Reviewer's report

Title: Omega-3 fatty acids status in human subjects estimated using a food frequency questionnaire and plasma phospholipids levels

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Reviewer: Allison Hodge

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This paper looks at correlations between intakes of omega-3 fatty acids estimated from FFQ data and the percentage concentrations of these in plasma phospholipid fatty acids. Moderate correlations were found for DHA and total omega-3 fatty acids in men and women, but for women only correlations were also seen for EPA and DPA.

Because of the interest in the potential health benefits associated with omega-3 fatty acids, practical ways to assess intake for epidemiological studies are important, and the use of biomarkers is a valid assessment of intake measurement for these. This paper could be made more useful for others needing to design an FFQ to assess n-3 fatty acids with more detail on the FFQ provided in the methods and considered in discussion. As it is this does not really add anything new to the field, except for those who wish to use the FFQ described to measure n-3 fatty acids.

1. An important limitation of this study is that the numbers are probably too small to look at whether correlations vary by age, smoking status, obesity or health status.

Minor essential revisions:

2. Page 4, line 52. The FFQ cannot estimate the effects of n-3 PUFA on health, but can enable such analyses.

3. The timing of blood sampling and dietary questionnaires is not provided in the methods. This is important as the amount and sources of n-3 in the food supply is probably changing as it is added as a supplement to things. Need to know whether the nutrient composition data used is from around the same time as the FFQ completion and how long blood samples were stored.

4. In methods comment on use of phospholipid fatty acids rather than other blood fraction eg cholesterol esters.

5. Page 6, line 82. ‘Frequency’, not ‘quantity’ is determined by asking how often foods were consumed.

6. Can you comment on how relevant the nutrient composition data from Minnesota are to Quebec?

7. In the methods section you don’t need the bit about the dietitian completing the FFQ with subjects on line 76 as it is in the section on dietary intake anyway.

8. To make this paper more useful for other people wanting to assess n-3 intake
it would be helpful to describe the food items in the FFQ that were main contributors to n-3 intake.

9. Was n-3 from supplements actually calculated and added to the amount from food?

10. Page 8, line 121. It does not make sense to say correlations were used to analyse the type of relationship between diet and plasma fatty acids. Do you mean the strength of the relationship?

11. Discussion, first paragraph. It is important to demonstrate that your FFQ appears to provide information on n-3 intakes that is consistent with the PPL levels; thus supporting the usefulness of the FFQ. It is not important to re-affirm that FFQs in general can estimate n-3 intake.

12. Discussion, para 2. When comparing your results to other studies please give the actual figures for plasma n-3s in those studies. This whole section on comparisons seems a bit difficult to follow. Give a bit more info in the studies in refs 23, 26 and 35, such as who the populations were. Don’t go back to the Oxford population after EPIC as this has already been compared in the first sentence.

13. The bit starting line 152 regarding differences in n-3 metabolism between men and women seems out of place here, unless different gender distributions across the studies being compared could contribute to the observed differences.

14. Page 11, line 157. Not clear that under-reporting relevant here. We already know that FFQ and biomarker n-3s are correlated, sometimes people under-report everything and the distribution of food intake is correct. Biomarkers for n-3 are not quantitative so cannot identify under-reporting.

15. The last sentence on page 11 needs to be reworded. ALA may not be correlated in plasma PL and diet because it can be metabolised, but the sentence could be simplified.

16. You need to be careful about interpreting the correlations between FFQ and biomarkers as evidence that the FFQ adequately estimated DHA and total n-3 intakes. You have no evidence that the absolute amounts estimated are correct, only that you have probably been able to rank people according to their intakes. This is relevant in terms of comparing your estimated intakes with recommended intakes. This is also relevant in relation to your comment in the final sentence about using the FFQ as a screening tool.

17. Page 12, line 195. You have only provided information on n-3 fatty acids, this cannot be used to evaluate FA status.

18. Ref 7 first author should be von Shacky, C.

19. Table 3. (n=100).

Discretionary revisions:

20. Are there any data available on the repeatability of the diet or blood estimates? This can be used to adjust the correlations.

21. Was there a sample size calculation performed? How were subjects
recruited?
22. Page 11, lines 162-165. Can these two sentences be combined? The main message is that sig correlations only for DHA and total n-3, then put correlation coefficients in brackets afterwards.
23. Don’t use ‘men and women subgroups’, either ‘men and women’ or ‘male and female subgroups’.
24. Rather than saying FFQs have been ‘built’ perhaps used ‘developed’.
25. Page 4, lines 55-57. Not sure how doubly labelled water or energy expenditure could help validate n-3 fatty acid intakes.
27. When comparing dietary n-3 intakes of men and women, consider reporting as % energy or per Kj as we know men ate more overall.

**Level of interest:** An article of limited interest

**Quality of written English:** Needs some language corrections before being published

**Statistical review:** No, the manuscript does not need to be seen by a statistician.

**Declaration of competing interests:**

'I declare that I have no competing interests'