We thank the two anonymous reviewers for their valuable and critical reading of the manuscript, which were useful in improving the scientific quality and clarity of the work. A point-wise response is given below, with the original reviewer comments in italics, and our answers in blue and labelled as “Response”.

Kind regards,
Kirsty Edgar and co-authors

RESPONSE TO REVIEWER 2

Overall comments:

I think that, overall, the manuscript is well structured and does a good job of gathering all the major ecological aspects of planktic foraminifera, but also, the current knowledge gaps. I consider it is a valuable contribution in the field of planktic foraminifera ecology/modelling as it summarizes the areas that need further research pretty well. Also, it highlights the trait-based approach as a useful tool to make reliable predictions for the impact of the environmental change on these organisms.

Concerning the language, it is well-written, however, some sentences are too long and complicated, making it hard to understand the message behind them. Also, I’ve noticed some technical writing mistakes such as double spaces, lacking commas and dots etc…

Response: Thank you for pointing this out. We will carefully review the manuscript to simplify overly long or complex sentences and correct any technical writing mistakes, as suggested.

I have mainly 3 concerns in regard to what I think the manuscript misses that should be an easy fix. I hope the following suggestions will help the authors to improve the manuscript.

- eDNA and eRNA analyses are mentioned in the: abstract, introduction and conclusion, however, there are nowhere to be found in the manuscript. I think these approaches should be given the same treatment as they are nowadays one the main ways we have to decipher some ecological aspects of the planktic foraminifera.

Response: We agree with this point and will expand this part in a revised and renamed section of the manuscript “*Suggestions for future research*” explaining what the technique is and its potential to contribute to the study of planktic foraminiferal traits. For now, most studies of eDNA on planktic foraminifera are largely confined to testing the approach for detecting diversity in ecosystems (e.g., *Barrenechea Angeles et al., 2020)* however, we will highlight the potential here. We would include new text along the lines of

“Environmental DNA (eDNA) metabarcoding, which analyses the genetic material present in the environment, such as sediment, or water, is a powerful new tool for identifying and monitoring biodiversity, biogeography and reconstructing ecosystems and ecologies (Ruppert et al., 2019). This technique can also provide insights into community composition over timescales spanning several hundreds of thousands of years or longer, improving our understanding of the relationships between biodiversity, environment and climate (e.g., Ambrecht et al., 2019). However, the bulk of eDNA foraminiferal studies to date have focussed on benthic foraminifera. For instance, this technique has significantly increased the diversity of organic walled and “naked” foraminifera that are rarely observed, have few morphological characters for traditional species delimitation, and don’t preserve well in the fossil record (Pawlowski et al., 2014). But eDNA holds great promise for investigating marine plankton, as it has the potential to overcome many of the data limitations that we currently face in this group with typically low standing stocks (de Vargas et al., 2015). It is a potentially

more effective means of detecting species presence in an environment than observations alone (Malviya et al., 2016; Ser-Giacomi et al., 2018; Barrenechea Angeles et al., 2020) but can also contribute much more broadly. For instance, providing insights into plankton population size (Andres et al., 2023), response to environmental change (Cao et al., 2022) or predator-prey dynamics (Ruppert et al., 2019). A combination of molecular and microscopic approaches can also yield new insights, e.g., in benthic foraminifera it allowed rapid determination of multiple different feeding strategies driving diversity and abundance in several foraminiferal taxa (Schweizer et al., 2022), a question which is typically restricted to analysis of feeding vacuoles and laboratory experiments. However, further method development is still required as some groups are not as well represented by eDNA technique as others, e.g., specimens may visually be present in sediments not found in the eDNA analysis, likely because of limitations of the primer to detect certain groups (Barrenechea Angeles et al., 2020, Hoshino and Inagaki, 2024)".

- The trait-based approach is pretty well detailed, and this manuscript presents it as a powerful tool for future predictions. Nevertheless, I feel that only the positive aspects of it are presented, not the limitations. In a synthesis paper I do not necessarily expect a discussion, but I feel that the limits of this approach, other than just the lack of quality input data, should at least be described/acknowledged. Also, it remains a model, which a simplification by its own nature. Therefore, the differences between models and in-situ observations should be better described.

Response: Thanks for pointing this out. We agree that acknowledging the limitations of the trait-based approach is critical. In the revised manuscript, we will discuss the limitations of models and their influence on modelling foraminiferal biomass stocks, where appropriate, including in the Introduction and as part of a new "*Suggestions for further research*" section. For instance, in the Introduction to also address similar comments by Reviewer 1, we will introduce text noting that

"While trait-based models have significant potential, they have inherent limitations. Like any models, they are a highly simplified version of natural systems. They may simplify complex ecological interactions and environmental influences, and the expression and importance of traits may vary by environment impacting generalisation. Additionally, trade-offs between traits are not always well understood or quantified, as for example, the absence or presence of symbionts in foraminifera. The eco-evolutionary model used by Gibbs et al. (2020), for instance, resulted in modelled trait evolution at rates orders of magnitude faster than observed in the fossil dataset, highlighting the need for caution when interpreting these model results. Despite these caveats, trait-based approaches present an exciting opportunity to leverage the exceptional fossil record of foraminifera to test our understanding of the modern range of environmental conditions and beyond, assess the universal applicability of traits and trade-offs through time and ultimately improve our understanding of evolutionary processes."

An example of addressing limitations of existing model-observation in Section 2.2.3 on Photosymbionts is here

"A recent study developing the ForamECOGEniE model to incorporate different symbiosis types (Ying et al., 2023) replicated patterns of asymbiotic and obligate symbiotic global distributions but underestimated the abundance of the non-spinose symbiont-facultative group particularly in the eastern equatorial Pacific. The challenge of modelling this group is the lack of information on what drives the symbiont-facultative group to acquire to lose symbionts. Without a clear ecological/physiological understanding, trade-offs cannot be incorporated in the model. Intriguingly, if the abundance of the obligate and facultative symbiotic taxa are combined there is a good fit between the model and observations (Ying et al., 2023). This may suggest that the groups overall exploit the same benefits from symbiosis. However,

determining the mechanistic underpinning is critical to understand the data, particularly the environmental/biological conditions under which symbiosis is active or not, and the benefits of the relationship to the host. Thus, culture experiments and further observation of this phenomenon are still required."

- Finally, it is stated numerous times that better datasets and experimental data is needed to improve our current knowledge of planktic foraminifera. However, the ideas and suggestions for improving them remain rather vague and, in that regard, how can the trait- based approach contribute to fill the missing data? Even after reading the manuscript a few times, one would struggle to come up with a strategy to overcome those gaps.

Response: We will expand on what types of new data are required to overcome the knowledge gaps and improve trait-based models in a new section called "*Suggestions for future research*". For example, for what is needed to move models forward we will detail, *"Therefore, key parameters needed to improve foraminiferal model performance include quantitative data of fundamental parameters such as: growth rate, respiration rate, half-saturation constant, and grazing preferences."* and for how models can directly contribute to constraining traits *"For modern species with high mortality rates in laboratory culture or difficulties maintaining multigenerational experiments (and high time and financial costs), trait-based models provide realistic setups to estimate potential physiological ecology in the absence of physical organisms (Grigoratou et al., 2019)."*

Considering the described comments, I think that this is a valuable and interesting contribution that should be considered for publication.

Response: Thank you!

Specific comments:

Lines 138-139. This sentence is odd. Larger sizes in the tropical regions and smaller in the equatorial region?

Response: Good spot. This should have read "equatorial upwelling" and will be amended.

Lines 172-174. This seems contradictory. Here, the effect of spines increasing the sinking rate is described as a benefit, however, in Table 2, negative buoyancy is described as a cost.

Response: The text should read that spines lead to a *decrease* (not increase) in sinking rate hence, further helping to counterbalance the negative buoyancy created by the shell in these taxa. Hence, it is considered a benefit in this regard.

*Lines 179-181**.** I think that this sentence should be re-written as it is correct, but not precise. Non-spinose species are omnivorous, they have the capacity of eating small prey such as phytoplankton, but they prefer an herbivorous diet (Hemleben et al., 1985). Also, the diet is known to change across the development of a given species. For example, O. universa shifts from herbivorous during its early and juvenile phase, to a carnivorous diet in the spherical adult stage (Schiebel and Hemleben, 2017).*

Response: We have summarised the dominant feeding preferences of the adult forms here, as we provide more details later in Section 2.2.1. However, we will include a brief clarification here, while aiming to avoid excessive repetition.

Lines 203-215. I feel that this paragraph misses the chance of describing 2 mechanisms that affect the calcification in an important manner: the presence or absence of photosynthetic *algae symbionts and the metabolic processes involved when forming new chambers during the individuals growth (Schiebel and Hemleben 2017; Takagi et al., 2020, 2021). The first process gives the species that host them a slight advantage as they modify the chemistry of the immediate water column, while the second mechanism is still under poorly constrained. However, I think that, as this is a review paper, these mechanisms should be described.*

Response: Good point. We will refer the reader to Section 2.2.3 on Photosymbiosis where we discuss the impact of photosymbionts on calcification. We will note in the text *"It is clear that specific traits within ecogroups, for example symbiosis (see Section 2.2.3), can positively impact the calcification response due to modification of the calcifying microenvironment and provision of an energy subsidy (e.g., Köhler-Rink and Kühl, 2005)."* We will also describe briefly the second metabolic mechanism that is poorly known but largely relates to enhanced respiration near the site of calcification that ultimately increases the total pool of inorganic carbon in the cell for calcification (e.g., Bentov *et al*., 2009 and Scheibel and Hemleben, 2017).

Lines 217-220. Too many concepts in one sentence makes it hard to read. Maybe separate?

Response: Thanks for flagging. We will split this sentence in two.

Lines 223-226. See previous comment, long and complex sentence. Maybe rephrase? Also, you consider this effects are "short-termed", however, the impact on the reproductive strategy, i.e. reduction of gamete/body size, is not that short term. Influencing the reproductive strategy of a species depends on a wide array of factors. I suggest rewriting this sentence in a more conservative approach.

Response: We will shorten and rephrase this sentence as suggested.

Lines 249-251. See previous comment surrounding spinose/non-spinose species feeding preferences. I think that the two paragraphs surrounding feeding strategy should be more homogenous.

Response: We will carefully review the content of both paragraphs as suggested and update the text to ensure better balance between the two feeding types.

Lines 261-262. This sentence lacks clarity, I would avoid mentioning "most zooplankton".

Response: We will remove reference to "*most zooplankton*".

Line 266. Considering the development of the paragraph, which is correct, I suggest to state that non-spinose species are omnivorous with a preference towards an herbivorous diet. By the way, that is what the reader could interpret by following the paragraph.

Response: We will update the text as suggested.

Lines 284-286. Again, long and complicated sentence. Maybe separate in 2?

Response: We will simplify and rephrase the sentence to *"Model simulation of trophic dynamics requires data on prey preferences, e.g., prey acceptance rates, protein acquisition from zooplankton versus phytoplankton, and average digestion and capture times."*

Line 340. Certain morphotypes of G. bulloides have recently been described as hosting bacterial symbionts (i.e. Synecocchus) (Bird et al., 2017). I would also correct in Table 2 when G. bulloides is described as hosting "none" symbionts.

Response: Bird et al. (2017) describe the identified cyanobacteria (*Synecocchus*) as endobionts in *G. bulloides* as the metabolic (if any) role of these are not yet known, i.e., are they really symbionts? They have also currently only been reported in one genetic type of *G. bulloides* IId and only from the eastern Pacific Ocean so it is unclear how widespread any association might be. However, we accept that it was an omission not to include any reference to this possibility in the main text and we will amend the main text in Section 2.2.3 on photosymbionts to include reference to the potential for a different type of symbiont, and particularly the need for future work to confirm the nature of the relationship. We will modify Table 2 to read “None*” and describe the possibility of symbiosis in the caption.

Line 365. Whole paragraph. See previous comment regarding the effect of photosymbionts on calcification. Further elaborating about it, as suggested earlier, in this paragraph could also be a possibility.

Response: This is a good point and we will develop the text on how symbioses may support calcification as requested, e.g., *“Algal photosymbionts also aid calcification by increasing the pH of the foraminifera’s immediate microenvironment above ambient seawater by utilising* CO_2 *during photosynthesis and therefore potentially enhancing calcification.”*

Technical comments

Response: We will make all of the suggested technical corrections in the final manuscript.

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