**AUTHOR RESPONSE**

**Reviewer 2**

**The manuscript entitled "The impact of essential climate variables on respiration rates in subpolar and polar planktonic foraminifera" by Armitage et al. reported the relationship between environmental parameters and respiration rates of polar and subpolar planktonic foraminifera species. Temperature effects were discussed in detail, and the authors showed that *Neogloboquadrina pachyderma*, a polar species that is often utilized for paleoenvironment reconstruction, has relatively stable respiration rates over a wide range of temperatures with low $Q_{10}$. This finding alleviates our concerns on potential respiration effect on foraminiferal test geochemistry with regard to *N. pachyderma*. They also conducted micro-Xray scanning to calculate biovolume more precisely, which allowed discussion on allometric scaling of respiration for generalization.**

**This study is important to gain our understanding of the basic metabolic activity of foraminifera under different temperature conditions, as well as ground the validity of species to be used in paleoenvironmental reconstruction. The manuscript is overall well-written, with detailed methods used and carefully discussed. However, I have several major concerns regarding the statistical treatment of the data and, in particular, the interpretation of the results. In several places, the analyses rely on limited datasets or assumptions that are not fully justified, and some conclusions appear to extend beyond what can be robustly supported by the data. I believe that addressing the points raised below—especially by reconsidering the statistical approaches and tempering some of the broader interpretations—would substantially strengthen the manuscript.**

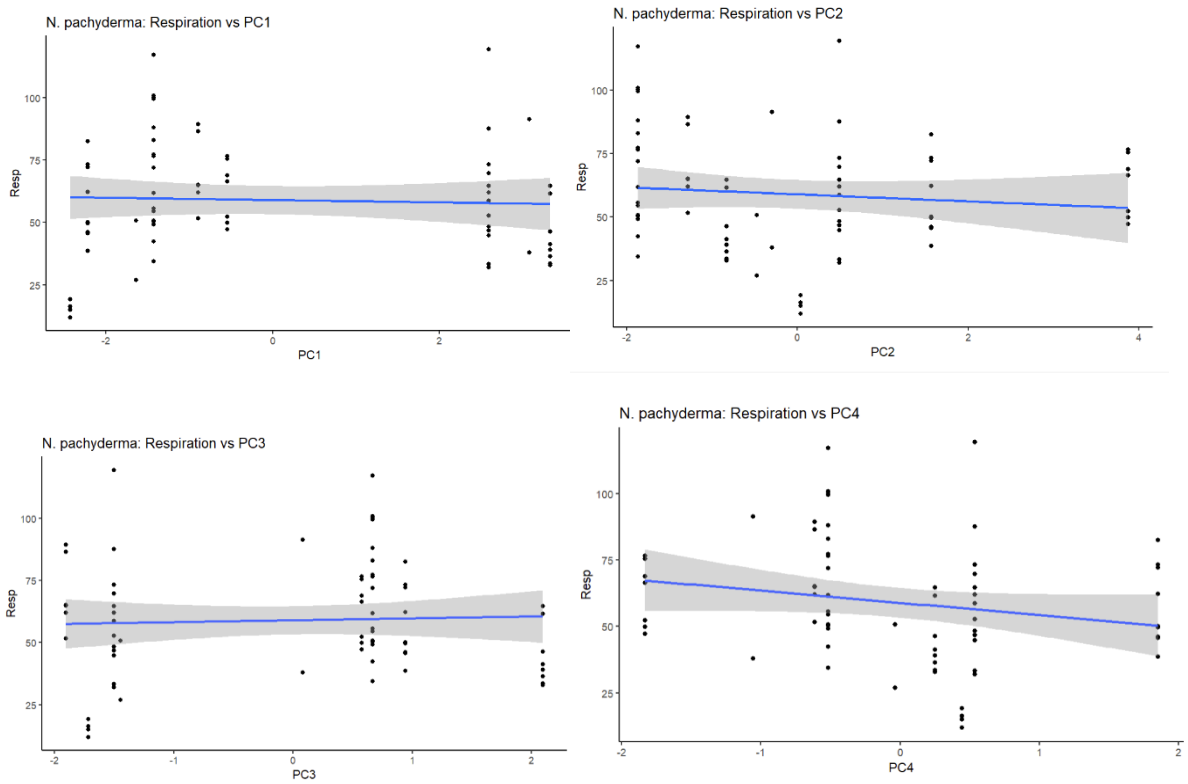
**AC:** We thank the reviewer for their positive assessment of the manuscript and for recognising the value of our study for understanding foraminiferal metabolic responses and their implications for paleoenvironmental reconstruction. We appreciate the constructive nature of the comments and fully acknowledge the concerns raised regarding statistical treatment and interpretation. We have carefully considered each point in detail and provide responses and planned revisions in the sections below. These changes will strengthen the manuscript and ensure that our interpretations remain well supported by the available data.

**RC1. Statistical analysis on respiration rates and other parameters**

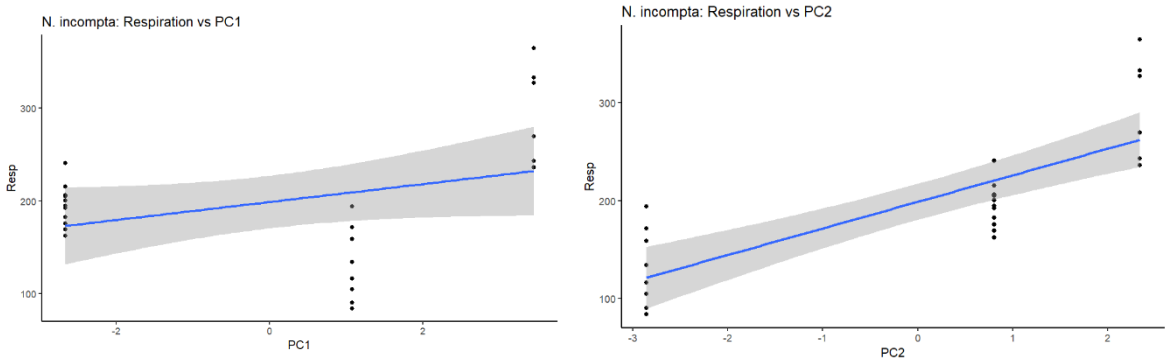
**The relationships between respiration rate and essential climate variables (ECVs) are evaluated primarily through separate pairwise correlation analyses (reporting $r^2$ and p values for each parameter, Table 3). While this approach may be useful as an exploratory analysis, it has important limitations that should be acknowledged. Many of the environmental variables considered (e.g., temperature, nutrients, salinity, DIC) are likely to be intercorrelated due to shared environmental gradients, such as water mass structure or seasonality. As a result, the reported correlations do not allow the independent effects of individual parameters on respiration to be disentangled. In addition, testing multiple environmental variables separately raises concerns about multiple comparisons, which may inflate the likelihood of detecting spurious significant relationships. I recommend either applying a multivariate framework (e.g., multiple regression or related approaches).**

**As I explain in the next part, correlation analysis for N. incompta needs to be reconsidered, since the datasets (based on 3 stations) cover a narrow range of each variable.**

**AC:** Following the reviewer's suggestion, we conducted a suite of multivariate analyses to evaluate whether respiration responds to combined environmental gradients. Principal component analysis revealed structured environmental axes in both species, with PC1–PC4 capturing 96% of environmental variance in *N. pachyderma* and PC1–PC2 capturing 100% of variance in N. incompta. Despite these clear multivariate gradients, respiration in N. pachyderma showed no relationship with any principal component (all $p > 0.39$), and scatterplots of respiration across PC1–PC4 space were insignificant (see Figures below). Multiple regression, partial least squares regression, and redundancy analysis all indicated that environmental variables explained approximately 0% of respiration variance (adjusted $R^2 \leq 0$; RMSEP ≈ raw SD; all confidence intervals overlapped zero; no improvement over null models). This demonstrates that respiration remained constant across the full multivariate environmental space sampled, supporting physiological robustness rather than masking of univariate effects by collinearity.




In contrast, *N. incompta* showed a strong multivariate environmental signal. Respiration was significantly related to the environmental gradients represented by PC1 and PC2, which together explained 75% of respiration variance (PC1 $p = 0.003$; PC2 $p < 0.0001$). This indicates that respiration in *N. incompta* is sensitive to integrated environmental structure, whereas *N. pachyderma* maintains a stable metabolic rate across environmental gradients. These contrasting responses highlight fundamental differences in metabolic plasticity between the two species.




In the revised manuscript we will include the multivariate analyses in the methods, results and discussion.

**RC2. For this species, respiration rates were measured at only three temperatures (10, 13, and 14 °C, in situ), covering a very narrow temperature range. In addition, the respiration rates recorded the highest at 13°C, and declined at 14°C. The calculation of Q10 based on the present data set appears problematic. Q10 assumes a monotonic, approximately exponential increase in metabolic rate over a sufficiently wide temperature range, under conditions where temperature is the primary limiting factor. Moreover, respiration peaked at 13 °C and declined at 14 °C, indicating a non-monotonic response and suggesting that the measurements may already span an optimal temperature or the onset of thermal stress. Under these conditions, the fundamental assumptions underlying Q10 are not met, and the resulting values are difficult to interpret physiologically. I therefore suggest either refraining from calculating Q10 or clearly stating that any estimated Q10 values are highly tentative and limited to a restricted temperature interval.**

**AC:** We agree that the narrow temperature interval and non-monotonic response limit the interpretability of the $Q_{10}$ estimate for *N. incompta*. We note however that the additional PCA analysis revealed a significant monotonic response of respiration to PC2 (e.g., representing temperature, salinity and alkalinity) which would support our interpretation and warrant calculation of Q10. Acknowledging the reviewers point though we will explicitly state that the $Q_{10}$ value is preliminary and needs to be confirmed over a larger environmental gradient.

**RC3. Spinose vs non-spinose interpretation.**

**The authors discuss the difference in $Q_{10}$ values between *N. pachyderma* and *T. quinqueloba* relating the morphology and trophic mode of the species. It is true that *T. quinqueloba* is a spinose species, but this species is a "short-spined" species that has completely different ecology and physiology from typical spinose-species like *Globigerinoides*, *Globigerina*, *Globigerinella*, *Orbulina*, etc. Specifically, *T. quinqueloba* is not a carnivorous species, nor a symbiont-bearing species, nor an oligotrophic-adapted species. Presence of spines is an adaptation for planktonic lifestyle, but since the non-spinose Neogloboquadrina species also share the shallow habitat as is presented in the sample metadata (Table 1), morphological difference (presence or absence of spines) is not meaningful to explain the $Q_{10}$ difference, I would say. As the authors noted at L443–445, it is true that non-spinose *N. incompta* showed relatively high Q10 (although it needs reconsideration as I pointed out above), which already collapses the validity of spinose/non-spinose comparison. I would say it's just species-specific.**

**AC:** We thank the reviewer for this valuable clarification. We agree that our previous interpretation placed undue emphasis on the spinose/non-spinose distinction and may not accurately reflect the ecology of *T. quinqueloba*. We will remove the morphology-based explanation and instead interpret the $Q_{10}$ differences as species-specific thermal responses. We will also note that the relatively high $Q_{10}$ in the non-spinose *N. incompta* supports this interpretation. A brief note on the two Arctic genotypes (Type IIa and Type IIb) of *T. quinqueloba* will be included for ecological context in section 4.1.

**RC4. Metabolic allometry and "crossover point". Representing the biovolume–respiration scaling relationship of planktonic foraminifera with data from other publications is interesting and potentially valuable. However, I don't fully understand the discussion on "crossover point" in L463–471. What exactly does the crossover point in Fig. 8 mean?**

**Moreover, I am concerned that the interpretation of the resulting scaling exponent may be overstated. The authors' statement that foraminiferal metabolism is somewhat "intermediate" between protists and metazoans more complex metazoans relies on cross-study comparisons that involve heterogeneous data sets, differing methodologies, and taxonomically broad groups. Given these uncertainties, the observed position of the foraminiferal scaling exponent relative to other organisms may reflect dataset composition or methodological differences rather than fundamental differences. I therefore suggest toning down this interpretation and framing it more explicitly as a hypothesis or conceptual possibility**

**AC:** We thank the reviewer for these helpful observations. We will clarify that the "crossover point" in Fig. 8 represents the intersection of fitted size-normalised respiration lines after temperature normalisation and does not imply a physiological threshold. We will also revise the interpretation of the metabolic scaling exponent and frame the comparison with protists and metazoans as a conceptual possibility rather than a firm conclusion, acknowledging dataset heterogeneity and methodological differences.

**RC5. Symbiotic ecology of *T. quinqueloba*.In Hemleben et al. (1989), it is indeed written that *T. quinqueloba* possesses symbionts, but no data are presented. Stangeew (2001) interpreted this species as symbiotic, based on the statement in Hemleben et al. (1989), which also does not show any evidence for the presence of symbionts on this species. Takagi et al. (2019) classified *T. quinqueloba* as a non-symbiotic species based on the absence of active chlorophyll fluorescence (photosynthetic activity). In this sense, "…their presence remains elusive (e.g., Takagi et al. 2019, ….)" is not appropriate. As far as I know, no positive data/evidence of the presence of symbionts for *T. quinqueloba* is available. Since the authors' observation is also in alignment with the absence of symbionts for *T. quinqueloba*, I think it's safe to say the specimens they used were non-symbiotic.**

**AC:** We thank the reviewer for this clarification. We agree that earlier references to symbionts in *T. quinqueloba* were unsupported and that more recent work (e.g., Takagi et al., 2019) demonstrates the absence of a systematic active chlorophyll fluorescence. Our own observations are consistent with a non-symbiotic ecology in the Arctic. We will revise the manuscript to clearly state that the specimens used in this study were non-symbiotic.

**RC6. Biovolume and empty final chambers. It is usually the case that the final chamber of collected foraminifera specimens is empty. In that case, biovolume estimation from the**

**whole test would cause overestimation, since the final chamber generally has the largest volume. In this study, was this point considered? Since the experiments were conducted at different time points from collection (within 24 hrs without food supply for CE23011, and fed specimens within 11 days for 2024 samples), specimens conditions might have been different. Ideally, filled or not needs to be checked, and the biovolume needs to be corrected by excluding the empty chambers. If this is not possible, at least, please make remarks on the cytoplasm volume, that it is not always equal to the cavity volume. In Burke et al. (2025), 75% of cavity volume was applied as biovolume. This is an alternative way to take into account the void part.**

**AC:** We thank the reviewer for raising this important point. In the revised manuscript, we will define the term "maximum biovolume" for clarity. We will state that *maximum biovolume* is the internal cavity volume bounded by the calcite test, representing the space occupied by living cytoplasm (including vacuolated cytoplasm and cytoplasmic linings) in asymbiotic planktonic foraminifera. While cytoplasmic density may vary within chambers, this metric captures the total living volume rather than carbon-equivalent biomass. Because cytoplasmic occupancy may vary during ontogeny, all volumetric estimates should be interpreted as upper bounds rather than instantaneous living biomass. We would also like to note here that the majority of specimens analysed here exhibited full chambers prior to measurements, as is common for asymbiotic species that tend to line all chambers more evenly. It is correct that symbiont-bearing species tend to have a cytoplasmic distribution that is heterogeneous, often leaving one or more chambers largely unoccupied, especially older chambers or those poorly illuminated.

When we compare our results to previously published datasets (e.g., Burke et al. 2025) we account for the 75% of the calculated cavity volume as biovolume. This ensured a like-for-like comparison across studies. We will clarify this distinction in the revised manuscript.

**Minor Comments**

**RC: Hemleben et al. (1989) and Stangeew (2001) citation corrections.**

**AC:** These corrections will be made.

**RC: Fig. 2 The illustration of the Unisense logos are confusing. Since it resembles to planktonic forams (maybe the logo derives from forams), I thought, at first glance, the specimens are located in those boxes. Please delete the logo. In addition, the cable of the "calibration chamber" is not connected anywhere. Is this correct?**

**AC:** We will remove the Unisense logos and correct the calibration chamber illustration in the revised figure.

**RC: Fig. 5 Why is the y-axis for panel (a) (Temperature) alone on a log scale whereas the others are in linear scale?**

**AC:** We appreciate the reviewer's comment. Temperature is well established to scale logarithmically with respiration rates (e.g., Burke et al., 2025; Lombard et al., 2009), whereas comparable empirical relationships do not exist for the other variables. We will clarify this in the revised manuscript.