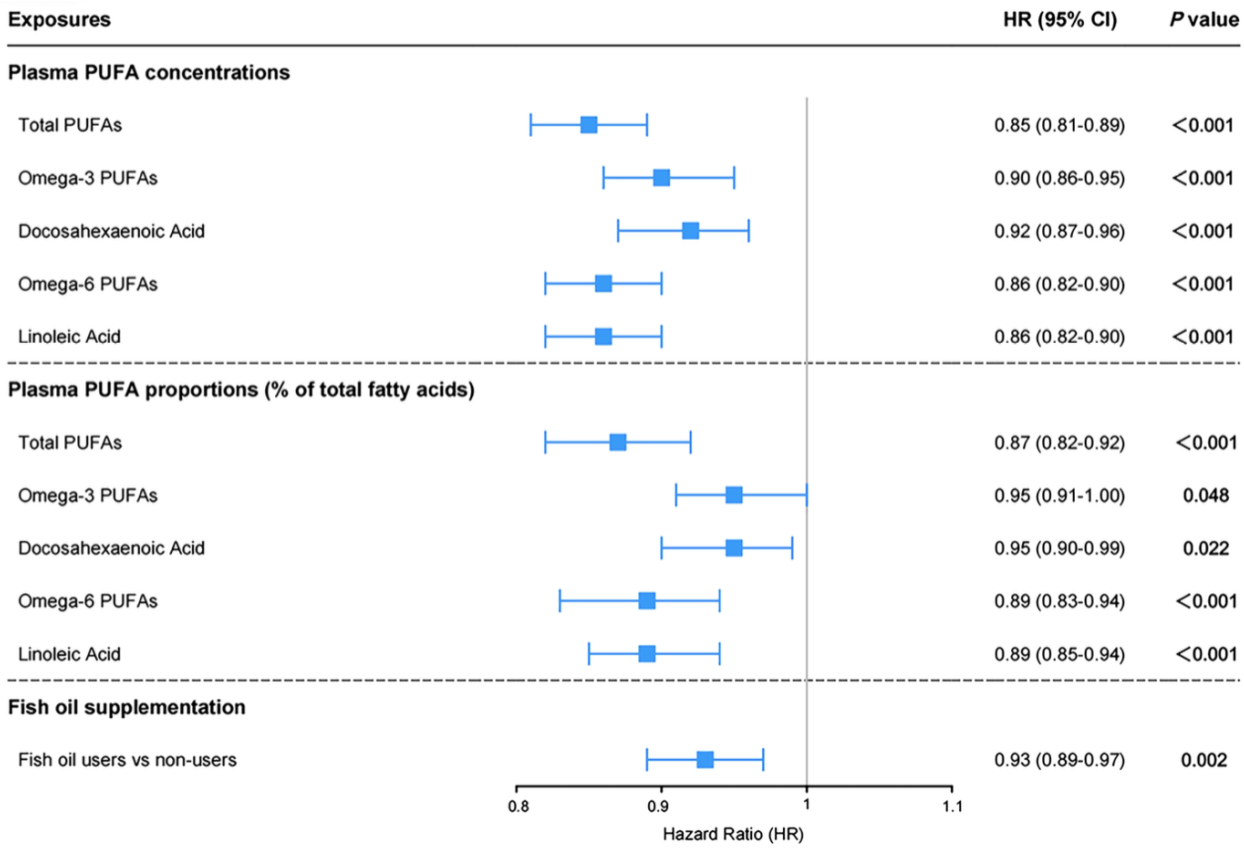
- Total PUFA were associated with a 15% [95% CI, 11% to 19%] lower risk.
- Omega-3 PUFA with a 10% [95% CI, 5% to 14%] lower risk.
- DHA with an 8% [95% CI, 4% to 13%] lower risk.
- Omega-6 PUFA with a 14% [95% CI, 10% to 18%] lower risk.
- LA with 14% [95% CI, 10% to 18%] lower risk.

When analysed as a proportion of all fatty acids in the plasma, the results were similar overall except for an attenuated strength of the effect estimate for omega-3 PUFA.

These findings remained similar after adjusting for age, sex, intake of oily fish, vegetables, fruit, social deprivation index, education status, and other relevant covariates for dementia risk. The findings also were unchanged after excluding dementia cases that occurred in the first 2yrs of follow-up.

***Associations Between FO Supplementation and Dementia Risk*:** FO supplementation was associated with a 6% [95% CI, 1% to 11%] lower risk of dementia, after adjusting for the covariates referred to above.

In a subgroup analysis by dementia type, FO supplementation was associated with a 14% [95% CI, 4% to 23%] lower risk of VD, but was not associated with lower risk of AD. In a sensitivity analysis excluding participants who also reported vitamin or mineral supplement use, the association with FO supplementation was attenuated and no longer significant.



***Forest plot** from the paper illustrating the effect estimates [**square box**] and 95% confidence intervals [**arms left-right from box**] for the exposures of interest in the present study. The top five rows indicate the risk estimates for the 5 plasma PUFA biomarkers assessed as total plasma concentrations; the middle five represent the 5 plasma PUFA biomarkers assessed as a proportion of all fatty acids in plasma. The final row is the risk estimate for FO supplementation.*

Mediation Between Plasma PUFA, FO Supplementation, and Dementia Risk*:** In the analysis to detect whether plasma PUFA measures mediated the association between FO supplementation and dementia risk, omega-6 PUFA mediated 4.39% of the association, total PUFA's 29%, omega-3 PUFA's 57.99%, and DHA 56.95% [discussed further under ***Key Characteristic, below].

***Analysis of Plasma PUFA, FO Supplementation, and Brain Structure*:** Higher plasma levels of total PUFA, omega-6 PUFA, and LA were associated with larger volumes of brain cortical and subcortical regions, brain regions damaged in dementia/AD. There were also associations between total PUFA, omega-6 PUFA, LA, and higher brain white matter structures related to cognition and psychological functioning. However, there were no significant associations for FO supplementation and brain structure measures.

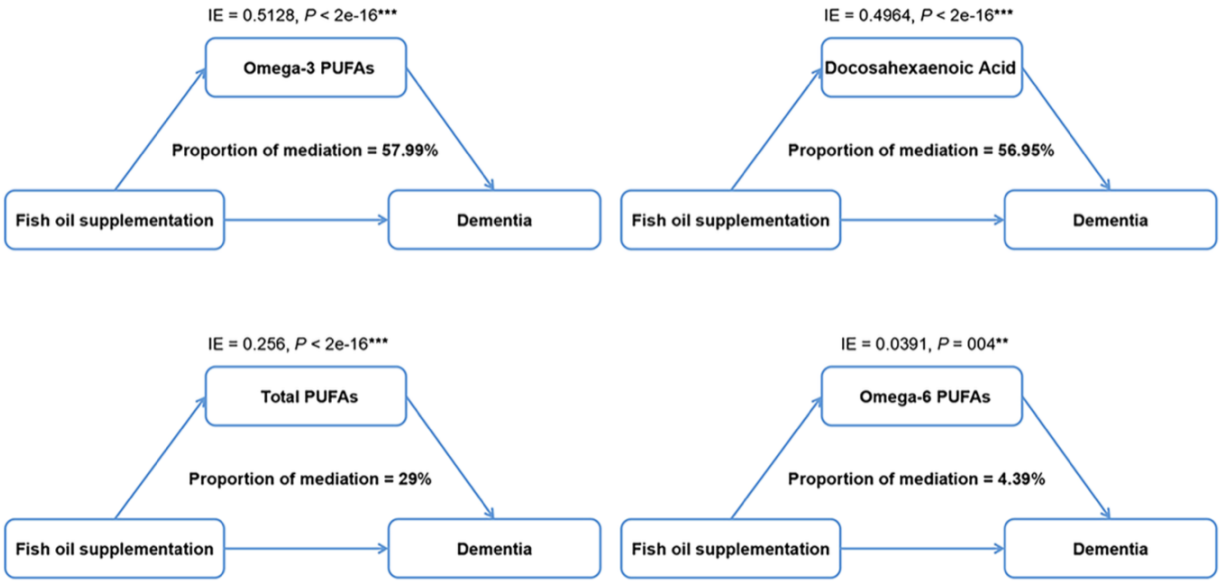
The Critical Breakdown

Pros: The study's aims were clearly stated, and included distinct analyses between biomarkers of PUFA status and FO supplementation as exposures. The mediation analysis provided additional validity of the associations for FO supplementation and dementia risk [discussed further under ***Key Characteristic***, below]. The sample sizes for both analyses were vast, particularly for the FO analysis with >400k participants. The sample was also balanced for sex, which is important given dementia risk is higher in women compared to men [7]. The analyses adjusted for relevant covariates for dementia risk, including adjusting for intakes of fatty fish as the main dietary source of long-chain omega-3 PUFA, and which is also associated with lower dementia/AD risk [8,9]. It also adjusted for *ApoE4*, a gene that regulates cholesterol metabolism in the brain and ~40% of AD patients carry this variant [10]. The results of the primary analysis remained largely robust to adjustment for these, and other, relevant covariates. The analysis also considered relevant subgroups [e.g., age, sex, *ApoE4* status] and sensitivity [e.g., exclusion of the first 2yrs of follow-up to minimise reverse causality] analyses.

Cons: Given that both the prevalence and incidence of dementia increase exponentially after 75yrs of age, the average age of the cohort in the present study of 56yrs at baseline was potentially too young relative to the outcome of interest to provide more robust statistical power. Given the age at baseline, the follow-up duration of ~10yrs, while looking adequate on paper, is slightly short for this specific outcome, and this is reflected in the relatively low incidence of dementia. Fish oil supplementation was categorised dichotomously with no additional information related to dose, and no repeated assessment to determine whether supplementation was maintained over the duration of follow-up. The biomarker analysis only assessed plasma PUFA, which is only a reflection of immediately recent [i.e., ~2-3 days] dietary intake, and are not a reliable indicator of longer term, habitual diet compared to other biomarkers [e.g., red blood cells or adipose tissue] [11].

Key Characteristic

Given the exposures of interest in the present study were not dietary intake of PUFA, but plasma PUFA as a biomarker and use of FO supplements, which would be expected to influence endogenous PUFA status, the use of a mediation analysis was insightful. The **figure** below illustrates the outcomes of these analyses for omega-3 PUFA [**top left**], DHA [**top right**], total PUFA [**bottom left**], and omega-6 PUFA [**bottom right**].



A mediation analysis considers how a third variable influences a relationship between two other variables. To understand how this is achieved, let's take the example of omega-3 PUFA. If you look at that figure, you'll see arrows pointing in a specific direction. A mediation analysis must start with a significant association between your two main variables, in this case FO supplementation and dementia.

Once that relationship is present and significant, the next step is to determine the relationship between FO supplementation and the third, mediating variable – in this case plasma omega-3 PUFA. If this is present and significant, the final step is to determine the strength of the association between the mediator and outcome, i.e., between plasma omega-3 PUFA and dementia.

In effect, this is testing whether the effect of FO supplementation on dementia risk goes through plasma omega-3 PUFA. If there is a full mediating effect, the effect of FO supplementation on dementia risk would completely disappear. If there is a partial mediating effect, the independent effect of FO supplementation on dementia is weakened, i.e., it is the mediator that explains a proportion of the overall effect of FO supplementation on dementia risk.

In this analysis, we can see that omega-6 PUFA mediated just 4% of the association between FO supplementation on dementia; but FO supplements don't contain any omega-6, so we wouldn't expect there to be much of any mediating effect here [and I'm not really sure why they ran this analysis].

Interesting Finding

There are several findings from this overall analysis that provide some interesting additional insights. The first relates to one of our stated **Cons**, above, which is the overall younger age profile of the cohort relative to the prevalence and incidence rates for the outcome of dementia. In a subgroup analysis by age of participants, stratified by over or under 65yrs, the association between FO supplementation and dementia was significant only in the >65yrs, a 9% [95% CI, 3% to 15%] lower dementia risk.

The second is the subgroup analysis by oily fish intake, stratified by <1 or >1 serving per week. In this analysis, the effect of FO supplementation was only significant in participants consuming <1 oily fish meal per week, with a 14% [95% CI, 6% to 21%] lower risk of dementia.

This is important, as an issue across all of nutrition research is that individuals with adequate nutrient status are unlikely to benefit from further increasing intakes of that nutrient beyond required levels [12,13]. And both biomarker studies [14] and intervention trials [15] suggest that those most likely to benefit from omega-3 supplementation are those with low dietary intake and endogenous omega-3 status.

The final interesting finding is the subgroup analysis by *ApoE4*, stratified by carriers or non-carriers; the association between FO supplementation and dementia risk was significant in *ApoE4 carriers*, but not non-carriers, with an 8% [95% CI, 1% to 15%] lower risk in *ApoE4-carriers*. This is instructive because *ApoE4-carriers* have lower omega-3 status and responsiveness to supplementation [16], while high-dose DHA supplementation may improve cognitive function in *ApoE4-carriers*, but not *ApoE4-non-carriers* [17].

Relevance

Although this study has the major strength of its enormous sample size, it is far from providing more definitive answers to some of the open questions on the potential protective effects of PUFA against dementia risk. The strongest, and most robust, effect estimates in the present study were for plasma PUFA, and FO supplementation was a more modest 6% lower dementia risk. The major limitation is what plasma PUFA represents as a biomarker, i.e., immediate dietary and supplemental intake [11,18].

Tracer studies have shown that the whole-body half-life of omega-3 PUFA is ~54 days following 3.2g/d omega-3 supplementation [18]. The fact that this study is a decade of follow-up, with supplementation only assessed as a "Yes/No" question and no further detail on dose, limits the inferences we might take. The fact that the effect of FO supplementation was no longer significant after excluding participants using micronutrient supplementation may also reflect the fact that the effects of omega-3 PUFA interacts with other nutrients, B-vitamins in particular [19].

Further, of this minor lower risk associated with FO supplementation, over half of this was explained in the mediation analysis by plasma omega-3 PUFA and DHA. Thus, the weight of the findings for the present study relates to the biomarkers of plasma PUFA status, with its inherent limitations for making inferences about dementia risk a decade later.

It is also interesting to note that while omega-3 PUFA tend to take the lion's 2share of focus for brain health, omega-6 PUFA and LA were associated with lower dementia risk in the biomarker analysis, and greater brain cortical volume in the imaging analysis. It is often overlooked that omega-6 PUFA do play important roles in the brain, and the brain appears to exhibit a particular ratio of DHA to AA, at least in developmental stages [(2)]. However, given that LA does not convert in any appreciable amount to AA [(20)], the precise relationship with LA in particular remains to be determined.

Application to Practice

Despite the continued lack of definitive answers, the weight of evidence is more consistently tipping in the direction of a benefit to long-chain PUFA of the omega-3 variety, which is evident in biomarker studies of brain structures and of dementia risk [(21,22)].

However, this does not appear to be a general benefit, and there do appear to be particular characteristics that may better predict a benefit to higher omega-3 PUFA intake, whether achieved from diet or supplementation; those who consume low, or no, oily fish; those with low total omega-3 intakes, individuals that are *ApoE4-carriers*, and elderly adults who are otherwise healthy and low in omega-3, or exhibit some signs of mild cognitive impairment.

Thus, depending on the population you may work with in practice, these factors should be considered in the evaluation of whether FO supplementation is required. Generally, for those who do consume oily fish, the recommendation of 1–2 servings a week is sound advice to cover long-chain omega-3 bases.

References

1. McCann JC, Ames BN. Is docosahexaenoic acid, an n□3 long-chain polyunsaturated fatty acid, required for development of normal brain function? An overview of evidence from cognitive and behavioral tests in humans and animals. Am J Clin Nutr. 2005 Aug 1;82(2):281–95.

2. Colombo J, Carlson SE, Cheatham CL, Shaddy DJ, Kerling EH, Thodosoff JM, et al. Long-term effects of LCPUFA supplementation on childhood cognitive outcomes. American Journal of Clinical Nutrition. 2013;98(2):403–12.

3. Brookmeyer R, Abdalla N, Kawas CH, Corrada MM. Forecasting the prevalence of preclinical and clinical Alzheimer's disease in the United States. Alzheimer's & Dementia. 2018 Feb 6;14(2):121–9.

4. Cummings JL, Morstorf T, Zhong K. Alzheimer's disease drug-development pipeline: few candidates, frequent failures. Alzheimers Res Ther. 2014;6(4):37.

5. Dangour, A. D., Allen, E., Elbourne, D., Fasey, N., Fletcher, A. E., Hardy, P., Holder, G. E., Knight, R., Letley, L., Richards, M., & Uauy, R. Effect of 2-y n□3 long-chain polyunsaturated fatty acid supplementation on cognitive function in older people: a randomized, double-blind, controlled trial. Am J Clin Nutr. 2010;91(6):1725–1732.

6. Quinn, J. F., Raman, R., Thomas, R. G., Yurko-Mauro, K., Nelson, E. B., van Dyck, C., Galvin, J. E., Emond, J., Jack, C. R., Weiner, M., Shinto, L., & Aisen, P. S.. Docosahexaenoic Acid Supplementation and Cognitive Decline in Alzheimer Disease. JAMA. 2010;304(17):1903.

7. van der Flier WM. Epidemiology and risk factors of dementia. J Neurol Neurosurg Psychiatry. 2005 Dec 1;76(suppl_5):v2–7.

8. Morris MC, Evans DA, Tangney CC, Bienias JL, Wilson RS. Fish consumption and cognitive decline with age in a large community study. Arch Neurol. 2005;62(12):1849–53.

9. Morris MC, Evans DA, Bienias JL, Tangney CC, Bennett DA, Wilson RS, et al. Consumption of fish and n-3 fatty acids and risk of incident Alzheimer disease. Arch Neurol. 2003;60(7):940–6.

10. Farrer LA, Cupples LA, Haines JL, Hyman B, Kukull WA, Mayeux R, et al. Effects of age, sex, and ethnicity on the association between apolipoprotein E genotype and Alzheimer disease. A meta-analysis. APOE and Alzheimer Disease Meta Analysis Consortium. JAMA. 278(16):1349–56.

11. Arab L, Akbar J. Biomarkers and the measurement of fatty acids. Public Health Nutr. 2002 Dec 22;5(6a):865–71.

12. Morris MC, Tangney CC. A potential design flaw of randomized trials of vitamin supplements. JAMA - Journal of the American Medical Association. 2011;305(13):1348–9.

13. Cunnane SC, Chouinard-Watkins R, Castellano CA, Barberger-Gateau P. Docosahexaenoic acid homeostasis, brain aging and Alzheimer's disease: Can we reconcile the evidence? Prostaglandins Leukot Essent Fatty Acids. 2013;88(1):61–70.

14. Coley N, Raman R, Donohue MC, Aisen PS, Vellas B, Andrieu S. Defining the Optimal Target Population for Trials of Polyunsaturated Fatty Acid Supplementation Using the Erythrocyte Omega-3 Index: A Step Towards Personalized Prevention of Cognitive Decline? J Nutr Health Aging. 2018 Oct 31;22(8):982–8.

References

15. 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 May;97(5):1134–43.

16. Tomaszewski N, He X, Solomon V, Lee M, Mack WJ, Quinn JF, et al. Effect of APOE Genotype on Plasma Docosahexaenoic Acid (DHA), Eicosapentaenoic Acid, Arachidonic Acid, and Hippocampal Volume in the Alzheimer's Disease Cooperative Study-Sponsored DHA Clinical Trial. Journal of Alzheimer's Disease. 2020 Apr 7;74(3):975–90.

17. Danthiir V, Hosking DE, Nettelbeck T, Vincent AD, Wilson C, O'Callaghan N, et al. An 18-mo randomized, double-blind, placebo-controlled trial of DHA-rich fish oil to prevent age-related cognitive decline in cognitively normal older adults. Am J Clin Nutr. 2018 May;107(5):754–62.

18. Plourde M, Chouinard-Watkins R, Rioux-Perreault C, Fortier M, Dang MTM, Allard MJ, et al. Kinetics of 13C-DHA before and during fish-oil supplementation in healthy older individuals. Am J Clin Nutr. 2014 Jul;100(1):105–12.

19. Jernerén F, Elshorbagy AK, Oulhaj A, Smith SM, Refsum H, Smith AD. Brain atrophy in cognitively impaired elderly: the importance of long-chain v-3 fatty acids and B vitamin status in a randomized controlled trial. American Journal of Clinical Nutrition. 2015;102:215–21.