

09<sup>th</sup> May 2023

Dear Editor,

You notified us on January 2, 2023 that we were encouraged to review and resubmit a revised version of our manuscript entitled “Distinct impacts of resource restriction and warming on growth, reproduction and survival”.

We thank the editor for its constructive comments and we are sorry for the delay in resubmitting the manuscript. To investigate how the effects of temperature and food restriction on the measured life history traits influence population fitness (i.e. population growth rate), we used our empirical measurement in an Integral Projection Model. It took us more time than expected to compute the model and analyse its predictions. However, we believe that this adds novelty to our work and also helps discussing the adaptive values of phenotypic responses to warming and food (see below and main text for details). In addition, we believe that the revised version satisfactorily addresses most of those comments (see our detailed point-by-point explanations below) and more concisely explains our main findings, which should be of a broad interest for the *PCI Ecology* readership. In addition to addressing the points made by the editor, we have also made changes to improve the flow and clarity of the text without any impact on our main findings.

We look forward to hearing from you at your earliest convenience.

With kind regards,  
Simon Bazin on the behalf of all co-authors.

**Editor's comment:**

2-January-2023

The comments of Referee 1 can be summarized in the way that the reviewer lacks, firstly, a presentation of the research in a broader context and, secondly, a clear emphasis on what new the research brings to the subject. I appreciate the improvement made by the authors in the first of the indicated contexts. The authors have referred more extensively to the literature related to the subject. However, there is some insufficiency regarding the second point, namely the importance of the results presented in light of the research topic. The authors argue that the most important novelty resulting from the work is that the study was performed on a species that is large and is a predatory vertebrate. I am not convinced that this is a sufficient argument, because in that case one could perform similar studies for each species separately and use the same argument accordingly. Then, all knowledge would be limited to "case studies". What I miss is putting the results interpretation in the right context. I have a suggestion, which the authors could use at their discretion. The presented results could be interpreted in the context of the complexity of body size control in different organisms. To date, similar studies have been done on rotifers, copepods, insects and others. By comparing the results based on organism-specific body size control system during the lifecycle, one can make attempts to summarize the similarities and differences found so far. This topic is actually a very clear and important gap in research on the TSR, raised in discussion in many papers while it is just touched in the

presented manuscript (actually, in the last sentence of Abstract). Some other ways of interpretation are of course possible.

Our response: we thank the editor for the detailed review of our manuscript. We agree that it would be very relevant to compare our results to previous studies on other taxa to determine if a general response pattern emerge. Accordingly, we have provided a supplementary table where we summarize the findings of previous studies investigating the effects of food restriction and temperature on size at maturity and/or adult (asymptotic) size. We did not perform a meta-analyse as there are too few studies on the topic to perform robust statistical analyses. Instead we refer to the table in the discussion and discuss how our results differ from (or agree with) previous findings (lines 392 - 403).

These previous studies measured the effects of temperature and food on life history traits, but we do not know how the combined effects on traits translate to the mean fitness of individuals. We used an innovative approach of implementing the experimentally measured traits into Integral Projection Models (IPMs) to determine how combinations of traits influence generation time and mean fitness, thereby providing a better understanding of trade-offs between traits. On the other hand, we decided to keep our novelty argument that the study was performed on a species that is large and is a predatory vertebrate. Vertebrates differ from invertebrate in many ways (phylogeny, morphology, physiology) and it is not straightforward that previous results on invertebrate could be translate to a vertebrate species. As such our study is the first to provide information on how the combined effects of temperature and food on life-history traits translate to population mean fitness in a vertebrate species, and thus this constitutes a novelty. We thus decided to keep this argument even if we now have another type of original contribution with the addition of the supplementary table for the across taxa comparison (as suggested by the editor).

Summarizing the comments of Referee 2, the major points referred to discussing the results in the wider evolutionary perspective, e.g., proximate vs. ultimate mechanisms, other limiting factors, size at maturity vs. asymptotic size. The authors made attempts to address these points, but I found them insufficient. The least satisfactory seems to be a reference to size at maturity vs. asymptotic size. I suggest to refer to existing literature in this regard (it is indeed limited, but some information is available). In the detailed comments below I aimed to clarify some issues to facilitate the interpretation of data in the more evolutionary context. Referee 2 made also some comments regarding statistical issues and those I found sufficiently addressed.

Our response: According to Atkinson (1994), TSR refers to body size at a given developmental stage. We clarified in the text when we refer to size at maturity, adult size, or asymptotic size (lines 70, 108, 318-322, 391-403, 446).

We combined the experimentally measured traits into IPMs. These models allow us to calculate the mean fitness of individuals, which allowed us to interpret our results in a broader evolutionary context (lines 457 - 478).

**Editor**

L. 30-31 - “food restriction appears to amplify TSR by decreasing initial growth rate in the cold treatment” – as invoked by Referee 2, the strength of TSR generally refers to changes in body size and not in growth rate.

Our response: we removed this sentence.

**Editor**

L. 34 – though I strongly applaud referring the results to fitness measures, I am not convinced that the reference to “live fast die young” is appropriate/necessary in the case of this study. In the context of response to higher temperature it is quite trivial, because temperature accelerates the biochemical reactions in living organisms and such a result is the only expected.

Our response: as suggested, we removed the text referring to the “live fast die young” strategy.

**Editor**

L. 56-57 – there are also some, though indeed limited, studies on ultimate mechanisms, like the one by Walczynska et al. 2015 (the one suggested by Referee 2; 10.1016/j.jtherbio.2014.11.002).

Our response: as suggested, we included this information and added references about studies on ultimate mechanisms (lines 54 - 59).

**Editor**

L. 63-65 – the TSR is weaker in terrestrial systems but it does not implicate that oxygen is not limiting there, but rather that there is quite many other interfering factors, among which the most important seems to be seasonality (e.g., Verberk et al. 2021).

Our response: modified as suggested (lines 62 - 66).

**Editor**

L – 65-66 – “At the individual level, body size shift can be explained by the impact of temperature on the growth of ectotherms” – I do not understand this sentence. It is either trivial, or an unwanted shortcut.

Our response: we merged this sentence with the following to clarify that TSR can explain the individual body size shifts.

**Editor**

L. 68 – as invoked by Referee 2, originally TSR was referred to size at maturity and not to asymptotic size.

Our response: According to Atkinson (1994), TSR is referred to body size at a given developmental stage. We clarified this in the text.

**Editor**

L. 70-72 – to put it simple, ultimate factors refer to fitness, namely, to evolutionary meaning of the studied phenomenon. The current sentence is too complicated and not exactly to the point.

Our response: modified as suggested (lines 56 - 57).

**Editor**

L. 74-76 – again, TSR refers to body size response, which is of course accompanied by the whole growth and development pattern, but body size remains in the centre of interest.

Our response: we clarified that TSR is about body size shifts and could be the result of different developmental strategies.

**Editor**

L. 80-83 – I do not understand the reasoning presented in this sentence.

Our response: we clarified this sentence by specifying that we are referring to the links between growth trajectories and fitness related traits.

**Editor**

L. 108-122 – This paragraph should be removed from Introduction, because the consequences of the examined effect for the food web interactions were not the aim of the study. Definitely, it is still an important point for Discussion section.

Our response: as suggested, we removed this paragraph from the Introduction, but we discuss about food alteration via the indirect effects of temperature on the phenology and geographic changes and its consequences for food web interactions in the Discussion (lines 479 - 488).

**Editor**

L. 124-129 – Such effects were also studied in rotifers (Kielbasa et al. 2014; 10.1002/ece3.1292) and in diatoms (Walczyńska and Sobczyk 2017; 10.1002/ece3.3263).

Our response: as suggested, we added these references.

**Editor**

L. 142-146 – In this sentence there is a mixture of what was actually addressed in this study and what could be interpreted out of the results.

Our response: we removed this sentence since we also removed the paragraph to which it referred.

**Editor**

L. 150-155 – these lengthy sentences should be shortened to one precise message.

Our response: modified as suggested.

**Editor**

L. 147-160 – again, a mixture of goals and interpretations, while in this section the sound, reliable hypotheses are expected.

Our response: as suggested, we clarified our hypothesis (lines 134 - 138) and removed the speculative part about the potential implications of our results in the context of global change.

**Editor**

L. 171-174 – Referee 1 raised an important point about the thermal range investigated. I found the authors' response to that comment insufficient. First of all, if the optimal temperature for the species is 25 °C, then 30 °C in a thermal treatment is above the optimum and should rather be considered a suboptimal temperature. I totally understand the logistical limitations which sometimes dictate the choice of treatments in the planned study, but the most important then

is to correctly interpret the data. In this particular case, the difference between the two thermal regimes is that at 20 °C the response is assumed to be fully plastic, because it is within the 'optimal thermal range' for the TSR (Walczyńska et al. 2016; 10.1016/j.jtherbio.2016.06.006, the work suggested by Referee 1), while at 30 °C, being above this range, some compensatory/alternative physiological mechanisms are expected to be launched. Actually, such a distinction makes the interpretation of the patterns found at each temperature much easier.

Our response: in Walczyńska *et al.* (2016), the species-specific 'optimal thermal range' for TSR was defined as the range between the temperature at which the population growth rate becomes positive, and the temperature at which the population growth rate is maximal. Outside this thermal range, the TSR pattern may no longer be observed, although according to the results of Walczyńska and its collaborators, the TSR can be maintained for temperatures slightly above the optimal temperature. Yamamoto (1975) and Hirshfield (1980) reported that the optimal temperature for medaka reproduction is 27 °C, suggesting that the population growth rate is maximal at this temperature. Moreover, we showed with IPMs that the asymptotic per capita population growth rate is higher at 30 °C than at 20 °C, suggesting that our experimental temperature range is non-stressful for this species.

For these reasons, we believe that our experimental temperatures fall within the 'optimal thermal range' for TSR and that our results are not the product of a response to thermal stress. We clarified this in Material and methods (lines 181 - 190).

#### **Editor**

L. 192-193 – Does it mean the highest pre-maturity mortality in this regime?

Our response: we have had high mortality from unnatural causes in one aquarium for this treatment. We pooled the remaining fish in the two other tanks. This does not correspond to a natural pre-maturity mortality.

#### **Editor**

L. 273-279 – I find this interpretation of Fig. 2 quite misleading. In reference to the theory presented in Fig. 1, the comparison of crossed vs. nested patterns should be made for the pairs ad\_20 vs. ad\_30 and res\_20 vs. res\_30, because this distinction is expected for comparison across temperature. Then, the effect of food should be compared for the patterns found for both ads vs. both reses.

Our response: as suggested, we clarified the interpretation of Fig. 2 (lines 318 - 325).

#### **Editor**

Figure 2 – age at maturity could be additionally displayed in the figure for comparison.

Our response: as suggested, we added vertical bars to represent the age of the first sexually mature fish in each treatment.

#### **Editor**

L. 302-306 – If I am correct, survival was estimated starting from the age at maturity. In that case, it should be stated clearly, here and throughout the manuscript, and I suggest considering the change of the name of this trait to 'life expectancy since maturity'. Otherwise it is misleading, especially that, referring to my comment above, the highest pre-maturity mortality was found in the res\_20 regime.

Our response: survival was indeed estimated starting from the age of the first observed fish sexually mature (i.e. 60 days). We decided to keep the name of this trait as “survival” because we think it better represents the measured trait, and it is now clearly stated in Material and methods (lines 201 – 202) that survival was monitored daily from 60 days.

#### **Editor**

L. 324-331 – this paragraph resembles very much the one presented in Introduction. Please, choose one part of the text, the one in which it will be more suitable. Also, the reference to stronger TSR response with larger size according to Forster et al. 2012 is generally correct, but it is not an argument in this case. Forster et al. 2012 compared species which are close on a phylogenetic tree. For an obvious reason, it would not be correct to expect that fish species have stronger TSR pattern than, e.g., rotifer species, just because they are larger.

Our response: as suggested, we chosen the more suitable part of this paragraph to shorten it. We now have an additional novelty with the addition of IPMs, which allows us to study the influence of trait combinations on population dynamics. Furthermore, we decided to keep our novelty argument that the study was performed on a species that is large and is a predatory vertebrate for the reasons presented above.

#### **Editor**

Discussion in general – perhaps there are different “schools” in this regard, but I was trained that discussion should start from presentation of the most important findings from the study in the wider perspective. The speculative part should be following this introductory one.

Our response: as suggested, we restructured the discussion by first discussing our most important findings after a short reminder of the context, and then we finished the discussion on the more speculative part.

#### **Editor**

L. 339-354 – This section should be much more concise, as it is speculative and does not refer directly to the main aim of the study. I appreciate the discussion on the consequences of thermally-induced body size changes in a predatory species for the whole community or a trophic web, but this should be much shorter.

Our response: as suggested, we shortened the discussion on the consequences of temperature-induced body size shifts for the whole community or a food web, and merged this paragraph with the last one to avoid repetition, and to clarify the discussion on where and how these results could be implemented (lines 479 - 500).

#### **Editor**

L. 364-368 – Which curves did the authors mean to be nested in this sentence? Please, see my comment above about comparing the crossed vs. nested curves. It is not clear what the authors found surprising.

Our response: we were referring to the growth curve at 20 °C for restricted fish that appears to be more nested (i.e., below the curves for non-restricted fish) than at 30 °C. We clarified this in the text (lines 403 - 405).

#### **Editor**

L. 383-400 – This part is far too long. Information is just repeated using slightly different words. Referring to the oxygen as a limiting factor, one of the most important differences

between oxygen and food availabilities is that the former generally decreases with increasing temperature (at least relatively to demands), while the latter generally increases with increasing temperature (in ecologically relevant conditions). Walczyńska and Sobczyk (2017; 10.1002/ece3.3263) discuss the differences in thermally driven nutrition vs. thermally driven oxygen effects on plastic body size response.

Our response: as suggested, we shortened this part and we shortly discuss about the differences in thermally driven nutrition vs. thermally driven oxygen effects on plastic body size response (lines 416 - 423).

#### **Editor**

L. 420-429 – This paragraph is highly speculative and not really understandable. Instead, one would expect a joint discussion of the experimental effects on different life history traits, namely size at maturity, age at maturity, fecundity, clutch size and survival (life expectancy since maturity?). What about the trade-offs, which are mentioned in Introduction?

Our response: we added IPMs to study the impacts of temperature and food restriction on traits (growth, size at maturity, age at maturity, fecundity, and survival) and how these affect population growth rate and generation time. Thus, we were able to quantify the impact of trade-offs between traits in a broader evolutionary perspective. We discuss the results of this model in Discussion in lines 457 - 478.

#### **Editor**

L. 431-432 – Again, TSR refers to body size and not to growth rate.

Our response: see response above.

#### **Editor**

L. 447-449 – This assumption should be definitely more elaborated. My feeling while reading this manuscript was that the food-restriction applied in the study was not a real restriction. The authors should provide some references in which such a way of feeding was proven to be a restriction (through, for example, slower growth rate or smaller final size). Shortly, why this particular way of food regimes distinction was chosen?

Our response: based on our observations, the digestion time of a meal for a medaka is about 7 hours. By feeding the fish 1 day out of 2, we caused them to fast for a minimum of 24 hours. At 20°C, this significantly influenced fish survival, as well as fecundity and growth. This show that our food restriction treatment as thus consequences. If our feeding method had no impacts, we would have not observed any effect of the feeding treatments on the measured traits.

#### **Editor**

L. 472-490 – This paragraph is again too speculative and too wordy. Perhaps it should be matched with the very similar paragraph which was presented at the beginning of Discussion to provide a short, reliable and concise information on where and how these results could be implemented.

Our response: see response above.

## References

Atkinson, D. 1994. "Temperature and Organism Size: A Biological Law for Ectotherms?" *Temperature and Organism Size: A Biological Law for Ectotherms?* 25: 1–58.

Hirshfield, Michael F. 1980. "An Experimental Analysis of Reproductive Effort and Cost in the Japanese Medaka, *Oryzias Latipes*." *Ecology* 61 (2): 282–92.  
<https://doi.org/10.2307/1935187>.

Walczyńska, Aleksandra, Anna Kiełbasa, and Mateusz Sobczyk. 2016. "'Optimal Thermal Range' in Ectotherms: Defining Criteria for Tests of the Temperature-Size-Rule." *Journal of Thermal Biology* 60 (August): 41–48. <https://doi.org/10.1016/j.jtherbio.2016.06.006>.

Yamamoto, T. 1975. "Medaka (Killifish)." *Biology and Strains* 365.  
<https://cir.nii.ac.jp/crid/1574231874426383360>.