

Response to Reviewer Comments Round 2 – Khan et al.

Thank you for the opportunity to respond to the review. This response letter is formatted to give each editor/reviewer comment (numbered) followed by a numbered response to each comment in blue and quotes from the main text in italics.

Comment #1 (Editor):

The reviewer was extremely pleased with your resubmission and only very minor corrections are needed before this can be accepted.

Well done.

Response #1:

Thank you, it is rare to get this sort of feedback in review and we really appreciate it. **All the suggested changes have been made.**

Reviewer comments:

Comment #2:

The Authors did excellent work and successfully addressed all my comments. I am satisfied with all the corrections. Changes made during the revision significantly improved the work. I also very much appreciate additional PERMANOVA tests conducted by the Authors to compare with the MGLM results.

I found added parts of the manuscript interesting and helpful for the readers.

During the second review, I found only a few technical or editorial errors. Therefore, I advise minor corrections to the manuscript, and I think that it can be considered for publication in the journal.

Response #2:

Thank you, it is rare to get this sort of feedback in review and we really appreciate it after the useful comments from the reviewer initially.

Comment #3:

Lines 29 and 76-81: I advise writing out the whole name of the listed statistical methods, e.g. permutational analysis of variance (PERMANOVA) when used for the first time in the manuscript. The same with MANOVA, LDA, nMDS, etc.

Response #3:

This has now been done. All first mentions of statistical methods are now listed in full. The relevant lines showing these changes are:

“...using distance-based methods (eg. permutational multivariate analysis of variance (PERMANOVA); ^{4,23,24}) or model-based methods which assume a gaussian error distribution (eg. multivariate analysis of variance or Linear discriminant analysis; ²⁵). Ecologists typically also use these distance-based methods to form ordination plots to visualise the multivariate groupings in a low-dimensional plot (e.g. non-metric multidimensional scaling (nMDS) plots or Canonical Discriminant Analysis; ^{26,27}).”

Comment #4:

Correct to “mean-variance relationships” through the manuscript.

Response #4:

This has been done. “mean variance” was changed to “mean-variance” 8 times.

Comment #5:

Line 147: please state clearly that the same groups of fish as in the previous study (reference 43).

Response #5:

This has now been clearly stated. The line reads:

“The present study was carried out with the dual aim of firstly, assessing variation in otolith chemistry and shape between the same groups *C. striata* in India as ⁴³ to test for further evidence of regional separation, and secondly, demonstrating the use of MGLMs with otolith chemistry and otolith shape data.”

Comment #6:

Line 235: less than 10% reported checking this assumption

Response #6:

This line has been changed and now reads: “revealed less than 10% of papers reported checking this assumption.”

Comment #7:

Lines 402-406: Please rewrite and possibly split this sentence to improve clarity.

Response #7:

This sentence has been split into two and reworded. It now reads:

“Mean-variance misspecification can also lead to confounding of dispersion and location effects in ordination plots³² (which we have verified in this study). This confounding can result in misleading or hard to interpret results when attempting to identify which response the effect is driven by or even a failure to detect multivariate effects unless it expressed in a high variance response”

Comment #8:

Line 429: delete “based”

Response #8:

The repeated “based” has been deleted. The line now reads:

“Both these ordination methods provide an alternative to traditional distance-based ordination methods which we have shown to be misleading by failing to account for mean-variance relations.”

Comment #9:

Figure 1: Indicate Y-axis title.

Response #9:

The y-axis title has been added. The plot now looks like this:

