Interactive comment on “Assessing annual variability in the shell thickness of the pteropod Heliconoides inflatus in the Cariaco Basin using micro-CT scanning” by Rosie L. Oakes and Jocelyn A. Sessa

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Reviewer comments have been copied below and author responses are listed underneath them. Page and line numbers refer to the tracked changes version of the manuscript.

Reviewer # 1 - General Comments: The aim of this study is to describe annual variability of different shell growth parameters (thickness, diameter, number of whorls, amount of shell material) of the pteropod Heliconoides inflatus in the Cariaco Basin (Venezue-
Ian Shelf). Additionally, shell condition was analyzed applying the Limacina Dissolution Index (LDX). Pteropod samples were collected over a year period from a sediment trap and compared to prevailing carbonate chemistry and nutrient conditions with the goal to entangle driving abiotic or respectively biotic factors of the various measures. The authors found that food availability has a greater control on shell formation than aragonite saturation and that shell condition was not altered with time spent in the sediment trap cup. Hence, the results can serve as baseline data to better quantify the response of this highly vulnerable organism group to ocean acidification (OA) by disentangling abiotic from biotic factors that impact on shell formation.

I think this study is very interesting and addressing a very important question in relation with consequences of OA on highly vulnerable thecosome pteropods. It gives strong in situ evidence that food availability and energy constraints have a major potential to mitigate abiotic stress and shows nicely that various shell parameters indicative for growth and calcification did not depend on the saturation state of aragonite, at least not in the range observed (always above 2).

From my understanding, the purpose of the study was twofold: 1) How does length of time (preservative) in the trap impact shell condition and potentially lead to false conclusions in the OA context? 2) Do changes in water column properties affect shells and how or which? Hence, point 1 looks at dead organisms, point 2 affects live organisms in the water column (including the carbonate chemistry history pteropods experienced in the past). In this context, my main criticism is that the author did not distinguish between processes that happened when pteropods were still alive (in the water column) and already dead (in the water column and the sediment) particularly with respect to potential shell degradation they observed on the preserved samples. Did the authors simply assume that shell integrity was intact as long as organisms dwelled in the water column alive? Might indeed be reasonable to assume but the authors need to state clearly in their ms what their opinion on that is and whether/when they talk about live or dead organism. Furthermore, one important issue with sediment trap samples is that
pteropods might have entered them as “swimmers” not as dead individuals that simply sank into the trap. This problem should be mentioned in the introduction and picked up later in the discussion again, would that impact the conclusions to draw from the Results?

Author response: We agree with the general comment about the need to discuss processes affecting pteropod shells when the organism is live, dead, and in the preservative. We have added information to the abstract and introduction, and have reformatted our discussion to better outline these processes and the likelihood of shell alteration occurring at each of these stages. Abstract: Pg. 2, lines 10 – 13, Introduction: Pg. 4, line 33 – Pg. 5, line 11, Discussion: Pg. 12, lines 5 – 24). We have also discussed the potential impact of swimmers on the samples in the introduction (Pg. 5, lines 1 – 4) and discussion (Pg. 12, lines 24 – 30) sections.

Reviewer #1 - Specific Comments

Reviewer comment: The title does not reflect the study content well enough. For example micro-CT is not even mentioned in the abstract and shell thickness is only one out of a set of measured parameters mentioned in the abstract. LDX is much more prominent in the abstract instead. Also, I think the title should reflect that the ms is about sediment trap samples of H. inflatus. Please change title accordingly to maybe something like this: “Assessing abiotic and biotic impact on annual variability of shell condition of the pteropod Heliconoides inflatus in the Cariaco Basin: shell dissolution index, size and thickness as revealed from sediment trap samples.”

Author response: Thank you for pointing that out. We have changed our title to, “Determining how biotic and abiotic variables affect the shell condition and parameters of Heliconoides inflatus pteropods in the Cariaco Basin”, based on your recommendation.

Reviewer comment: L4–6: This study does not deal with natural variability of pteropods (in terms of abundance of which “variability” is usually understood if not stated otherwise), neither is it discussed. Either remove this sentence or rephrase to harmonize
with the variability you are actually focusing on (shell growths parameters).

Author response: We agree that the use of ‘variability’ could have been interpreted in multiple ways throughout the text. We have changed the text in the abstract to clarify our focus on shell growth parameters. It now reads, “… the biotic and abiotic factors influencing their shell formation and dissolution in the modern ocean need to be quantified and understood.” (Pg. 2, lines 5 – 7). We have either changed the wording, or expanded on the meaning of our use of the word ‘variability’ through the manuscript.

Reviewer comment: L11: remove “with”

Author response: done

Reviewer comment: L14/15: Are the authors talking about dead or live individuals?

Author response: We agree that the differentiation between processes that affected live and dead individuals was unclear. We have restructured the abstract to clarify the mechanisms that can alter shell condition when the animals are live, dead, and in the preservative. “The shell condition of pteropods from sediment traps have the potential to be altered at three stages: 1) when the organisms are live in the water column associated with ocean acidification, 2) when organisms are dead in the water column associated with biotic decay of organic matter, 3) and when organisms are in the sediment trap cup associated with the abiotic alteration by the preservation solution.” (Pg. 2, lines 10 – 13).

L19: : : : in shell characteristics of H. inflatus of trapped pteropods: : :

Author response: We acknowledge the importance of conveying the fact these samples are from a sediment trap in the abstract. We have added a section on mechanisms affecting shell condition in specimens from sediment traps (Pg. 2, lines 10 – 13) and have added additional mentions in the abstract.

Reviewer comment: Section 1.2: The authors should shortly mention the problem of collecting live pteropods (“swimmers”) in sediment traps and how that could have
affected their work approach and results. (Alternatively it might be mentioned on P4 last paragraph).

Author response: Good point - we have added discussion of swimmers entering sediment trap samples in the introduction (Pg. 5, lines 1 – 4), and have mentioned how our results may have been impacted by swimmers in the discussion (Pg. 12, lines 24 – 30). Introduction: “A further complication of sediment trap data is that interpretation can be skewed by the presence of ‘swimmers’, i.e., specimens that were alive when they entered the trap (Harbison and Gilmer, 1986). This is a particular concern with pteropods as they sink to avoid predation (Harbison and Gilmer, 1986) and therefore may enter into the trap while still alive.” Discussion: “These results could have been further complicated by the presence of swimmers, which would have entered the trap live and therefore would not have undergone any dissolution in the water column. If there was an increase in swimmers entering the traps at one time of year relative to another, it could be interpreted as less water column breakdown during these months. The most pristine shells in this study entered the trap in June and December, suggesting that there was not a seasonal pattern to swimmer frequency. We therefore assume that the number of swimmers entering the sediment trap is constant throughout the year and therefore does not affect the seasonal trends reported above.”

Reviewer comment: Throughout the ms, they need to make clear whether they talk about live or dead organisms.

Author response: We agree that the distinction between processes that affect live and dead shells was not clear in the original manuscript. We have worked to clarify how different processes affect live, dead, and preserved shells: Abstract: Pg. 2, lines 10 – 13, Introduction: Pg. 4, line 33 – Pg. 5, line 11, Discussion: Pg. 12, lines 5 – 24.

Reviewer comment: P4L6: Lischka and Riebesell 2017 (Polar Biol, Volume 40) also studied metabolic response of pteropods (oxygen consumption).

Author response: Thank you for reminding us of this work - we have added the refer-
ence (Pg. 4, lines 11 – 12)

Reviewer comment: P4L24: through misses the “r”

Author response: corrected

Reviewer comment: P5L33: remove comma between body and whorl

Author response: In molluscs, the final whorl is also known as the body whorl. The sentence is grammatically correct as it was, “…the final, or body, whorl.” (Pg. 6, line 17)

Reviewer comment: P6L10: Please detail at what temperature and for how long shells were dried.

Author response: We have added details about drying to the methods section (Pg. 6, lines 28 – 29): “Calcareous plankton were wet-picked, and left to dry in a 40°C oven for 24 hours, before being, dried, and stored for faunal analysis (E. Tappa pers.comm.).”

Reviewer comment: P10L31/32: Could any changes detected originate in the time prior collection in the trap during live in the water column?

Author response: This is an interesting question. We assume that there was no in-life dissolution as the water is permanently supersaturated with respect to aragonite, and because we don’t see any evidence of patchy dissolution, such as is seen when pteropods undergo dissolution in-life (e.g. Peck et al., 2016, 2018). We have clarified this in the text (Pg. 12, lines 7 – 11): “The water in the Cariaco Basin was supersaturated with respect to aragonite throughout the study. The thin aragonite shells of the pteropods are therefore chemically stable in the water column so it is unlikely that they underwent in-life dissolution. Furthermore, there is no evidence of patchy dissolution in pristine shells, or those which have undergone dissolution, such as has been observed in pteropod shells undergoing in-life dissolution in naturally undersaturated environments (Peck et al., 2016; 2018).”
Reviewer comment: P11L7/8: How can the authors know, pteropods were dead already? How likely is it that shell deterioration happened on the live organism? The assumption that any shell degradation took place only when organisms were dead already, is this simply based on the assumption that under aragonite supersaturated conditions no shell deterioration happened? If so, state clearly and support your view.

Author response: See response above

Reviewer comment: P11L20: : : : in the overall trend: : : (remove “is no”)

Author response: done

Reviewer comment: Fig. 4: It would help the clarity of the figure if September, June, December (mentioned in the text) could be indicated on the x-axis.

Author response: Good point – see updated figure (now figure 5)

Reviewer comment: Fig. 8: Italics for Heliconoides inflatus

Author response: changed

We thank Reviewer #1 for their thorough and constructive review of our manuscript. Their comments have helped us to improve the scope and clarity of our work

Fig. 1. Figure 5: Shell condition over the study period