Comment on bg-2021-34
Anonymous Referee #1

Referee comment on "Anthropogenic CO$_2$-mediated freshwater acidification limits survival, calcification, metabolism, and behaviour in stress-tolerant freshwater crustaceans" by Alex R. Quijada-Rodriguez et al., Biogeosciences Discuss., https://doi.org/10.5194/bg-2021-34-RC1, 2021

Overall, this manuscript provides timely and necessary data for understanding how CO$_2$-induced acidification may affect freshwater organisms. While the manuscript is well written I do have a number of concerns and suggestions that if addressed will improve the quality of the final paper. A large part of my concern is that the authors are throwing away a chance at putting this study into a larger ecological context. Researchers study the effects of climate change on organisms to determine their adaptibility but very little is mentioned about this in the manuscript.

1. **Length of experiment**- In the methods section, the authors need to state the duration of their experiments. I noticed on the results table that for some measurements, the time axis extended to 8 weeks while in others 7 days, so I'm assuming that different experiments were done for different lengths of time? If so this needs to be explicitly stated somewhere in the methods.

2. **Line 37**: "To date there are no comprehensive studies investigating various physiological and behavioral effects of realistic future levels of CO$_2$-mediated acidification in calcifying freshwater invertebrates". This is false. David et al. (2020) investigated the effects of CO$_2$-induced acidification on the invasive freshwater gastropod, *Viviparus georgianus* over a 12 week period (Journal of Molluscan Studies 86:259-262). They used shell repair as a proxy for physiological performance. The findings of that study was the first to assess the effects of CO$_2$-induced acidification in a calcifying freshwater invertebrate and regardless of whether the authors think it was 'comprehensive' or not, that reference should still be somewhere in the manuscript and the results compared with theirs.

3. What was the rationale for using the Chinese mitten crab for this study? No information is provided on the study species in terms of life history, fecundity, etc? The authors mentioned that it is a model organism for studying climate change but do not actually explain why.

4. Very little is mentioned on the invasive status of Chinese mitten crabs in other parts of the world and nothing is mentioned about the implications of the findings of this study on the management/control of invasive populations of the species. This is what I meant when
I mentioned earlier that the authors did not put their findings into a broader context.

5. **Line 65:** The authors mentioned that these crabs were 'purchased'; does that mean that they were bred in a controlled setting prior to arrival? If so, how sure are you that their physiological responses are reflective of what happens in natural populations?

6. **Line 71:** Is oatmeal and mollusc meat the type of food the crabs would usually eat in their natural environment? Is that a best practice for doing controlled experiments on crabs in the lab? (If so please provide reference).

7. If you collected juveniles and cultured them under these stressors, why not measure the growth rate? In fact, why wasn't size measurements taken prior to each experiment?

8. What was the sample size for each experiment. This needs to be included either in the methods or when reporting statistics in the results section.