Dear Editor,

Thank you for your detailed, constructive and helpful reviews! Your thoughtful comments will certainly improve the clarity and accuracy of our manuscript, and have caused us to reexamine certain decision more deeply and to explain our reasoning more explicitly. Please find our responses below. We have copied editors and reviewer comments in italics, and our responses are below each italicized reviewer comment. Where line numbers changed substantially, we have included the revised line number (e.g., “now line 531”) for easier cross-referencing to the new line numbers in the revised draft.

One question we have for the editor and peer reviewers--would you all be comfortable with us putting this Response to Reviewers (including the great comments from the Editor and Peer Reviewers) in our public repository for this manuscript? We completely understand if the preference is to keep this confidential, which we’ll happily oblige in that case. We wanted to ask for the purposes of full transparency and also for providing a teaching tool that I could use in the future for early career researchers who are new to the peer review process and what it might look like in greater detail.

Please feel free to email me ([helena.mcmonagle@wellesley.edu](mailto:helena.mcmonagle@wellesley.edu)) if you’d like a version of our revised manuscript and supplementary file that has all tracked changes from the peer review process shown.

Sincerely,

Helena McMonagle

On behalf of co-authors Joel Llopiz, Amy Maas, Deborah Steinberg, Annette Govindarajan, and Timothy Essington

**Response to Editor comments:**

*Thank you for your work on this interesting study and your submission to ICES. Both reviewers and I agree that the manuscript provides timely and important information and has strong potential to be a valuable contribution to the literature. To strengthen the paper, the reviewers raise thoughtful points below. We look forward to considering the manuscript for publication following revision according to these comments.*

Thank you for this feedback! We appreciated all these thoughtful suggestions, and believe that our revisions have substantially improved the manuscript by addressing these peer review comments.

*I agree with the reviewers that important details from the supplemental material should be moved to the main text of the manuscript. In addition to making the changes asked for by the reviewers, please briefly describe the methods of Appendix A in the main text associated with line 148. Please also briefly summarize the gut contents in Appendix F in the results.*

Line 148 (now line 164): We agree it would be good to briefly describe the DNA barcoding methods in the main text rather than fully relying on the Appendix A and we have done that now starting at line 170.

Regarding the suggestion about Appendix F: Great idea. We have briefly summarized the results of the gut content analysis right after Figure 6 (currently lines 506-511).

*From a writing perspective, there are a few places where stronger resolving sentences could improve the clarity and impact of the paper. For example, in lines 319, 504, 513, and 549, the messages of these paragraphs could be explicitly stated to conclude these sections and connect to the implications of these points.*

Thanks for the suggestion to be more explicit in our conclusions. We have provided more explicit take-home messages to these paragraphs:

* Previous line 319, now 384-388, regarding the zooplankton counterfactual issue,
* Previous lines 504 and 513, now line 617, regarding take-home messages from the paragraphs previously ending in lines 504 and 513 regarding biomass estimation using nets, eDNA and acoustic technology
* Previously line 549, now 719, regarding the unrealistic but common assumption that predation and other mortality automatically results in downward fish carbon transport

*Please check the figure presentation for color-blind accessibility to support a broader readership. For example, in addition to the color codes for family on Figure 3, add text labels grouping each family.*

Good reminder about color-blind accessibility. We have used ggplot’s “Dark2” color palette, which is color-blind friendly. We agree with your suggestion to also add text labels where possible and have added these labels in the caption of Figure 3.

*Thank you for publishing your data and code, these efforts greatly enhance research reproducibility and transparency, and we appreciate your work to provide these valuable resources. The reviewer’s comment about a repository with a doi number would help increase the long-term availability of these and the impact of the study.*

Appreciate the feedback about open and reproducible science. This is really important to us too, and we will make data available via SeaBASS (managed by NASA) with a permanent DOI upon the proof stage prior to publication (in case any further edits from the peer review process are required before that point).

*Minor comments:*

*L470: Briefly explaining here why this study site requires this remineralization depth would help the reader understand applicability of these findings to other sites.*

We have reworded and added to this explanation in line 408.

*L579: This discussion of the mixed layer depth variation is an important consideration that could be strengthened if the idea were set up in the introduction and then directly connected to the current data in the discussion.*

We have further explained this variation in ocean mixing in line 531 (see comment below).

*L72, now 81: Explain briefly how this natural variation could occur, ex: seasonality, region*

We have edited this to include temporal (e.g., could be seasonal or annual differences) and regional variation.

*L149: Check phrasing on “highest taxonomic level” – should this read lowest level? A clearer phrasing could be “most specific taxonomic identification possible.”*

Thank you! We have now clarified this point about taxonomic level (see response to Reviewer 2), and have added your suggested definition of “most specific” taxonomic identification in line 163.

*L181: Add this point to the abstract, clarifying that larvae are excluded from the study.*

Done. We have now been explicit in the abstract (line 23) that we examine adult fish.

*L492, now 602-615: Please include additional discussion of the implications of patchy distributions both for the interpretations of these findings and for future research.*

We have included some discussion of the implications of patchy distributions for both interpreting our findings and for future research starting line 604-619, and also in a later paragraph starting line 636.

*L378, now line 428: To enhance transparency and reproducibility, please briefly explain why this is the appropriate statistical test either here or in the methods.*

We have further explained why we chose the Kolmogorov-Smirnov test here for differences between groups.

*L421, now 479:* *Briefly defining t1, 2, and 3 here in the legend would improve interpretability of this figure.*

We have added this description in line 479.

*L470, now 408: Briefly explaining here why this study site requires this remineralization depth would help the reader understand applicability of these findings to other sites.*

This is a result from Siegel et al., 2021 (e.g., shown in Fig. S2F). At our study site (and nearly the entire globe), carbon sinking to ~3000 m can be assumed to be 100% sequestered for a time horizon of 100 years. We have re-worded this sentence for clarity and briefly explained why this study site requires a deeper depth of sinking to be fully sequestered (overturning circulation at this study site, lines 408-412).

*L579: This discussion of the mixed layer depth variation is an important consideration that could be strengthened if the idea were set up in the introduction and then directly*

Agreed. This is further addressed in the paragraph that includes line 408.

**Response to Reviewer 1 comments:**

*General comment: It’s fishes, not fish (when plural for >1 spp).*

Thanks for catching this. We have edited this in the text where we mistakenly put fish. When we are listing fish in comparison to zooplankton, we keep this term as a general category of marine life as it is clear in these places that we are referring to fish in general (e.g., “We sampled fishes and zooplankton” sounds a little clunky in our opinion). We also leave “fish” where it is used as an adjective, e.g., “fish carbon flux” or “fish biomass” as we understand you are referring to the use of fishes as a noun.

*Methods general comments: I think it would be helpful here to report on the overall model structure early in the methods so we know what the relevance of the various terms are. There’s a couple of places where I have questions that aren’t answered until a later paragraph, usually due to information being split up through the section. It’d be helpful to include some of this info at the same time as it’s raised (e.g., you mention that a MOCNESS is used, but don’t state the sizes / capture efficiency assumptions etc. until a later paragraph).*

We agree, and other reviewers requested additional detail upfront in the Methods section as well before it is introduced in a later paragraph or even supplement. We have added a substantial amount of detail to the Methods section.

*It would be useful to briefly include a description of how the mortality pathway is estimated, just so we don’t have to jump between papers, especially given that this is the parameter that’s arguably hardest to measure.*

We have further explained our mortality flux estimate in lines 367, 389 and 405.

*Zooplankton flux: Perhaps it’s explained more fully in your other paper, but I think the logic here for why you include zooplankton vs. fishes in the way you do needs some more explanation. I think the assumptions you’re making here about zooplankton being consumed only by fishes is a particularly big leap that ignores other high-biomass migrant grazers (e.g., crustaceans), and gives a lot more zooplankton mortality to fishes than would occur in reality. If this is simply a proxy for zooplankton mortality overall, maybe it doesn’t matter so much, but if that mortality feeds back into the fish production that could be a far bigger issue. Either way, I’m left*

*confused after reading this section and I think the inclusion of zooplankton overall needs a bit more justification / explanation for me to understand how it fits in.*

Agreed. Another reviewer had a similar comment and we definitely want this to be clear. This zooplankton counterfactual issue is truly a gap in how fish carbon flux is typically calculated and compared to other carbon fluxes. We have added a substantial amount of explanation in lines 384-388.

*Appendices: I read the appendices after the methods, and there’s some useful information in there that would likely improve the main manuscript. Specifically, much of the information in Appendix C is extremely helpful, and a summary of it in the main text could really help explain why you did / did not present certain datasets. Similarly, some information from Appendix G would be useful to include in the main text (see specific comments relating to zooplankton).*

We have added substantially to the Methods section to move some of the details from the supplement into the main Methods text. We summarize Appendix C starting at line 202. We have summarized the results in Appendix G in Fig. 7. Beyond that, detailed description of the zooplankton carbon flux at this study site is beyond the scope of this paper on fish carbon flux, and the zooplankton results will appear in greater detail in subsequent publications from this cruise effort.

*Table S1: I would expect this in the main text, given that this is primarily a modeling study and these are the parameters and data sources used.*

We would have liked this to appear in the main text too, but this table is nearly 3 pages long and thus too large to include in full in the main text given formatting constraints (though we would welcome a correction to our assumption here from the editor). Also, a very similar table in terms of content and format appears in McMonagle et al., 2023. We make the supplement as accessible as possible by linking it in the journal publication as well as storing this table in our Github repository in the copy of the supplement there. Parameter values from this table are also available in our published code. We do briefly mention the parameter range used for *q*, capture efficiency of the nets, given that this is such an influential parameter according to our results.

*I also think that Figures S1 and S2 are probably important enough to move to the main text, perhaps replacing Fig. 8, or Figs. 4 – 5 as appropriate. Again, since this paper is a modeling study, I think it’s important to present the model within the main text and move more of the supporting data figures to appendices if you are short on space. I will leave this decision to the authors however.*

These figures already appear in the main text of McMonagle et al. 2023 and are nearly identical there, and thus we have chosen to include these in the supplement. We have summarized the results of the sensitivity analysis though in line 559 and 608, so that a broader readership not looking to compare their results with ours in greater detail can still obtain the main results of these figures. Figure 8, however, is a completely new result that is not already presented in McMonagle et al., 2023 because in our previous paper, we did not examine uncertainty in fish biomass (that study was looking at bioenergetic-related parameter uncertainty of a single migrating or non-migrating fish).

We considered removing Figs. 4-5, but given the huge influence of biomass uncertainty, we have chosen to leave them in because it shows that catch per tow is highly variable and that a larger net does not necessarily have the impact on catch efficiency that one would intuitively assume. We believe these results could be useful for a slightly different audience interested in constraining uncertainty in fish biomass estimation from net sampling. We have made additional comments regarding this implications of the patchy distribution that we seemed to see, as shown in Fig. 4, in line 617.

*Line 107 – 108: “retentive” implies that you believe some fauna will be entrained into the anticyclonic eddy – is that a fair assumption? This assumption could be justified in the intro a bit more I think, if it’s relevant to your topic. If the eddy has effects on the fauna associated with e.g., downwelling / greater water clarity, we might expect changes in the vertical distributions and migration behaviors of some fauna (see e.g., https://www.science.org/doi/10.1126/sciadv.1602468). It is not clear to me if / how the potential effects of this eddy are considered in the rest of the manuscript. Please add further clarification as to which samples occurred inside / outside /on the edges of this feature.*

We unfortunately do not have sufficient sampling effort nor a clear enough relationship between catch and distance to eddy core to draw conclusions about biomass with relation to location within/near eddy to go into greater depth on that in this manuscript. However, future research using EXPORTS data from this North Atlantic study site may find it relevant that the three-ship experiment was based around an anticyclonic eddy. Given that this explains our coordinates throughout the cruise, we decided to leave in that the sampling was essentially following an anticyclonic eddy, though this predictor of biomass is not pursued further in this manuscript. To avoid this sentence being misleading, we have removed “retentive” and just retained basic mention of the anticyclonic eddy within/near which sampling occurred (line 121).

*I don’t believe the potential effects of this eddy are considered again in the manuscript, but could they explain any of the differences between MOC-1 and MOC-10 catches for example?*

We had a similar question so we looked into this. However, even when we towed in the same time and place with both ships/net systems, we still see large differences in catch between the two net systems.

*Line 129-130: It’s not immediately obvious why you calculate areal biomass from volume data. I’m guessing this calculation is intended to get you an estimate of fish density in some volume under a a 1m2 area? I think this detail could be clarified better because I expect .*

Our reasoning for this unit choice for biomass is simply that the biomass measurement is ultimately used for a fish carbon flux estimate, and carbon flux per unit square meter is a more common unit used in the literature (as we add in lines 150-151 in response to your suggestion). We agree it is somewhat counterintuitive compared to biomass per unit volume, but this unit choice allows for better comparison to the broader literature of carbon flux via the biological carbon pump (e.g., via phytoplankton growth and sinking), which is typically presented in areal units. Our code contains calculations and code for figures that presents these results in units of biomass per unit volume filtered as well in case anyone is interested in digging into our results in greater detail.

*Lines 135 - 136: I don’t see “day” and “night” defined in the text at all. Were these centered on solar noon / midnight or some other times? Adding clarification as to when the tows were made in relation to sunrise / sunset would be useful information to have. On a similar note, were the tows made from deepest to shallowest, or shallowest to deepest?*

We were not always able to time our tows to occur at the same time during the day and night on both ships, in part because there were several other types of deployments occurring on both ships in addition to the net deployments. We further describe the net sampling in line 133: “As is typical for MOCNESS tows, samples were collected while the net was being retrieved from its deepest to shallowest sampling depth.” We further define day vs night in line 136: “Day tows are defined as those that occurred after the downward migration of the deep scattering layer was complete (i.e., after this layer as shown on an echogram reached its deepest depth), and before the upward migration began in the evening. Night tows are those that occurred after the upward migration was complete and before the downward migration began in the morning.”)

*Line 184 – 187: This section isn’t completely clear to me, but it seems much of the methodological explanation is in an appendix and then a number of figures are presented in the results section. I’d suggest either moving everything relating to catchability to the appendix, or include a more complete explanation of this process in the main text rather than splitting the information across multiple places.*

We hear this point, but we made this decision reluctantly after considering the relatively long length of the manuscript already. The data filtering section of our methods is already a page long, and the more detailed description of our data filtering decisions described in Appendix C do not substantially impact our catch results (e.g., excluding extremely rare species does not substantially change our results, as we further explain in the main text in our revised line 479-483). However, for the sake of reproducibility in case anyone ever wanted to repeat our results step-by-step and understand each decision made (and documented in our code in moving from raw to processed data), we have included Appendix C to aid potential future users of our data. Adding the entirety of Appendix C to the main text would add 2 pages to the manuscript main text length, which we don’t believe is necessarily essential for obtaining the main results of the paper.

*Lines 190 – 193: I think this needs more detail. How did you know what was fish / non-fish? How / when / where were the acoustic surveys made that would allow you to estimate migration rates of just the fishes? I don’t believe the acoustic data are ever referenced again, so perhaps this can just be removed.*

We have added in lines 222-223 that we assume that the dominant deep scattering layer is representative of the bulk of fish migrators. We believe that the use of acoustic data here provides an improved estimate from relying solely on literature values, though in reality we certainly agree with your point--deep scattering layers contain far more taxa than just fishes and not all mesopelagic fishes contain gas-filled swim bladders that appear clearly on echograms.

Also, we have now mentioned the acoustic echograms again in our methods where we define day vs night delineations.

Acoustic data was used to the extent possible (albeit roughly) to avoid having to rely solely on literature estimates of fish swim speeds, which is what has been done in the past for published studies of fish carbon flux. Acoustic data and analysis is certainly not a focus of the paper, and for reasons related to imprecision of calculating swim speed from the echograms as you point out, we still allow swim speeds to vary substantially in our sensitivity analysis. Further detail about how swim speed was estimated from echograms can be found in our data folder in the public repository (data > Swim\_speed\_from\_acoustics.xlsx), which we now mention in the Methods section as well for easier navigation to that information for readers who really want to dig into these details and for reproducibility.

*Line 203: What was the assumed net efficiency and why was that value chosen?*

We have added this in line 248, and the assumed net capture efficiency is also given in Table S1 (though we agree that this also merits some additional text in the main body of the manuscript’s methods). We have also added sources that appear in Table S1. As shown in our sensitivity analysis results, uncertainty in this parameter is highly influential in overall biomass uncertainty, and the reality is that this parameter is very poorly constrained in the literature due to the inherent challenge of measuring it.

*212 – 216 (now 244): I struggled to follow this section on a first read. I think it would be helpful to move the parameterization of the net efficiency up to this paragraph rather than leaving it to later.*

We have now added additional explanation about capture efficiency parameterization in line 248. We agree this is well worth including at this point in the manuscript.

*Fig. 2: Please reference the colors in the legend for additional clarity, and check the colors used here for color-blind accessibility. I suspect some people will struggle to distinguish at least the green and pink here.*

We used colors from ggplot’s “Dark 2” for these figures, which is a color-blind friendly palette.

*275 – 277: I think that considering additional mortality within the mesopelagic and epipelagic zones is important, but it seems like the aim described in these lines is to add some extra mortality to the zooplankton population. By limiting your consideration to “fish-based” mortality (per your observed biomass estimates), you’re certainly underestimating the real mortality from all causes, and I can’t help thinking that the simpler solution is simply to add a mortality-at-depth term to your model rather than try to estimate all the parameters around (only) non-migrating fishes. Gelatinous zooplankton and invertebrate (especially crustacean) grazers are likely to be really important consumers at depth as well as fishes, and ultimately, if the carbon is being consumed by a modelled non-migrator, it is effectively all staying at depth anyway.*

We agree that this point about a counterfactual to fish carbon transport via predation on zooplankton needs to be clarified. Our Editor made a similar suggestion. Unfortunately, the data to quantitatively model mortality flux is simply not available at this study site, or in any other published fish carbon flux study to our knowledge. However, we have more explicitly explained our decision about how we account for zooplankton mortality flux in lines 365 to 389.

*Line 292 – 301: You adjust your MOCNESS catches to account for catchability, but not the sediment traps (which will also underestimate total flux for a variety of reasons: see e.g., https://scor-int.org/Publications/WG116.pdf). It would be useful to briefly describe the type of sediment trap used here.*

We have described some information about the sediment trap sampling in lines 349-354, and we add additional detail in this paragraph. We refer readers to Estapa et al., 2023 for additional details about sediment trap sampling (which we pull from for comparison in our study, but which is outside of the focus of our study).

*Line 299 – 301: It’s not clear here that the Thorium method was used in the modelling. Please clarify / remove reference to the unused method as appropriate.*

We have clarified how the Th-234 results are used in our manuscript in lines 354 and 361. The Thorium method is used for creating Fig. 7

*Line 304 – 305 (now 365): The paragraph begins by referring to zooplankton, but then refers to “these fishes”. Please clarify this bit. Later, the paragraph seems to switch from MOC-1 to fish sampling and I’m struggling to follow it.*

We made a typo here, thanks for catching it. We have added text to clarify in this paragraph and have fixed our typo.

*Line 304 - 307: I’m struggling to follow this section. Can you clarify here exactly what is being compared and how?*

We have added quite a bit of explanation for clarity in this paragraph in lines 364-389, which will help readers understand the decisions we made for comparing fish to zooplankton carbon flux (when in reality, these carbon fluxes are interrelated via predation).

*Line 309 – 310 (now 373): I think it’s fine in this paper to say that you focus on fishes and zooplankton specifically, but I don’t think it holds that you “need” to include zooplankton here.*

True, we have taken out “needed”.

*317 – 319 (now 382): Presumably you’re only referring to migratory zooplankton and so diurnal epipelagic consumption is considered irrelevant. Is that correct?*

Migrating mesopelagic fishes could be consuming diurnal epipelagic zooplankton as well, which is further described in McMonagle et al., 2023.

*Results:*

*General comments: Since your zooplankton and sediment trap data are described in the methods, I’d expect them to be presented here (e.g., flux rates to different depths; zooplankton t-plot), but they’re hardly discussed. Given their importance as part of your comparison, I think some text about these elements should be added to the main text.*

We do not go into detail regarding sediment trap results beyond our short summary (which we added to in this version) starting at line 351 because this work was not ours, but rather is presented in a published study that we cite (Estapa et al., 2023). Similarly, the zooplankton active flux results are being prepared for publication elsewhere (Maas et al. in prep). Discussion on the taxonomy and distribution of the zooplankton communities is being fully discussed in a third manuscript (Steinberg et al. in prep). As the focus of this paper is the uncertainty in the contribution of fish to the biological pump, we limited our text solely to comparisons of magnitude and temporal variation to the other pathways measured in the EXPORTS project.

*In addition, as much as it’s not a focus of your study, I would be interested in knowing what proportion of the catches were comprised of non-fish taxa (for zooplankton and micronekton). A quick look at the data files suggests you were catching a decent number of crustaceans (although I found it hard to tell which files were showing what!).*

The mesozooplankton were dominated by crustaceans such as copepods and ostracods; the micronekton were dominated by hyperiid amphipods and gelatinous organisms including pteropods (numerous pseudothecosomata as well as some large euthecosomata) and cnidarians. These taxonomic findings are currently being written up and will be the focus of a dedicated manuscript (Steinberg et al. in prep).

*Line 331: I think it’s fantastic to have the R code etc. available through GitHub (which is beautifully commented!), but I’m curious if the data have been stored anywhere more permanent as well (i.e., somewhere they can be assigned a doi)?*

Yes, the data will also be available via NASA’s SeaBASS database. We have added this to our data availability statement.

*Line 353: See previous comment, but is this “areal” measure really representing density within a 1000 m3 column? It’s fine if this is standard terminology, but I’m struggling to see how you could calculate density except as a volume. Please clarify.*

We have added some explanation of our areal biomass metric in line 150-151.

*NB: You provide density per volume filtered in Fig. 6, which is what I’d normally expect.*

Correct. We use these volumetric units here because this figure is showing biomass differences per depth interval, so we have not yet here multiplied by the depth interval that is used to get volumetric biomass into units of areal biomass. We believe that this is the most comprehensible unit to use for this figure, which is really just meant to show dominant migration behavior for each fish family. While in many contexts volumetric biomass units are more intuitive, we have chosen to stick with areal units to allow for comparison to the carbon flux literature.

*Line 376 – 378 (now 478): I’d expect this result, given that you’re comparing 90% of the data to 100% of the data. A more interesting comparison from my perspective would be the MOC-1 sizes vs the MOC-10 sizes.*

We agree that this result is not surprising. We presented the results of the statistical test to justify the data filtering choice (i.e., to show that removing rare species doesn’t impact our overall results). We include differences in MOC-1 vs MOC-10 sizes in Fig. 5.

*Fig. 6: This is only MOC-1 data? I’d like to see MOC-10 included as well (probably as a second panel). NB: I found the explanation for this in an appendix, but in my opinion it should be included in the main text to explain why the results are only for a part of the dataset.*

Good idea. We have summarized and added these reasons for using the MOC-1 data for Fig. 6 in the caption for this figure.

Also, thank you for your close reading of not only our manuscript but also our appendix. It has certainly improved the manuscript in multiple dimensions including readability, reproducibility, accuracy, and thoroughness of explanation.

*Fig. 8: This figure needs units on the x-axis. As-is, I’m struggling to understand what I’m looking at. It’s also labelled as uncertainty (which I read as “variance”), but seems to represent the range of outputs generated across the parameter spaces. Please clarify.*

Our definition of uncertainty is rooted in the Monte Carlo sensitivity analysis. Given that the Monte Carlo simulations provide 1000 results, we then need to define our lower and upper range in some way, which we do using quartiles (a little different from variance). There is no unit in the x-axis because this axis represents categories--these categories are which type of parameters we allow to vary in the Monte Carlo simulations. We have described the x-axis further in this figure caption. The y-axis shows the distribution of estimated carbon transport under different simulations that explore different contributors to uncertainty.

*Section beginning on Line 457 (now 578): I think a lot of this text should be in the methods section. Please move as appropriate so this is just results.*

We have moved most of this results paragraph to the Methods section (starting line 399). We had tried a detailed explanation of the calculations in both sections in our editing process, and in the end we agree with your suggestion to keep all this information in Methods. We add in the results (line 601) that readers can refer back to how we came to these values by looking at our methods, and we provide a reference to the code where these calculations are done.

*Discussion:*

*General Comments: The discussion starts with a discussion of parameter uncertainty and then moves into a discussion of the potential flaws in the data sources in a way that I read as a little negative. I’d suggest starting the discussion by summarizing your main / most important results first (e.g., carbon flux estimates) and then move into considering the implications of the dataset used and the various assumptions used in the model. A reminder of the main results will also be helpful for those readers who inevitably skip the methods and results sections! Discussion of sources of error / variation should come later in my opinion.*

Great idea! We have provided a more detailed response to this suggestion above in our response to the editor, with specific line numbers and descriptions for each addition related to the main takeaway point/interpretation in the discussion section.

*I would also like to see a bit more consideration of the rest of the biota that weren’t included in your current model, such as the potential impacts of larger size classes, invertebrate micronekton, gelatinous zooplankton, the epipelagic diurnal foodweb etc. I don’t mention this expecting you to include them in your model at all, but acknowledging the other components of the ecosystem I think will help put your results into a wider context. There’s much more out there than just fishes!*

We have added this discussion, and a rough calculation for comparison to whale carbon flux (lines 678-697). There are limited examples in the literature of carbon flux associated with higher trophic levels that were available in units that we could use to compare to our study, but this is a good starting point for much more detailed future research in response to your suggestion.

*Lines 506 - 507 (now starting line 646): I think this paragraph is an interesting one for sure, but I’d like to see more discussion here in terms of how your data choices could have influenced the model results (i.e., do you think your model outputs are more / less conservative as a result of your choices) for example.*

We have added clarification to the take home message in this paragraph. It is not clear whether our results are more or less conservative because some decisions could have resulted in an overestimate while others may have resulted in an underestimate.

*Lines 515 - 529 (now 661): The title doesn’t seem to match the content of this section, which appears to consider temporal variance in the sample data.*

We have revised the topic sentence to this paragraph to align more with our results. We describe the limitations of our study given that it is a snapshot in time; at a non-bloom sampling time, the contribution of fish is likely much higher. To our knowledge there is no study examining seasonal differences in fish-mediated carbon flux, but this could be a worthwhile area for future study if ultimately it is average annual carbon fluxes and sequestration rates are the most relevant estimates from a climate perspective.

*Line 527 – 528 (now 673): Here you state your estimates are likely relatively lower than the annual average, but express in the next paragraph that your estimates are likely higher than reality across the board. Perhaps this section can be reworded a bit to clarify the more subtle contexts here because as-is it reads as somewhat contradictory and I’m not sure whether to take your findings as too high or too low overall.*

See comment above about over vs underestimation. While we have tried to describe the likely impact of our modeling choices on the bottom line carbon flux result, we cannot say whether we are left with an over- or under-estimate. We have removed part of this paragraph to avoid a contradictory message while emphasizing the context in which this study occurred.

*Line 556 (now 727): I think the role and type of zooplankton included in the model needs to be clarified throughout the manuscript because up to this point I was under the (possibly incorrect) impression that migratory zooplankton were modelled similarly to fishes (i.e., their contribution to active flux was estimated as implied in the methods (lines 303 - 319)).*

Zooplankton are not included in our model--rather, the output of an ongoing zooplankton carbon flux study using data from this same cruise was used to put fish carbon flux into context of the broader biological carbon pump, which includes the contribution of zooplankton. We believe that the additional descriptions in the revised manuscript of how zooplankton play into this study will help readers.

*Section beginning on line 551 (now 722): Do you have suggestions here for future research priorities that could be used to reduce uncertainty in our estimates / future management of the oceans?*

We have added our suggestions per your and the Editor’s suggestion. At the moment, biomass related uncertainty seems to be here to stay, but future technologies (or refinement of existing technologies) may constrain biomass uncertainty in the future. We are not in a position to speculate more on this.

*Line 575 (now 746): If 1000 m is the minimum limit for long-term carbon sequestration, do you think these kinds of mesopelagic studies are of value? I would say they do, but it would be nice to see this addressed in the discussion a little, since you raise the issue!*

According to Siegel et al, 2021, 1000 m isn’t necessarily the limit for long-term carbon sequestration--however, scientists and policy makers would be misguided to assume that 100% of the carbon that is transported to shallower depths (e.g., 200 m) is sequestered when this proportion could be much lower. The depth at which carbon is transported, form of carbon, and the oceanographic context at that site (see additional comments on ocean circulation for example) and ultimately, carbon sequestration time horizons ought to be considered more rigorously. We believe this is particularly important to emphasize given the renewed interest and investments in marine carbon dioxide removal research and methodologies, which we think our discussion communicates without overstepping.

**Response to Reviewer 2 comments**

*My comments mostly comprise of grammatical, technical, and content clarifying suggestions, with some minor structure changes. See below for complete review with line-specific comments.*

*Lines 1-2: Your Abstract focuses heavily on uncertainty, but your Title does not reflect this; given the Journal’s guidelines, I would adjust either your Title or Abstract to more accurately reflect the other.*

Good point about consistency with regard to the uncertainty findings in the title and abstract. We have revised the title to include this finding.

*Lines 17-19: Revise sentence starting with “However”. It reads as a run-on and either needs to be split into two sentences, or needs to be shortened to remove verbosity.*

Ah yes. We fixed this sentence to avoid a run-on.

*Line 19: What is a “rich” dataset? You clarify this later in the manuscript (lines 83-85) but it is unclear here – it also does not connect to what the majority of your abstract is about (uncertainty).*

True, that’s not clear here. Given that we’re reaching the abstract word limit, we removed the term “rich” and clarify in the next sentence that this is a unique dataset, contributing to the novelty of this paper. We explain that it is unique because the expedition provided data for estimating carbon flux associated with fish, zooplankton, and sinking particles at the same time, location and depths (200 and 500 m). For example, a dataset with only fish biomass data that is depth-stratified would not be able to examine uncertainty in the relative contribution of fish to the biological carbon pump as well as a dataset that collected both fish, zooplankton, and passive sinking particle lux at the same time and place at the chosen flux boundaries.

*Line 20 (now line 22): You may need a transition sentence between “carbon pump pathways” sentence and “We calculate parameter uncertainty […]” i.e., related to previous line comment: How is uncertainty related to rich dataset?*

Given word limits for the abstract, we think that changing the order of these clauses will help to better tie in these two concepts. That is, the concept that a data-rich sampling effort is needed to investigate uncertainty, and the finding that, despite a unique dataset and high sampling effort, we still are left with fairly high uncertainty ranges around the mean for both absolute and relative fish carbon transport estimates.

*Lines 29-30: General comment for end of abstract: You spend a considerable amount of time in your discussion/conclusion discussing caveats considering sequestration depth vs. timescales, and what carbon is truly sequestered by fishes… I think this is a very important part of the paper – perhaps the most important part – that should be highlighted somewhere at the end of the abstract!*

We would have liked to spend more time highlighting this takeaway point too, which we also believe is relevant to discussions about carbon sequestration, carbon offsets and marine carbon dioxide removal. Given word limit constraints, we have briefly highlighted this point in lines 31-32 of the revised abstract.

*Lines 35-36 : You mention mesopelagic fishes dominate global biomass of fishes – what % (approximately) is this relative to total global fish biomass? This might be helpful for contextualizing the updated estimate.*

We agree that it would be helpful for readers to know the relative percentage of global fish biomass that could be mesopelagic fishes to help conceptualize just how high the revised estimates are. We have added this in lines 40-43.

*Lines 43-44 (now lines 49-50): You mention release of carbon via egestion of fecal matter, respiration of CO2, and mortality from predation. Consider rewording this sentence for clarity with respect to predation: Is carbon being “released” via predation in the form of a fish fall? Or is it being re-packaged in tissues of a predator, which is then egested, respired, etc.?*

We have changed the term “release” to “transport” so that this makes more sense for predation pathways. The latter of your descriptions is more how we define mortality--mortality flux includes both predation and fish falls. We have clarified that the mortality flux includes both of these pathways.

*Lines 43-44 (now lines 49-50): You mention egestion of fecal matter, CO2 respired, and predation mortality; you do not mention calcium carbonates which are brought up later in the manuscript; I would include here as well.*

Good idea. We added carbonates to this list.

*Line 48: “the magnitude of fish carbon transport” change to “fish-mediated carbon transport”*

Done.

*Line 50 (now line 56): “improved understanding of the role that mesopelagic fishes play is needed” reads as verbose; reword to be more streamlined if possible… perhaps simply rearranging the entire sentence (Starting with “Before the consequences…” Lines 49-50) to something like “Improved understanding of the role that mesopleagic fishes play is needed to…”*

That’s better! Done.

*Line 51: “the magnitude of carbon transport” change to “the magnitude of fish-mediated carbon transport” for clarity, since the following sentence talks about climate-relevant C sequestration and could be confused with C sequestration in general. “Fish-mediated” carbon transport is used elsewhere in the manuscript so I would add for unity as well.*

We agree with the suggestion to be clear and consistent with language and have added “fish carbon transport”. However, we tried changing all instances of “fish carbon transport” and “fish carbon flux” to “fish-mediated carbon transport”, and we found that this reduces readability due to the long noun phrase, especially for the more complex sentences in the manuscript. There are about 50 instances of these terms throughout so we want to be concise but also clear that we are using all of these terms interchangeably. Including all three terms (fish-mediated carbon transport, fish carbon transport, and fish carbon flux) will allow readers to understand that our use of these terms is comparable to elsewhere in the literature.

Thus, upon your suggestion, we now define our use of these terms in the Introduction line 89 of the revised draft with “fish-mediated carbon transport (hereafter also referred to as fish carbon transport or fish carbon flux)”, choosing this sentence because it’s not as long and complex as others earlier in the introduction. We believe this will help clarify terminology for readers while also enhancing readability throughout. We have even heard “fish flux” in scientific discussions of the topic, but chose to avoid this even shorter term due to its ambiguity.

*Line 52-53 (now line 60): Are there any references for 100 years being the typical horizon for climate policy? IPCC report or something similar? Unless it is mentioned in Siegel et al. 2021, then disregard.*

Yes, this is mentioned in Siegel et al., 2021 (“*leakage of sequestered carbon evaluated over typical planning horizons (50–100 years) is often the dominant fraction of the amount injected for depths <500 m”)*

*Line 58 (now line 66): This is the percentage I was looking for, I would move this into your introduction (see comment for Lines 35-36).*

We have edited and removed these percentage values to avoid repetition. The editing is because if global mesopelagic fish biomass is the updated minimum of 2 Gt, not the old estimate of 1 Gt, and other fish biomass estimates that generally exclude mesopelagic fishes estimate 1 Gt, then these percentages should be roughly 67-94%.

*Lines 61-62 (now line 71-72): While mesopelagic fishes perform DVM, and this active transport makes them prime candidates for effective C export and sequestration, I would mention that empirical rates are also lacking for epipelagic forage/large pelagic fishes, or all fishes in general, so this is still up in the air. Mesopelagic fish are disproportionally more studied/quantified than epipelagic or large pelagic fishes – see Saba et al. 2021 table on compiled fish carbon studies.*

Agreed, fixed!

*Line 72 (now 82) Clarify fish biomass to mesopelagic fish biomass.*

Done.

*Figure 1: I would include a distance scale in the inset if possible to help contextualize the distance/size of the tows.*

We like the suggestion to include a scale bar, but found this was hard to fit in the small inset and difficult to read the scale bar distance. It may also be confusing as to whether the scale bar refers to the inset or main map. Instead, we have described in the legend the vertical distance of the inset rectangle to provide readers with a sense of distance scale beyond just the latitude scale bar on the y-axis.

*Line 141* (now starting line 163)*: Under Fish identification, size distribution, and biomass section, I would clarify what you mean by lowest and highest taxonomic level possible. Lowest/highest meaning down to genus and/or species? Or lowest meaning lowest resolution (i.e., family, order…) What were the highest-level subsampled fishes used for/why was this subsample ID’d to the highest level possible? Clarifying what you mean by lowest/highest level would help with this interpretation.*

Good catch, thanks. We have now defined lowest as being the level as close as possible to species level, and have made sure that we are consistent with this terminology--the use of “highest” later in this paragraph was my mistake. I personally don’t find those terms (highest vs lowest) to be very intuitive myself so great idea to simply define what we mean there.

*Line 160 (now 189): Add a period after “shrinkage”.*

Done.

*Line 180 (now 209): Change “identified from MOCNESS-10 tows, this focus” to “identified from MOCNESS-10 tows, this focuses”*

We fixed this wording to clarify.

*Line 201 (now 238):* *Aereal biomass density is presented earlier when you explain C flux per aereal biomass density of fish per day, but is not defined until line 227. I would define this component earlier with the first equation, before simulated tows. I think this is especially important to clarify given it is also used in your simulated tows before you define it.*

We agree it would be good to briefly define areal biomass density as soon as it is mentioned. We have edited this paragraph accordingly to incorporate this suggestion.

*Line 215 (now 255): “Simulated tow” feels a bit under-explained, and could benefit from some clarity. Is this standard practice for determining uncertainty around tow catch biomass – do the data need to be assumed as normal to do this; are the data already normal, etc…. Is this used in other publications?*

Fair point! We have further explained why we simulate tows in subsequent sensitivity analyses from a true distribution estimated using empirical data. A normal distribution is a simple approach whose limitations (e.g. tends to have too few observations in the tails, compared to so, a Student’s T) are well known. We lack sufficient data to draw conclusions about the underlying probability distribution of tow caches.

*Line 255 (now 299): “[DOC] independent from egestion” – do you mean carbon leeching from fecal pellets? I would clarify this as those reading may not be intimately familiar with these mechanics, given this is an interdisciplinary*

*topic.*

Definitely worth explaining for a wider audience. We now provide examples of what we mean by fish DOC sources.

*Line 256-262: Excellent and pithy explanation of inclusion of specific dynamic action related respiration. I agree with this methodology, and really nice to see it reflected in the gut of the catch; your respiration component is solid.*

Thank you!

*Line 276-277 (now 295-298): Sentence starting with “We also consider…” could benefit from being reworded for improved clarity. Ending with “...zooplankton that may have otherwise migrated and released carbon above the flux boundary” would be sufficient.*

That sounds good. We have edited for clarity as suggested.

*Lines 278-280 (now line 323-325): This sentence is confusing; could benefit by clarifying “ingestion of energy needed to meet the sum of energy requirements and energy lost to respiration…” this makes it more of an equation, which is what the Kitchell equation is: conservation of mass balance. When you say “energy requirements”, do you mean for maintenance only? i.e., the fish is not growing (or starving)? Or do you mean maintenance plus a certain amount of growth? If the latter, how are you estimating growth? When you say “standard base energy budget for fishes”, you should clarify that this is a set of equations that need not be species-specific. I could see a situation in which this is misunderstood for a “mean” budget of all fishes or the like.*

That’s correct, that this is a Kitchell style bioenergetics approach where consumption is estimated from energetic needs. We like the suggestion to edit as “the sum of energy requirements” and then go on to explain in greater detail what these energy requirements are, and how we define these categories of energy loss. We also clarify a little further in this paragraph what is meant by standard base energy budget, both in terms of this being a set of equations and in terms of this being used to describe non-species specific energy flows for an individual fish (not a mean for all fishes).

Line 288 (now 338): *I would clarify that sinking particle flux is non fish- or zooplankton-mediated flux; fish also contribute to this sinking particle flux with fecal pellet egestion and it seems you account for egestion in your methodology, as well as in zooplankton flux component.*

Agreed. We have clarified our definitions and calculations in this paragraph.

*Lines 304-307 (now 365): The sentence starting with “The marginal carbon…” is confusing, consider rewording for clarity. The “to that which may otherwise be transported” is where I get thrown off. The comma after per capita can*

*be removed. What flux boundary are you selecting?*

We reworked this paragraph so that the zooplankton carbon flux calculations will be clearer to readers now.

*Lines 313-314 (now 378): Are there predation/non-predation mortality rates published that could tease this interaction between zooplankton and their predators apart? Do we know predation or normal mortality rates of zooplankton (by natural senescence) that could be used to separate non-mesopelagic fish mediated predation mortality from natural zooplankton senescence? If not, that’s OK… I understand observed rates in the mesopelagic are hard to come by, and this is otherwise a fair assumption!*

Rates distinguishing different types of mortality are not available for zooplankton to our knowledge.

*Line 359, Fig 4: A small suggestion, I would switch the colors. I would make 1 sq meter MOCNESS green and 10 sq meter MOCNESS blue, because it matches with the order they are presented in the graph legend – when you read the graphs left-to-right, the first color you encounter should be the first color in the legend.*

Good idea. Since the MOC-10 data appears sooner from left to right in Fig. 5, we have switched the order in the legend so that the MOC-10 (10 sq. meter mouth net) legend label appears first from top to bottom in the legend.

*Line 365: I don’t see an in-text reference for Figure 5 in the results section. I think it was meant to be referenced in Line 376 (now 478).*

Thank you for catching this! We have included reference to Fig. 5 in what is now line 479, and have also reworded this sentence for clarity.

*Line 453 (now line 571):* *When you say the 90th percentile range for bioenergetics or biomass uncertainty alone is “similar” – what is meant by similar? Visually similar? Is this alone sufficient enough to come to this conclusion, or is there a statistical test that can be done to confirm?*

By similar 90th percentile ranges, we mean that the minimum and maximum estimates (after excluding the most extreme estimates from the Monte Carlo simulation) are similar when considering either bioenergetic or biomass uncertainty. We have clarified this in the figure legend (line 574).

*Line 466-467 (now moved to lines 404-405): Excretion of PIC is not mentioned in your introduction when talking about fish carbon production; you should mention it so the reader has a clear understanding of all the components you are about to include in your calculations: POC egestion, PIC excretion, and DIC respired, and mortality pathways.*

Good point, as choices around which among these pathways are considered in fish carbon flux papers vary widely in the literature. Unfortunately we couldn’t include this list in the abstract as it brings us further past the word limit. We have included this in lines 107-109 in the introduction instead.

*Line 471-475 (now 535): “Lower range estimate” is used to describe both the 5th and 95th percentile from Monte Carlo simulations. Did you mean “higher range estimate” for the 95th percentile? Additionally, for clarity: the 10-50% estimate comes from the Monte Carlo simulation (percentiles), or is this a general estimate used to give a relative range?*

We did mean to say higher instead of lower there, in relation to the upper limit of carbon sequestration. We’ve added clarification about where the 10-50% comes from, which is an estimate based on an assumption that not 100% of carbon transported to either the 200 m or 500 m flux boundary in particulate form could be expected to sink to sequestration depths such as ~3000 m. Only at these depths of ~3000 m could we safely assume that 100% of carbon is sequestered for 100 years for this study site, based on results from Siegel et al., 2021 (Fig. S2F). The reason we cannot base this result solely on Siegel et al., 2021 is that the Siegel paper examines the fate of carbon at various injection depths, but the rate at which particulate carbon sinks from where it is released by these fishes (e.g., at a 200 m or 500 m flux boundary) is unknown.

Line 537 (now 705): *Are there other organisms that are well-known to predate on migrating zooplankton, to make your generalized pathway scenario a bit more specific?*

Yes, there is evidence for that in our dataset, which we have included in Appendix F. We added this reference here.

Discussion section general comment: *Overall comment on Discussion: This section is excellent, and I think these recommendations/commentary on sequestration timescales vs. depth of export is valuable. Like I said in the beginning of my review, I think you need to alter your abstract a bit to better reflect the hard work put into the full body of the paper. I think even a very brief, one or two word mention of recommendations based on the large uncertainty in bioenergetics/biomass parameters is worth putting in the abstract.*

Thanks so much for this feedback. We have done our best to incorporate some of this discussion content into the abstract.