by Isa Schon, 10 Jan 2023 16:33
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Minor revison

I would like to thank the authors for their extensive response to the reviewers' comments and the changes of the manuscript. Sorry that the evaluation of the revised version took some time – the xmas period is particularly difficult for the reviewing process.

Reviewer 3 found that your revised manuscript could be recommended now.

Reviewer 1 and 2 of the first round were unfortunately no longer available to reassess the revised version and I did not want to invite new reviewers for the second round of reviewing as this usually leads to problems and I consider as unfair. Instead, I have gone through the comments of reviewers 1 & 2 and your replies in the rebuttal myself.

While all comments have been addressed in the rebuttal letter, not all comments also led to changes in the revised manuscript. I would like to ask the authors to do so for the points specified below. This is especially relevant for the concerns on homology of mechanisms, which was raised by both reviewer 1 and reviewer 2. Likewise, for certain points, especially reviewer 1 asked for a more balanced discussion or interpretation, which I feel has not been addressed in the revised manuscript. In view of this, my editorial assessment is minor revision. I outline the reviewers' comments below, which I feel need still to be addressed:

Dear Dr., Schon,

Thank you very much for your comments on the manuscript. We submit the revised manuscript and provide a detailed point- by- point reply to the comments. We hope that we addressed all the comments suggested above, and the manuscript is now acceptable. Once our manuscript qualifies/acceptable for publication, we kindly like to request a PCI recommendation from the editor for submission to PCI friendly journals.

With best wishes,

Philipp H. Schiffer (For all authors)

Abstract, line 35:

1) I agree with reviewer 1 that the expression “genetic and biochemical programs” is not a good choice of words; please use instead the appropriate expression which you mention in the rebuttal letter (“combination of genetic and biochemical pathways that are upregulated upon preconditioning”).

We thank the reviewer for this suggestion. We modified the sentence in the abstract as recommended.

2) Phylogenetic placement and species description of reviewer 1:

1. No where in the manuscript is explained which species concept is applied to justify that the novel parthenogenetic Panagrolaimus strain is a new species. Please add this important information.

Thank you for the suggestion. We have now explicitly stated that we used the phylogenetic species concept (elevated to the genomic level by using many genes) in the text (Lines 141-16). We also made this clear in the sub-heading of the corresponding paragraph in Results (Line 165).

2. I still feel that the possibility that the basal position of the Siberian strain in the phylogeny could indicate that this strain is a hybrid parent is not sufficiently addressed, neither in the rebuttal nor in the revised manuscript. The newly added lines (178-182) do still not discuss this possibility. I would like to ask the author to provide the possibility of a hybrid parent at least as alternative explanation in the manuscript, even more so as the bootstrap support of the outside grouping is really low in Figure S3.

We thank the reviewer for this suggestion. As recommended, we now included the alternative explanation in the discussion now (Lines 270-273).

3. Please provide more information on the abbreviations now used in the new version – for example - what is GRAMPA (line 178)?

We now added the abbreviation for GRAMPA (Gene-tree Reconciliation Algorithm with MUL (Multi labelled)-trees for Polyploid Analysis) in the manuscript (Line 178).

Point 4) C. elegans dauer of reviewer 1:

While the new version of the text does now partly address this comment, the fact that larvae are not metabolically active is still not specifically mentioned. Not all readers will understand what "hypometabolic" means.

We thank the reviewer for mentioning this. We now included an explanation of hypometabolic state of dauer larvae (Lines 207-209).

Point 5) Panagrolaimus developmental stage of reviewer 1:

I still find the term "mixed population" confusing is this is normally not used to describe populations with different larval stages. The authors corrected this only in one place in the manuscript and I recommend to include the same corrections also everywhere else where this is mentioned.

Indeed, the term "mixed population" is confusing. We now replaced the term "mixed population" and corrected it to "mixture or populations of all larval stages and adults" (Lines 214, 216).

Point 6) Homology of mechanisms of reviewer 1 and point 2. of reviewer 2:

While indeed some suggestions for additional research have been added in the revised version of the manuscript in lines 301-303, these still do not include RNAi or inhibitor-based experiments nor is it clear from the text that this kind of experiment is planned for the future. I would like to recommend to the authors that they should add these specifics not only in the rebuttal but also in the manuscript.

I also cannot see how the "correlation is not causality" comment has been addressed. This is not the case in the mentioned lines (190, 200-201).

We thank the reviewer for this comment. As recommended, we added a new paragraph in the discussion providing all the details as we mentioned in the rebuttal (Lines 297-307). We now modified lines 191, 203-04 to mention that *Panagrolaimus n. sp* might utilize partially similar mechanisms to survive cryptobiosis, as our conclusion was based on the biochemical evidence of trehalose accumulation. We now addressed the "correlation is not causality" comment by mentioning that the presence of homologous genes in two species does not necessarily demonstrate their functionality in both.

Comment on line 132 of reviewer 1:

While the authors explained in the rebuttal that the strains were grown for multiple generations in several labs and that the strain was no longer frozen when received, none of this information made it into the manuscript although this is absolutely vital information and absolutely needs to be added.
We thank the reviewer for raising this point. We now included this information in the genomic DNA isolation methods part of the manuscript (Lines 334-337).

Suggestion of reviewer 1 to remove lines 241-243 and 244-251:
I agree with the reviewer that these lines would better fit the introduction and found the reply by the authors not convincing.
As suggested by the reviewer, we now removed the lines 241-43 and moved the lines 244-51 to introduction.

Sorry for asking this again but I would recommend another careful language check editing of the new parts of the manuscript as lines 178-182. Also for example in line 270, “fine” is not a suitable word.
We appreciate the recommendation and apologize for the colloquial phrasing in certain parts of the manuscript. We now had a native speaker carefully proofread our manuscript.