

Interactive comment on “Short and long term consequences of larval stage exposure to constantly and ephemerally elevated carbon dioxide for marine bivalve populations” by C. J. Gobler and S. C. Talmage

Anonymous Referee #3

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General comments

The manuscript by Gobler and Talmage explores the effects of different regimes of exposure to elevated carbon dioxide on the larval stages of the bivalves *Mercenaria mercenaria* and *Argopecten irradians*, as well as the carry-over effects to the juvenile stages. The study provides new and relevant experimental evidence on larval survival, calcification rates, RNA:DNA ratios and growth rates. The hypothesis of trans-life-stage ‘legacy effects’ or “carry-over effects” of exposure to elevated CO₂, although not novel, seems to be supported by the dataset presented. The questions addressed

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are all relevant and well within the scope of the journal. The study is well structured, clearly presented and advances significant contributions to the field, so I recommend its publication in Biogeosciences. There are however, some minor aspects needing revision or clarification, which are detailed below.

Specific comments

Methods:

p.15906, L.12: The origin of the larvae used in the experiments is not given. Describe briefly how they were obtained. p.15906, L.15: What was the density of T-iso cells provided? p.15906, L.22: It is not clear what the expression “1% of its original concentration” refers to. Please clarify

p.15906, L.25-28: Given that no difference in larval performance was observed in experiments done with and without antibiotics I do not understand the rationale for using antibiotics.

p. 15908, L.7-8: The assumption about the relative importance of shell dissolution and deposition seems questionable. In fact, in previous studies by these authors (Talmage and Gobler, 2009, 2010), larval bivalve shells exposed to elevated concentrations of CO₂ have been considered highly vulnerable to dissolution. Please clarify this issue in the discussion.

p. 15909, L.25-27: Were 40 individuals used per treatment or in total? According to Figure 5, four replicate beakers were used before and after the transfer across treatments. Please clarify. p. 15910: How many experimental units and how many individuals per unit were used for each treatment level in the long term growth experiment. p. 15910, L23-24: The purpose of the ranking procedure prior to data analysis is not clear. Were non-parametric statistics used?

Results

p. 15914, L6-7: The stated objective here is was to measure post- settlement growth,

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but results are given for two post-spawning periods. This is confusing because, if I understood correctly, the first measurement for the 12 week period was done while the individuals were exposed to the different CO₂ treatments and the second measurement was done after the transplants to the field. In contrast, both measurements during weeks 13-26 were already done in the field.

p. 15914, L14-15: According to Figure 7, in September the shell diameter at 390 μ atm was approximately 16 mm. Individuals reared at 390 and 750 μ atm were probably already similar in size by January.

Discussion

p. 15915, L2-4: I think some clarification on the experimental design of the long term growth experiment is needed to support the claim about the carry-over effects (see comments above on the methods section). p. 15916, L7-9: Given the methodological assumption that shell dissolution is negligible relative to shell dissolution, the results observed here should be largely attributed to reduced accretion of new shell. Please clarify.

p. 15919, L3-4: The conclusion about the faster growth of juvenile individuals exposed to high CO₂ as larvae is confounded by the fact that, although the individuals were transplanted to the field after 47 d, growth was measured from 0-12 weeks. See comment above (p. 15914, L6-7). p. 15919, L19: On carry-over effects, see also:

Parker LM, Ross PM, O'Connor WA, Borysko L, Raftos DA, Pörtner H (2012) Adult exposure influences offspring response to ocean acidification in oysters. *Global Change Biology* 18:82–92

Dupont S, Dorey N, Stumpp M, Melzner F, Thorndyke M (2012) Long-term and trans-life-cycle effects of exposure to ocean acidification in the green sea urchin *Strongylocentrotus droebachiensis*. *Marine Biology*: 1–9. DOI 10.1007/s00227-012-1921-x

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