

We thank reviewer #1 very much for their constructive comments and suggestions on our manuscript which will certainly help to increase the clarity and significance of our study. Please find below our point-by-point response in *italics*.

**RC1:** ['Comment on bg-2022-157'](#), Anonymous Referee #1, 12 Aug 2022

The article entitled "Zooplankton community succession and trophic links during a mesocosm experiment in the coastal upwelling off Callao Bay (Peru)" presents a "Timing" of new or timely research results (of special scientific interest), which have not yet been communicated to the public.

The authors studied micro- and mesozooplankton in mesocosms simulating an upwelling under extreme and moderate conditions of the OMZ (N:P) signature for 50 days. To check for possible differences, they also collected samples at a point in the Pacific Ocean close to the mesocosms (control sampling). They obtained values for the abundances of the main species observed, as well as values for stomach content (gut fluorescence), fatty acids, elemental composition (C, N, O, P) and stable isotopes ( $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$ ).

The results obtained may contribute to the knowledge of these first links of the marine food web of one of the largest eastern boundary upwelling systems (EBUSs) the Humboldt Current System, where small and medium pelagic fishes, main predators of the study organisms, are abundant.

The potential and strength of this work are undoubted, however, one of the great weaknesses found in this work has been the lack of a study scheme carried out with the mesocosms that include the study area (location). Not including this information in the work itself has made it very difficult to understand the procedure carried out.

*Response: We regret that the lack of study scheme and location area made it difficult to understand the whole experiment set up and working procedure. For a revised manuscript version, we therefore suggest to include a general schematic of a mesocosm together with a map of the study location and positions of the mesocosms off Callao and the sampling point in the nearby Pacific.*

Likewise, there is a lack of coherence between the hypotheses, the objectives (practically non-existent), the results and the conclusions that are vague or do not fit the results obtained. Therefore, I suggest this work for "Major Revision".

*Response: Both reviewers see a lack of coherence between results and the conclusion, moreover reviewer #1 sees the need of elaborating towards clearly formulated hypotheses or objectives. Our main objective was actually formulated in L77/78: "With an experimental approach we aimed at improving our mechanistic understanding of the interplay between upwelling processes and coastal plankton dynamics in the northern HCS." But we understand that our wording of the objectives and our approach was apparently not enough to the point. For a revised manuscript we would therefore revise these sections, formulate clearly our objectives and align the conclusions better to be coherent with the objectives and actual results (see also our response to some more comments of the reviewer on that subject further below).*

In the attached PDF you will see my suggestions for improvement, both in the main text and in the figures and tables. You will also see specific comments and technical corrections, in order to encourage the robustness and strength of this work.

**Citation:** <https://doi.org/10.5194/bg-2022-157-RC1>

*Response: Thank you very much for the detailed commenting in the pdf. Below, we have collected the comments and suggestions made by the reviewer in the pdf in order to reply to them:*

L1: I suggest "is one of the most..."

Response: *In terms of fisheries yield, the HCS is the most productive, therefore we would prefer not to change the wording in a revised manuscript version.*

L1: To be consistent with the rest of the document it is necessary to include an EBUS "S".

Response: *Yes, correct, we would include an "s" in a revised manuscript.*

L9: different...

Response: *different... will be included in a revised manuscript.*

L12: Any statistical evidence to prove it?

Response: *The differences in  $\text{NO}_x^-$  were relatively small ( $2.2 \mu\text{mol L}^{-1}$ ) but significant ( $p < 0.05$ ) (Bach et al. 2020). We suggest to change this part of the sentence to: "... but differences in nutrient concentrations established through OMZ water additions were only borderline significant.*

L22: Please, be more precise with your conclusions. Make them more aligned with the objectives and results achieved.

Response: *For a revised manuscript we would revise our conclusions. Please see further below for our detailed answer(s) to the further comments made by reviewer #1 to our conclusions. With respect to the respective sentence in L22 we suggest rephrasing to "Concluding, further mechanistic studies are needed to unravel links between intensification and increased frequency of upwelling and expanding/shoaling OMZs and trophic relations of the pelagic food web more precisely" for a revised manuscript.*

L26/27: Reference, "one of the", and space

Response: *The statement of the HCS being the most productive in terms of fisheries yield refers to Bakun and Weeks 2008 and Chavez et al. 2008 that are cited at the end of the next sentence. To make this clearer, we could connect the two sentences into one: "Among the four major...fisheries yield, that despite only moderate ..., sustains extraordinarily large pelagic fish stocks (Bakun and Weeks 2008, Chavez et al, 2008)".*

*Accordingly, as the HCS is actually the most productive in terms of fisheries yield, we would prefer to refrain from replacing "is the most" with "one of the" but remove the space after "yield" in a revised ms.*

L30: an

Response: *As suggested, we would delete "an" in a revised manuscript version.*

L37: This reference was mentioned in the first place, it should be "a".

Response: *We would correct this to Ayón et al 2008a in a revised manuscript.*

L72–89: Material and Methods section

*Response: L72–76 provide some literature information on drivers of zooplankton composition off Peru. We guess these lines were accidentally included in the section marked yellow and the reviewer comment actually refers to L77–89?*

Please rewrite the hypothesis, objectives, aligned to the question (title).

*Response: We agree and would rephrase the last paragraph in the introduction to better carve out our objectives and suggest the following text for a revised version: “Within the framework of a collaborative research project, in austral summer 2017, a large-scale in situ mesocosm experiment was performed in the coastal Peruvian upwelling region off Callao to investigate impacts of upwelling on pelagic biogeochemistry and plankton communities. An upwelling event was simulated by addition of two different types of OMZ waters to each four of the mesocosms. Within this collaborative effort, the aim of the present work was to improve our mechanistic understanding between upwelling processes and coastal plankton and food web dynamics. Our two main objectives were 1) to describe how the mesozooplankton (MeZP) community responds to an upwelling event with water masses of different OMZ signatures and 2) to describe trophic links in OMZ influenced waters. To address the first objective, we monitored the temporal development of the MeZP community in the mesocosms over a 50 d period. The second objective was addressed by analyzing the fatty acid composition and gut fluorescence of dominant copepods, by determining the stable isotope (SI) and elemental composition (C:N) of dominant zooplankton taxa, and by correlating dominating zooplankton as revealed in the present study with the occurrence of phytoplankton simultaneously monitored during this mesocosm experiment by project collaborators (Bach et al. 2020, Bernales et al. in prep.).” Furthermore, for a revised manuscript, we suggest to move the sentence in L81–84 “Our experiment coincided with a concurrent strong coastal El Niño,... at the start of the experiment.” to the start of the discussion section 4.1.*

L92: To better understand the procedure of this paper it is advantageous to have an scheme about the mesocosm setup and a figure about the location area... if hard to understand where are located any of this mesocosm... Although the information is in another paper, I suggest to include a brief information here...

*Response: As requested by the reviewer we would include a map showing the study site and location of the mesocosms as well as mesocosm schematic.*

L94: Any specific net? (details, name...)

*Response: It is not a specific net with a specific name. It is simply a 3 mm mesh attached to the upper and lower end of the mesocosm bags to exclude larger organisms from the enclosed water column during the time of deployment. These meshes are removed as soon as the mesocosms are closed at the bottom with the sediment trap and the bags lifted above the surface, i.e. when the experiment starts. For better understanding, we could include this information in a revised manuscript version.*

L97: Response: the comma after i.e. would be removed in a revised manuscript.

L98: Station (hereinafter St.)

*Response: We would introduce the abbreviation St. as suggested in a revised manuscript.*

L116: Please be consistent in all your work.

Response: We are not exactly sure what the reviewer means here? This sentence describes that the zooplankton was sampled with an Apstein net of 100  $\mu\text{m}$  mesh size from the mesocosms (i.e. it shouldn't be confused with the 3 mm mesh in L94).

L155/156: The reference should go here.

Response: As suggested, we would introduce the abbreviation "GF" after fluorescence and mention the reference Mackas and Bohrer (1976) after "method".

L165/166: Include "GF" when you use gut fluorescence in the first time, for example line 155

Response: As suggested, we would delete "gut fluorescence" here and use the abbreviation "GF" instead in a revised manuscript.

L167: gut fullness or gut fluorescence

Consistency in the terms and between the main text and the text of the figure legend

Response: Gut fullness and gut fluorescence are indeed two different things. In this case, we really mean fullness (i.e. the extent to which the gut is filled with food, Fig. 1), because the GF method needs to be applied during times of maximum fullness of the gut. Accordingly, gut fluorescence is the response measured to estimate on the extent of gut fullness.

L167: Results section

Response: In fact, during preparation of this manuscript we considered mentioning this part in the results section, but then had the impression this information was needed earlier to explain why sampling of mesozooplankton for GF determination was performed during the midst of night. For this reason, we would prefer to keep this sentence in the M&M section.

L170: 100 or 200? Please be consistent in all your work.

Response: Please note, we used two Apstein nets equipped with two different net bags of two different mesh sizes: 100  $\mu\text{m}$  for the regular collection of mesozooplankton samples for abundance determination (L115/116), and in case of copepods used for GF determination we used an Apstein net bag equipped with 200  $\mu\text{m}$  mesh size (for most possible gentle collection of organisms and to exclude as many as possible smaller zooplankton as for this purpose we were only interested in the larger adult female copepods). To help the reader recognize this difference, we would suggest to rephrase the sentence to: "MeZP samples were collected with the same Apstein net as mentioned above but equipped with a 200  $\mu\text{m}$  net bag..." in a revised manuscript.

L180: Any reference about this procedure?

Response: The procedure follows recommendations by Båmstedt et al. (2000) in "ICES Zooplankton Methodology Manual", p322–330. We would add a sentence at the start of this paragraph stating that determination of gut clearance rates followed Båmstedt et al.

L181: Consistency throughout the work, check, in some cases you use "sp" and in other cases "spp".

Response: We regret to have been inconsistent with the use of "sp" respectively "spp". Of course, we would correct this in a revised manuscript.

Fig. 1: GF or Gut fullness to be included in the axis.

Response: We would include GF to the y-axis title in a revised manuscript.

Fig. 1 caption: include units

Response: We would include "(ng Pigment Ind<sup>-1</sup>) after Gut fluorescence and remove "." After spp.

L191: Response: We would include "gut" to "The relative... fluorescence..."

L192: Are these units correct?

Response: In the literature gut fluorescence is often given as "ng pigment per individual". However, the unit "ng pigment per  $\mu\text{g DM}$ " is the standardized version that better reflects size variation and possible variation in the body constitution among the female copepods, which we prefer to use.

L198:

Response: We would replace "gut fluorescence" with "GF".

L251:

Response: "O" would be included in the heading

L264: Consistency of section title and main text.

Response: We agree and would include "O" in the section heading.

L291: And how does it affect the results and the study?

By putting this sentence, it seems that something is not right, don't you?

Response: This simply means that for the mentioned taxa, samples were not taken on each day or in each mesocosm. This is often the case in biological experiments, and the statistics used does account for that. We would add this information to the respective sentence: "For the two polychaete species as well as the microzooplankton, the sampling design was incomplete, i.e. samples were not taken on each day or in each mesocosm, but the statistics used does account for this fact."

L294: I suggest removing the full stop at the end of "Ind". Individuals per Liter; Ind L-1

Response: The full stop would be removed in a revised version.

L310: ... to the values found in the mesocosms, also after day 18 (Fig. 3).

Response: As suggested by the reviewer, we would add "compared to the values found in the mesocosms, also after day 18" to this sentence.

L310: The difference seems negligible (has a statistical test been carried out, to see if there are significant differences?)

Response: This statement relates to the lower curves of the upper graph of Fig. 3. "Others" in the Pacific (black triangles) have some higher percent contribution than "Others" in the mesocosm treatments also after Day 18 (black triangles fall clearly above the red and blue triangles of the mesocosm treatment means). We didn't do a statistical test because we have only one Pacific sample per time point. For the mesocosm treatments, confidence intervals are shown along with the means.

To make this clearer, we suggest to add "A" and "B" to the two subfigures of Fig. 3, and correspondingly add "(Fig. 3A lower curve)".

Fig. 3: text missing on x-axis

Response: Thanks for making us aware of this shortfall. The x-axis text would be added to a revised Fig. 3.

Fig. 3 caption: Very confusing to understand without information on the location scheme. Suggestion, why not create three columns or rows and put in one Pacific (control sampling) Second column or row (Mesocosms in extreme conditions) Third column or row (Mesocosms in moderate conditions) In this way we can still achieve a better interpretation of the results in the two subjected conditions.

Response: We agree and would rearrange Fig. 3 as suggested in a revised manuscript.

Fig. 5: cursive

Response: Species name would be formatted in italics.

Fig. 5 caption: Consistency

Response: "Sp." and "spp." inconsistencies would be taken care of.

Fig. 6: is cursive? Cursive

Response: Italics would be changed to normal font ( $L^{-1}$ , please note: the " $\mu$ " is not cursive).

For a better interpretation I suggest first, to put the axis with the same range as the "Copepoda" figure, and second, to see the curve better, you can put the "Copepoda" figure in the upper panel and the "Polychaeta" figure in the lower panel.

Response: The maximum copepod biomass is twice as high as the polychaete biomass. Putting them on the same scale will make the second panel rather illegible. We would therefore prefer not to change the scaling of the figure, but would agree to rearrange the two subplots to have Copepoda above the Polychaets.

Fig. 6 caption: I suggest modifying by "Copepoda" as shown in the figure. Consistency, please check throughout the work. I suggest modifying by "Polychaeta" as shown in the figure.

Response: Copepods would be changed to Copepoda and polychaets to Polychaeta.

L396: Include figure 7 after it is indicated in the text.

Response: The layout of a final manuscript would be the responsibility of the Biogeoscience production office. In general, we would also prefer to have Figures as close as possible to the referring text passage. Of course, we could try to place Fig. 7 closer to its text in a revised manuscript, but it will always depend on the remaining format how well this can be realized (and will not be the final layout anyways).

L401: I suggest including as supplementary material.

*Response: GF data will be made available via the PANGAEA database. Therefore, we would suggest to include the PANGAEA doi link at this place in a revised manuscript version?*

L408: Insert Table 1 after this paragraph.

*Response: Usually, reference to Tables and Figures, respectively, follows after the first mention of any of their content in the text. Therefore, we would prefer to refer to Table 1 here together with the first mention of results summarized in Table 1.*

Fig. 7: correct

*Response: The "L" would be changed to normal font.*

Fig. 7: I suggest placing the "Pacific" curve on top of the other curves, as the dots are not visible.

*Response: Would be changed as suggested in a revised manuscript.*

Fig. 7: Please, include the units here.

*Response: Units would be included as suggested in a revised manuscript.*

L424: Insert Table 2 after this paragraph.

*Response: As mentioned above, we prefer to refer to Tables (and Figures) at first occurrence of any of their contents in the text. But we would re-check with Biogeoscience format guidelines what the correct way would be.*

Table 1: Please correct the units to be the same as in the main text. Include the units in the text of the table legend.

*Response: We would correct the units to be the same as in the main text and include units in the legend.*

L437: spp., Sp.

*Response: We would check the whole document for consistency in a revised manuscript version.*

L466: ?

*Response: Thanks for spotting this wrong Table reference. Of course, this should be Table 2 and would be corrected in a revised version.*

L476: I suggest including as supplementary material.

*Response: We agree and would include pair plots showing Pearson correlations (per mesocosm and for data pooled per OMZ treatment) between measured GF of copepods and phytoplankton/microzooplankton protist groups as supplementary material together with a revised manuscript version.*

L476: This is the first time "protist" is used in main text, please include in parenthesis the groups of protists, already referenced in the text, consistency.

*Response: Protist groups would be included in this text passage.*

L480: P value?

*Response: As agreed already with one of the previous comments, we would provide supplementary material with Pearson correlations and would include the requested p-values (respectively Pearson correlations) in this paragraph.*

L485: statistical information should be included

*Response: P-values, respectively Pearson correlations would be included here, too.*

L495: Why not include as supplementary information?

*Response: These data will be publicly made available on PANGAEA and the doi link would be included in a final manuscript version.*

Fig. 8 caption: Include what the grey dashed lines mean.

*Response: In a revised version we would add "Vertical dashed lines indicate the three phases according to Bach et al. (2020)."*

L508: Include Tables 3 and 4 before discussion.

*Response: This is again more a matter of the final typesetting with Biogeoscience production office. If we would change it for a revised manuscript version, it wouldn't be final anyways.*

L509–511:

The first paragraph of the discussion is information that is not observed or presented in the results of this work, it is information that has already been published previously. It should be rewritten for the avoidance of doubt.

In the results you have not mentioned "phytoplankton" so, although we can understand what you mean, we have to use the same line throughout the paper.

Please try to modify the text of the discussion to be in line with the results obtained.

*Response: We are not exactly sure, if we understand correctly what the reviewer means here, especially with "we have to use the same line throughout the paper". Although we have referred to Bach et al. 2020 several times in this paragraph it is apparently not clear without doubt that what we summarize here to introduce some of the key results of the entire experiment is results presented in Bach et al. To enhance clarity, we would certainly rephrase this text passage starting with the mention that in this paragraph we report results by Bach et al. to provide some basic information on the main outcome of the entire study for the following discussion. With respect to the phytoplankton, in the revised objectives (last paragraph of the introduction), we mention now correlation with phytoplankton. We think, this will help the reader to see the recurrent theme.*

Table 3: [ $\mu\text{g Ind}^{-1}$ ]

*Response: We would correct the unit to make it consistent throughout the document.*

Table 3: I suggest including which are polychaetes and which are copepods, at least between parentheses or in a new column on the left, for better interpretation.

*Response: We would revise Table 3 and indicate which are copepods and which the polychaets.*



Table 4: Isn't it "molar"? Please check consistency with the main text, lines 505.

Response: Yes, correct, it is molar and we would add "... as well as their molar ratios."

L516: Result section

Response: This summarizes results from Bach et al. to describe some wider context of the entire study for the following discussion, i.e. it represents not our results but is introductory information to help put our results into context with the entire study. Therefore, we would like to keep this passage here.

L538: say which group.

Response: We would replace "on this group" with "on copepods" in a revised manuscript.

L558: There is no mention of "eggs" in the results section.

Response: Egg production was not measured in our study, but as mentioned in this sentence we frequently noticed egg carrying females of *Oncaea* and *Hemicyclops* during processing of zooplankton samples. We suggest to include "(personal observation)" at the end of the sentence for clarification.

L658: The actual text that is in the conclusion should be in the discussion session.  
Please list 3 or 4 conclusions you draw from your work in this section.

Response: We agree and would include the conclusion text in the discussion (for example L659–668 following L591, and L669–675 following L657). With respect to the second part of this comment, please see our response to the comment in L675 below.

L663: Egg production has not been studied in this work, therefore this sentence cannot be a conclusion of the results achieved.

Response: We would rewrite the conclusion as requested by the reviewer and, as mentioned in response to the previous comment, would move this sentence to the discussion.

L675: Any conclusions on "Zooplankton community succession and trophic links during a mesocosm experiment in the coastal upwelling off Callao Bay (Peru)"?

Response: We have reconsidered our conclusion section and would suggest the following text for a revised manuscript version: "Our experiment provided some interesting insight on the response of a zooplankton community and their trophic relations to an upwelling event with OMZ waters. In particular grazing rates (gut fluorescence) and fatty acid compositions revealed that feeding of dominant copepods on autotrophic food (diatoms) was insignificant and the question arose whether copepods were starving. However, our methodical approach does not allow conclusion on the potential importance of omnivorous feeding. Likewise, the question whether starvation was responsible for reduced fecundity of copepods or whether the loss of eggs (of broadcasting copepods) to the oxygen minimum layer in the mesocosms was the main reason for the low numbers/lack of copepod nauplii found in the mesocosms remains open. To answer this question, future studies need to better constrain the nutritional condition of the copepods (body size/mass, grazing experiments: importance of omnivorous feeding?), the importance of microzooplankton grazing (dilution experiments) versus copepod grazing in the food web, and determine copepod fecundity through determination of instantaneous *in situ* egg production rates and hatching success. Measures of nutritional condition would also help a profound interpretation of isotopic signatures in relation to starvation of organisms. In this way, trophic links in a plankton community under OMZ influence

*could be investigated in much more detail and more profoundly, but implementation of further methodical approaches would also require much more time and manpower.”*

L684: Last author

*Response: We think this is more a matter of personal taste and would prefer to keep it as it is (also it is coherent with other papers resulting from this study published in this BG special issue).*

L783–788: The order of these two references by Franz et al. should be changed

*Response: References were automatically ordered by the Biogeoscience bibliography style according to the journal specific style.*