Letter to the Editor

Response to Bannenberg and Rice

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This manuscript is a response to concerns expressed in a letter by industry-based scientists Bannenberg and Rice in response to our recent narrative review. In the review, we largely discussed why supplementation with n-3 PUFA rich oils might have benefits to the body composition and metabolism of the offspring of overweight or obese pregnant women. Bannenberg and Rice raised concerns about a number of points that may be perceived as negative about the quality and functionality of commercial fish oils. We provide a refutation to their comments and a brief review of recent evidence regarding the n-3 PUFA content, and oxidative state of supplements available to consumers. From a clinical research perspective, there remains a need to exercise caution. An oil containing less n-3 PUFAs than expected may be ineffective, and lead to incorrect conclusions that n-3 PUFAs lack efficacy. Oxidized fish oil may be ineffective or even cause unwanted harm. Although we must not overinterpret limited evidence from animal models, we have a responsibility to minimize risk to study participants, especially those most vulnerable, such as pregnant women. Prior to selecting a fish oil to be used in a clinical trial, it is essential to independently verify the n-3 PUFA content of the oil, and that the oil is unoxidized.

Response to Bannenberg and Rice Letter to the Editor Dear Editor, In response to the comments of Bannenberg and Rice in their letter to the editor,[1] we have addressed the 4 points they have raised regarding our article, providing additional published evidence that refutes their concerns.[2] In our article, we largely discussed why supplementation with n-3 PUFA–rich oils might have benefits to the body composition and metabolism of the offspring of overweight or obese pregnant women. We briefly raised a number of points about quality and safety, which were not central to our narrative review but are the focus of comments by Bannenberg and Rice, employees of the Global Organization for EPA and DHA (GOED), a promotional industry group.

Bannenberg and Rice[1] challenged the evidence that fish oil supplements frequently contain lower amounts of n-3 PUFAs than labelled. There have been many surveys of n-3 PUFA content in fish oils. Across these studies, the mean percentage of labelled content measured has varied between 68% and 109%, with between 40% and 91% of products having less n-3 PUFA content than labelled, and between 0% and 88% having $< 91\%$ of labelled content.[3–13] Industry-sponsored studies have generally reported a higher rate of products meeting their claimed content[6,7,9] than have independent studies,[3,5,8,10–13] and studies published after 2015[6–10] have reported that products were more likely to meet their claimed content. However, even taking into account only studies published in 2020 (the most recent evi-

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dence available), a store-purchased supplement in the United States or Finland was 40% to 54% likely to have less n-3 PUFA than labelled content and 9.5% to 20% had $< 90\%$[9,10] of the labelled content. Bannenberg, Rice, and others[1,6] have argued that n-3 PUFA concentration should only be measured using methodology validated by industry, describing highly cited methods such as that first reported by Lepage and Roy[14]: as nonstandard or not validated but providing no substantive criticism. However, in a recent industry-sponsored study using multiple laboratories, there was marked interlaboratory variability of up to 20%.[9] This indicates that industry endorsement of a method does not ensure precise measurements.

Our interest is not whether oils reach local regulatory compliance thresholds, as stated by Bannenberg and Rice,[1] rather, it is that a discrepancy of this magnitude is potentially important in situations where achieving high levels of n-3 PUFA intake is necessary to achieve a desired therapeutic target. For example, in the context of fish oil in the pregnancy of women who are overweight or obese, it may be necessary to give a dose of 3 g/day to improve maternal insulin sensitivity (which is the mechanism that we have hypothesized might lead to improved body composition and metabolic outcomes in children).[2] Insufficient content may have affected the results of previous clinical trials in which supplement content was not specifically measured. Therefore, despite the criticism of GOED, it is prudent, as we propose, for researchers to independently validate the dose of n-3 PUFAs in oils that are to be ingested and, subsequently, to analyze circulating concentrations.

Bannenberg and Rice[1] raised concern that a previous study of overweight male participants, published by our group, showing that supplementation with krill oil induced insulin resistance[15] could not be generalized to overweight pregnant women. Because insulin resistance is one of the key metabolic derangements underlying the link between obesity and cardiometabolic risk,[16] use of krill oil by an at-risk group such as overweight men has the potential, in our opinion, to exacerbate cardiometabolic risk. We acknowledge that it is unknown whether this effect would also be present in overweight pregnant women, but as discussed in our narrative review, overweight pregnant women represent a group who are at substantially increased risk of metabolic dysfunction, particularly in the second half of pregnancy.[2] A supplement that increases insulin resistance in pregnancy could potentially precipitate gestational diabetes, which is associated with adverse consequences for mothers and their babies. Thus, with an abundance of caution, we would recommend that although evidence is limited, women who are pregnant and overweight should avoid the use of krill oil. Furthermore, we would emphasize that given the serious nature of any potential risks to the mother or unborn child, studies in this area should proceed only with the utmost care.

Bannenberg and Rice[1] identified an additional concern that we failed to acknowledge recent studies that show "good oxidation status" of fish oil products. There are now many studies examining the oxidative state of consumer fish oil products. Two independent studies published in 2015 showed excess oxidation to be very common in store-purchased supplements, with 50% and 92% exceeding at least 1 of the recommended cutoffs for peroxide value (PV), anisidine value, or total oxidation.[4,17] Since this time, independent studies ($n = 84$ products reviewed) have shown that 17% to 38% of supplements have excess peroxide value, and 0% to 25% have excess anisidine value,[8,10,18] whereas industry-sponsored studies ($n = 107$ products reviewed) have shown 0% to 28% exceeded peroxide value and 4% to 40% exceeded anisidine value.[6,7,9] Across all these studies, the average number of products exceeding a $PV > 5$ was 20%. Interestingly, in an audit of oxidative parameters of oils submitted by industry for analysis (including 1900 measurements), where oils might have been expected to have been less oxidized than store-purchased products, 14% of supplements had a $PV > 5$ and 2.2% a $PV > 10$.[19] It is our contention that there remains variable and, at times, high rates of oxidation in fish oil products sold to consumers, and that this risk is likely to also apply to the oils used in clinical trials. Again, with an abundance of caution, we recommended in our review that researchers independently verify that an oil planned to be used in a supplementation study has a low level of oxidation.

Bannenberg and Rice[1] claimed that, in our review, we implied that "fish oils in general, or fish oils that modestly surpass GOED's limits for oxidative quality, are unsafe." This mischaracterizes our argument and misses an important point. The safety of oxidized fish oil has not been established. Currently, it is unknown what the safe level of oxidation of fish oil is; that is, the level at which the positive effects of n-3 PUFAs are not diminished and there are no ill-health effects. Although some regulatory recommendations are for a peroxide value of < 10, and it is more common for a level of < 5 mEq/kg to be recommended,[20] both of these cutoffs are entirely arbitrary from the perspective of health effects. The appraisal of fish oils by the US Food and Drug Administration as "generally recognized as safe" does not undermine our argument, because there is currently insufficient evidence to determine whether there is a level of oxidation for fish oils that should be considered safe. More work is required to determine the safe level of oxidation of n-3 PUFA–rich oils. In the

meantime, we make the conservative recommendation that studies of n-3 PUFA–rich oil supplements report the level of oxidation of their supplement, and after independent testing, select a supplement with a peroxide value < 5 mEq/kg, which is the cutoff for a quality supplement recommended by GOED and others.[20]

Bannenberg and Rice[1] stated we have implied that oxidized fish oils would have deleterious effects in human pregnancy, based on our previous study of supplementation of oxidized fish oil to pregnant rats. This is incorrect, although we do believe it is prudent for pregnant women to avoid consuming oxidized fish oil and for clinical trials using a fish oil supplement to independently prove it has low levels of oxidation before it is used. We showed in a rat model that a very high dose of highly oxidized oil induced maternal insulin resistance and high neonatal mortality.[21] This is proof of concept that oxidized fish oils can be harmful in pregnancy, but the finding cannot be generalized to women. The effects of oxidized fish oil in human pregnancy remain unknown, and it is also unknown whether the lower relative doses consumed by women are potentially harmful. In this context, it must be recognized (1) that oxidized fish oil should not be assumed to be safe in human pregnancy, and (2) a human study of oxidized fish oil in pregnancy would not be ethically appropriate. A detailed dose-response toxicity study in rat pregnancy is required and underway.

To conclude, Bannenberg and Rice[1] have taken exception to statements that may be perceived as negative about the quality and functionality of commercial fish oils. These statements are supported by careful review of the evidence. From the perspective of clinical research, there is a need to exercise caution. An oil containing less n-3 PUFAs than expected may be ineffective and mislead researchers to erroneously conclude a lack of efficacy. Oxidized fish oil may be ineffective or even cause unexpected harm. Although we must not overinterpret limited evidence from animal models, we have a responsibility to minimize risk to study participants, especially those most vulnerable, such as pregnant women.

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