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# ***Interactive comment on “Experimental assessment of environmental influences on the stable isotopic composition of *Daphnia pulicaria* and their ephippia” by J. Schilder et al.***

**Anonymous Referee #1**

Received and published: 9 February 2015

General comments.

As stated at the end of the introduction, this paper was intended ‘to experimentally examine whether offsets in  $\delta$  13C,  $\delta$  15N and  $\delta$ 18O values exist between Daphnia and their ephippia’. If this a question of interest for those who are working on past changes in Daphnia isotope composition using ephippia recovered from sediment archives. This is however a very small community and I do not think that this paper will touch a large readership. It does not really connect either to biogeosciences, and, as a matter of fact, there is very few references to any biological or geochemical processes within the whole paper (although it could be relevant to specify the working hypotheses in the introduction, i.e. according to which physiological hypotheses ephippia should exhibit

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isotope composition that would be differ from those of the whole carapaces/body).

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Besides, the experiment that has been performed is of very limited scale, at a point that unexpected results (such as those obtained for  $\delta$  13C at 20°C or very different Daphnia  $\delta$  15N in spite for similar  $\delta$  15N of the food sources) remain hard to explain. Because the range of tested conditions is narrow, the study does not provide any novelty as compared to previous papers on that topic, exception maybe for  $\delta$  18O.

Actually, the very annoying point of this paper is that everything is done to inflate and oversell the real content of the paper and the reviewer feels he is getting duped. The title is somewhat catchy, but 'environmental influences' actually refers to (i) test of two  $\delta$  13C food values, which differ by less than 1.8 per mil, (ii) two temperature conditions, one of which leading to conditions that 'may not affect Daphnia in their natural environment' and (ii) two  $\delta$  18O water values. Even if the experimental setting was ideal, it would have been only two conditions for each factor, and this would not be enough to be called 'environmental conditions'. It is even more dubious that the experimental design was not perfect. If the point was to test whether food  $\delta$  13C affect the isotopic offset, we would expect that a much larger range in  $\delta$  13C values for the food sources. My guess is that much more labelled sodium bicarbonate would have required in the algal growth medium to create such a range of  $\delta$  13C but this is understandable flaw because this can be usually difficult to anticipate. The experiment had been already conducted by the time that authors realized that labelling was too small to really serve the working hypothesis. In a sense, it is interesting to see that even such a small range of  $\delta$  13C values is detectable at the level of ephippia isotope composition, but this is not the way this is presented in the paper.

To remediate to the narrowness of the potential readership, authors try to increase the perspective of the experiment by relating to the need for the community of isotope ecologists to quantify trophic fractionation factors (p2577, from I 20). Yet, because the experiment has not been initially designed for such purposes, it does not provide any more information than those that have been specifically conducted some time ago

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(Impact of temperature by Power, 2003 ; food composition by Matthews and mazumder, 2008).

To conclude, authors have targetted a high-level, generalist journal but this experiment, even if everything had worked perfectly, does not have the potential to reach such a readership. Inflating artificially the purpose of the paper is not enough to fool the reader on the actual contribution of the research (may be just enough to upset him/her). This study has been designed for a very specific purpose, and therefore should be published in a very specialized journal. The experiment itself has been performed rigourously, and even though it has a small scale and produced sometimes unexplained results, I am very confident it could be published in the adequate journal (JOPL ?).

Specific comments.

Overall, the language is very understandable and the paper is clear. However, I found that the graphical representations of the results (fig 2 & 3) were not legible, and hampered the understanding. Fig 1 is not necessary, the text is clear enough. Fig. 5 also can be removed, as it presents very straightforward results.

Table 1: Significance detection in multiple paired comparisons requires accounting for Bonferroni's corrections.

Three different clones were used and they apparently did not contribute equally to ephippia production. Any clone effect on the isotope results?

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