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# ***Interactive comment on “An isotopic ( $\Delta^{14}\text{C}$ , $\delta^{13}\text{C}$ , and $\delta^{15}\text{N}$ ) investigation of particulate organic matter and zooplankton biomass in Lake Superior and across a size-gradient of aquatic systems” by P. K. Zigah et al.***

**P. K. Zigah et al.**

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## Responses to comments by reviewer no. 1

Comment: The manuscript by Zigah et al. (Biogeosciences Discuss. 9, 4399, 2012) exploratively investigates the elemental and isotopic composition of ‘zooplankton’ (everything caught by 300  $\mu\text{m}$  mesh size nets) and bulk particulate organic carbon (POC) in Lake Superior. By using a 1-isotope ( $\Delta^{14}\text{C}$  only) Bayesian mixing model, the authors conclude that algal carbon consistently contributed to a significant share of the

Full Screen / Esc

Printer-friendly Version

Interactive Discussion

Discussion Paper



zooplankton biomass, while terrestrial POC, sediment material or bacterial carbon did not. Allochthony calculations showed that the overall terrestrial contribution to zooplankton was occasionally high, with values up to 54%, and that the grand average of zooplankton allochthony was 10-20% depending on choice of model parameters (Table 5). Further, the Lake Superior data was compared with data from other lakes, showing that the discrepancy between zooplankton  $\Delta^{14}\text{C}$  and algal  $\Delta^{14}\text{C}$  increased with increasing basin:lake area and decreasing hydrologic residence time. The ms convincingly shows how  $\Delta^{14}\text{C}$  can be used to determine allochthony and to some extent discriminate between algal and other sources of zooplankton biomass. The  $\Delta^{14}\text{C}$  datasets that are presented are in many ways unique, and allow for novel conclusions about zooplankton reliance on different food sources in Lake Superior. Thus, this study makes a significant contribution to the field of biogeoscience.

Response: We thank the reviewer for his/her valuable comments on our MS.

Comment 1. My major concern is that a single isotope ratio ( $\Delta^{14}\text{C}$ ) is used to model the contribution to zooplankton by as many as four different sources. Recent Bayesian mixing models such as MixSIR and SIAR have tremendously increased the possibility to discriminate between multiple sources. However, the whole idea of working with more than two sources is based on the simultaneous use of multiple isotope ratios. If there is only one isotope ratio, but multiple sources, the two sources that represent the highest and the lowest 'endmember', respectively, can be mixed in different proportions, creating combinations that perfectly resemble all other sources. My recommendation would be to include more isotopes in the MixSIR model (at least  $\delta^{13}\text{C}$ ), remove sources from the MixSIR model or to abandon the MixSIR model.

Response: We have included  $\delta^{13}\text{C}$  in the MixSIR model and have rerun the model using both  $\Delta^{14}\text{C}$  and  $\delta^{13}\text{C}$  signatures as suggested. See pg 10, Section 2.4.

Comment 2. Radiocarbon data from a set of lakes in Québec (Canada) is presented as original data. However, as far as I can see, these data were already published

BGD

9, C3182–C3192, 2012

Interactive  
Comment

Full Screen / Esc

Printer-friendly Version

Interactive Discussion

Discussion Paper



Interactive  
Comment

by McCallister & del Giorgio (2008, Limnol. Oceanogr. 53:1204). The sampling in these lakes was also following a different protocol, e.g. using plankton nets with a much smaller mesh size (50  $\mu\text{m}$ ). My primary suggestion would be to bring in this data from the Québec lakes as published data, using the McCallister & del Giorgio (2008) reference. Alternatively, the authors need to motivate the choice of considering this data material as original. However, if the 'McCallister & del Giorgio' data can be considered original also here, I would seem logical that P. A. del Giorgio is at least acknowledged (or perhaps even included as co-author) in the present manuscript.

**Response:** We decided to present and discuss the individual  $\Delta^{14}\text{C}$  data from the small lakes in Quebec in this MS because McCallister and del Giorgio (2008) paper did not look at the individual lakes, rather only reported the ranges for all the lakes that they studied. Del Giogior was aware of and saw this MS before submission, and we meant to acknowledge him in the submitted manuscript. As suggested by the reviewer, we now bring in these data as published data. See Pg 8, lines 171-173, and Table 6.

**Comment 3.** In the methods description, lines 13-17, p. 4404, a previous study is cited, indicating that 90% of the zooplankton in Lake Superior is likely to be consisted by copepods. The manuscript needs to be much more specific here, by considering different groups of copepods that have fundamentally different feeding styles. It could be argued that the feeding styles within the copepod community (e.g., cyclopoids vs calanoids) in fact show bigger differences than those shown between cladocerans and copepods. Therefore, a more detailed description of the Lake Superior zooplankton community would be preferable. Further, the discussion could be strengthened by addressing the possible role of shifting zooplankton communities for the observed patterns in allochthony. For example, it could be argued that the allochthony was higher in the small lakes because these happened to have a high proportion of relatively non-specific filter feeders such as Daphnids.

**Response:** We have included a more detailed description of the zooplankton community in Lake Superior in the MS. See pg 6 & 7, lines 136-149. We have also discussed

[Full Screen / Esc](#)[Printer-friendly Version](#)[Interactive Discussion](#)[Discussion Paper](#)

the role of shifting zooplankton communities in the MS. See pg 25 & 26 lines 576-592.

Specific comments Comment 1. p.4400 l.5-7 The abstract reads: 'these subsidies [i.e., terrestrial carbon subsidies]: : play major roles in determining the fate of organic carbon'. I am not sure what this means. Is the terrestrial carbon determining the fate of itself?

Response: The revised abstract clarifies this. Please see pg 2. Lines 29-30.

Comment 2. p.4400 l.7 Perhaps a reference that supports the claim that terrestrial carbon subsidies may support economically important fisheries should be included somewhere in the manuscript

Response: The revised abstract no longer contain this assertion. Please see Pg 2, lines 29-30.

Comment 3. p.4402 l.10-11 'neither : : : or' should be changed to 'neither : : : nor' or 'either : : : or'

Response: We have replaced 'or' with 'nor'. See pg 4 line 83.

Comment 4. p.4403 l.15 'of' is missing between 'abundance' and 'radiocarbon'

Response: We have inserted 'is'. See Pg 5 line 114.

Comment 5. p.4404 l.6-9 The choice of zooplankton sampling protocol might need to be motivated. For example, a 300  $\mu\text{m}$  net will not retain protozoan zooplankton, small meta-zoan zooplankton such as rotifers or even small crustacean individuals. Further, since the zooplankton were not hand-picked, it can be argued that coarse POC might have contaminated the samples. These issues are presently not addressed.

Response: To make it clear that the zooplankton that we studied were of a particular size group, we now specify mesozooplankton throughout the MS as the zooplankton size group that were studied in Lake Superior. The zooplankton samples were rinsed to rid them of smaller particles and then collected onto GF/F filters. Visual inspection

**BGD**

9, C3182–C3192, 2012

Interactive  
Comment

Full Screen / Esc

Printer-friendly Version

Interactive Discussion

Discussion Paper



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Interactive  
Comment

of the filters indicates that zooplankton biomass was the major contributer to these samples, although a small amount of other carbon may indeed be present. Please note that the  $\Delta^{14}\text{C}$  values of mesozooplankton does not show (or support) measurable contamination by POC because 14C ages of POC samples are significantly older than those of the mesozooplankton, which actually track DIC radiocarbon signatures.

Comment 6. p.4404 l.13-17 It is not clear what the 90% refers to. Is it out of total zooplankton, metazooplankton or only crustaceans? Adults or including nauplia and juvenile cladocerans? Is it 90% by numbers or by biomass?

Response: This has been clarified. It is  $\sim$ 90% (by biomass) of the crustacean zooplankton. Please see Pg 6, line 137.

Comment 7. p.4410 l22 MixSIR is using the standard deviations of source isotope signatures to obtain reasonable estimates of uncertainties in the contribution by different sources to a consumer. Section 3.2.1 focuses on the endmembers themselves, but not explicitly on the standard deviations of these endmembers. It needs to be clarified how the standard deviations were managed.

Response: As stated in section 2.4 pg 10, line 221-223, the uncertainty of  $\delta^{13}\text{C}$  and  $\Delta^{14}\text{C}$  values that were used in the model are the analytical uncertainties based on analyses of multiple external standards, or multiple analyses on a graphite target in the case of the  $\Delta^{14}\text{C}$  values.

Comment 8. p.4412 l.16 suggestion is to remove 'a little'

Response: We have removed 'little'. See pg 13, line 281.

Comment 9. p.4413 l.14-17 It is not clear how the 1-isotope modeling approach gives any advantage in terms of being able to use other isotope ratios as independent checks on the robustness of the model output. First, a multi-source 1-isotope model can never be robust (see general concern #1). Second, the authors do not consider that these other isotopes ( $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ ) contain important information about sources. Thus, it

Full Screen / Esc

Printer-friendly Version

Interactive Discussion

Discussion Paper



seems a contradiction that they can be used to check model robustness.

Response: We have revised the paragraph to clarify the apparent contradiction. See pg 16, lines 357-358.

Comment 10. p.4414 l.5 remove 'then'

Response: Done. See pg 16, line 371.

Comment 11. p.4414 l.22 a suggestion could be to replace 'vs.' with 'or'

Response: Done. See Pg 17, line 381.

Comment 12. p.4415 l.27 This is an intriguing result that that is unexpected considering the close correlation between the  $\delta^{13}\text{C}$  of zooplankton and POM that have been reported, e.g. by Mohamed & Taylor (2009). It raises a few questions that could be addressed in the discussion: Is the studied ecosystem different than those previously studied? Is it rather the choice of using  $\Delta^{14}\text{C}$  that helps reveal that there in fact is no connection between the sources of POM and the sources of zooplankton biomass? Could it be that the zooplankton community in Lake Superior is dominated by very specific grazers that use only certain components of the bulk POC pool?

Response: We have included a discussion of the zooplankton community composition vs. allochthony in the MS. Please See pg 25 & 26, lines 577-593.

For Lake Superior, we observed, as stated in section 4.3 pg 20 & 21 lines 465-470, that the addition of radiocarbon data considerably refined the relationship between mesozooplankton and POM, and reveal that sources of mesozooplankton biomass carbon much more clearly than the stable carbon isotope data shows. Because of the much larger dynamic range of  $\Delta^{14}\text{C}$  (-1000 to  $\sim+200\text{\textperthousand}$ , compared to that of  $\delta^{13}\text{C}$  in organic carbon (-32 to -12%, as stated in pg 4 lines 94-95,  $\Delta^{14}\text{C}$  values are much better at source differentiation. Although Lake Superior ecosystem consist of a large fraction of specific filter feeding calanoid copepods, the stable carbon isotope data alone did not sufficiently reveal the dominant food source to the mesozooplankton biomass, es-

**BGD**

9, C3182–C3192, 2012

Interactive  
Comment

Full Screen / Esc

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Interactive Discussion

Discussion Paper



Interactive  
Comment

pecially during isothermal condition where the  $\delta^{13}\text{C}$  of the bulk POM was identical to  $\delta^{13}\text{C}$  of mesozooplankton, suggesting incorrectly that the mesozooplankton were indiscriminately feeding on everything (algae, detritus, protists, bacterial, terrestrial OC) in the bulk POM, or that there is a dramatic shift in the zooplankton community composition to non-specific filter feeders. It is the sensitivity of the  $\Delta^{14}\text{C}$  data to source variation that allowed the consistent filter feeding habit of the dominant calanoid zooplankton, as well as the use of algae or algae-feed microzooplankton by the other zooplankton groups in the lake to be observed/detected. It should be noted that even in the smaller lakes that we studied,  $\delta^{13}\text{C}$  values of zooplankton and POM considerably overlapped suggesting non-selective feeding style and or the dominance of non-specific filter feeders feeding on indiscriminately on the bulk POM (Fig 1 in McCallister and del Giorgio, 2008). However, the inclusion of radiocarbon data clearly revealed that even in the smaller lakes, algae was preferentially fed upon by the zooplankton (Fig 3 in McCallister and del Giorgio, 2008; this study), and that the potential predominance of non-specific filter feeders such as daphnids and cyclopoid copepods in the smaller lakes did not lead to a switch from algal food preference. It will be interesting to see what insight would come from wider use of radiocarbon in ecosystem ecology studies; we believe such data would likely refine the current understanding of the functioning of a lot of the other studied systems that were based solely on stable carbon isotopes.

Comment 13. p.4416 l.1-2 Here and elsewhere: The results section includes interpretations of what the results suggest etc., which makes it discussion-oriented. Other parts of the results are clearly methods-oriented, like section 3.2.1. As a consequence, the results section span over more text pages than it would need to do.

Response: P4416. Line 1-2 has been revised to avoid interpretation. See pg. 18, lines 415-416. We have moved the section 3.2.1 to methods as suggested. See section 2.4.1 We have also removed all apparent interpretations in the results sections.

Comment 14. p.4432-4433 Tables 3-4 An alternative to the ‘median contribution’ is to use the modal contribution. Often the probability distributions that are generated by

[Full Screen / Esc](#)[Printer-friendly Version](#)[Interactive Discussion](#)[Discussion Paper](#)

Interactive  
Comment

Bayesian models are complex and the median may not represent the contribution that is most likely. Thus, the mode has been suggested to be a better measure of central tendency (Parnell et al. 2010 PLoS One 5:e9672). On the other hand, the sums of the different contributions are relatively close to 1 in Tables 3-4, indicating that the median is working fairly well in this case.

Response: We have checked again, and the median values in Tables 3-4 were all very close to 1, so we agree with the reviewer that the median values are working pretty well in this case, and have chosen to report the median values from the model.

Comment 15. p.4432-4433 Tables 3-4 There is conceptual overlap between the different sources. If the bacteria have a high degree of allochthony, the bacteria are, in principle, part of the terrestrial POC pool. Further, also the sedimentary OC and Bacterial carbon/DOC can be of algal origin.

Response: We agree that all the carbon in the lake (and all aquatic systems) ultimately comes from terrestrial and/or in situ aquatic algal source. However, there are several subunits of the POC pool that could be distinguished beyond (on a much finer level than) the level of terrestrial vs. algal distinction. This has not been, or could not be done in most studies because those studies only used stable carbon (and nitrogen) isotopes; in such studies it is difficult (if not impossible) to distinguish POC sources beyond terrestrial vs. algal. However, radiocarbon signatures, as employed in this study, do allow for such finer distinctions to be made. Hence, given the unique radiocarbon signatures of the various carbon pools in Lake Superior, we are able to determine the terrestrial vs algal contributions, and in addition, to tease apart what fraction of the POC comes from the OC from the sediment, terrestrial, recent algal as well as bacterial sources. We have explained the need for such further and more detailed source partitioning in Lake Superior in Section 2.4.1 (previously 3.2.1).

Comment 16. p.4433-4434 Tables 4-5 There is typically a 20 units of percentage off-set (in either direction) between zooplankton autochthony (Table 5) and algal carbon con-

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tribution to zooplankton biomass (Table 4). This big difference needs to be adequately discussed.

BGD

9, C3182–C3192, 2012

Interactive  
Comment

Response: The revised Bayesian model estimates based on dual isotopes ( $\Delta^{14}\text{C}$  and  $\delta^{13}\text{C}$ ) no longer show such a huge offset. Please See the revised Table 4. The lakewide average offset is now  $\sim 8\%$  or 3% if the Isothermal SM site with large difference is excluded. The offset is only  $\leq 1\%$  when the creek POC is used as the terrestrial endmember. However, for the mesozooplankton allochthony estimate, the lakewide average offset was  $\sim 18\%$  or 11% (if Isothermal SM is excluded). The offset, however, is  $\leq 6\%$  when creek POC is used as terrestrial endmember. We have now included this bit in the results text. See pg 17, lines 383-390.

Comment 17. p.4438 Fig 3b I would remove the regression lines as no regression model exists here (the regression is almost as far from significant as it can be).

Response: We have removed the regression line as suggested. See the revised Fig 4b (previously 3b).

Comment 18. p.4439 Fig. 4b Perhaps this correlation should be denoted 'marginally significant'

Response: We have noted in the result text that the correlation is marginally significant. See pg 18, lines 418-420.

We have also stated this in the caption of Fig 5b (previously 4b). Please See pg 45, lines 913-916.

Comment 19. One recently published paper of seemingly relevance is not cited: Karlsson et al. (2012) Limnol. Oceanogr. 57(4): 1042-1048, DOI: 10.4319/lo.2012.57.4.1042 Just like the present manuscript, this study shows that autochthonous organic matter contributes 'disproportionally' to zooplankton biomass

Response: We agree this is a relevant paper and it has now been cited in this MS. See pg 3, line 56; pg 26 line 605.

Full Screen / Esc

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Interactive Discussion

Discussion Paper



Please also note the supplement to this comment:

<http://www.biogeosciences-discuss.net/9/C3182/2012/bgd-9-C3182-2012-supplement.pdf>

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Interactive comment on Biogeosciences Discuss., 9, 4399, 2012.

**BGD**

9, C3182–C3192, 2012

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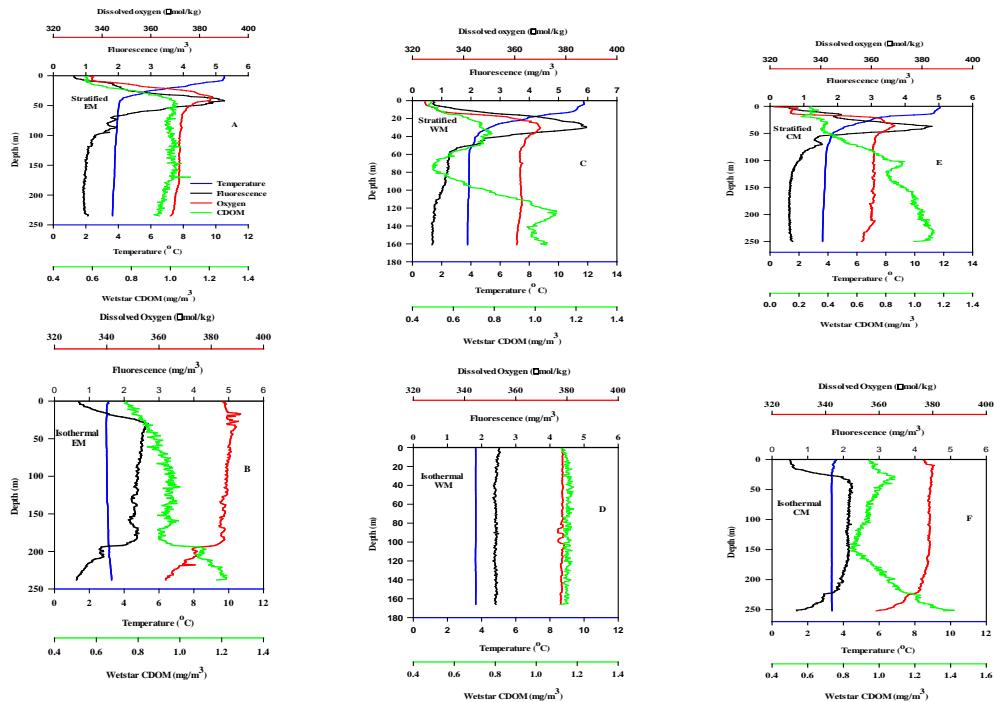
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Interactive Discussion

Discussion Paper



Interactive  
Comment

**Fig. 1.** Supplementary Figure 1

Full Screen / Esc

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Interactive Discussion

Discussion Paper

