

Dear Christian,

Thank you for considering this manuscript for peer review at PCI Paleo, and for handling the review process. The reviewers had several good suggestions, and I have followed all of them with one exception for which there isn't yet sufficient data. Please see detailed responses to all reviewer comments below (also in blue).

Sincerely,

Melanie

**From the Recommender:**

Dear Melanie,

I wish you the best for 2020!

Sorry that it took so long. One reviewer wanted to carry out a review but was very busy, but then there was some sort of family problem and he did not find the time. And then X-mas came and I went offline.

Your manuscript is extremely interesting and I think it will revolutionize the understanding of trilobite morphogenesis.

I did not put my mark in "I recommend..." yet because there are some minor issues that were pointed out by the reviewers.

Please check out their comments and proceed as usual, returning your revised version.

I am optimistic that in the next round, your manuscript will be nearly perfect!

Best wishes, Christian

**Reviews**

*Reviewed by Kenneth De Baets, 2019-11-12 15:41*

This is a very nice contribution proposing the first growth model for trilobites. The manuscript also nicely illustrates how model parameters can be estimated from empirical data. More importantly, the model is used to test the relative impact of varying particular parameters on body size as well as assess how estimates differ when subsampling the representation of ontogenetic stage and/or specimen number per stage (or more generally). I would like to recommend it, but i had some minor points which I would like to see implemented in the final version.

The main points are:

1) Introduction: I agree that the Raup papers were major landmarks, but the method is not so well-suited to analyze growth (Urdu, 2015). In this context, it would be worth pointing out here that many studies have focused on invertebrates with accretionary growth and developed newer models which better capture particular growth parameters (Okamoto, 1988a, b; Ackerly, 1989; Urdu et al., 2010a, b).

Thank you for pointing this out; I did not intend to imply that Raup was the last or best word on the topic. Indeed, the list of papers that have been published since his work in the 1960s speaks to a later point in the paragraph, which is that accretionary growth models have seen much more work than discrete models (*sensu* McGhee, in reference to molting organisms, and not to be confused with the distinction between continuous vs discrete time-step models for accretionary growth). I have modified the text to reflect this and to acknowledge more explicitly the work that has been done since (p. 2). I found the Urdu papers to be particularly good summaries of this history, and much better than the previous citation; they are now included in the manuscript.

2) Additional figures. It is appropriate to discuss the different schedules in trilobite (hypoprotomeric, synanthromeric, proarthrous), but it might be worth considering to illustrate the differences graphically and how these would be implemented in the model. Also I feel it might help to have a schematic trilobite in various ontogenetic stages showing/illustrating the main parameters used in the model.

I am happy to provide additional figures illustrating these concepts. In the revised MS, the different schedules are shown in Figure 1, and the parameters are illustrated in Figure 2. Plots showing the empirical dataset used for parameter estimation (formerly Figure 1) is now Figure 3. The model output is now shown in Figure 4, followed by a new Figure 5 (see response to Reviewer 2 below). All subsequent figures have been re-numbered appropriately. The order for these figures has stayed the same.

If it is assumed that the transition from the meraspid growth gradient occurred at the onset of epimorphosis, then the different schedules are implemented via the  $R$  parameter in the model. This is a reasonable hypothesis but has yet to be tested. I have added brief discussion to the description of  $R$  (page 6).

3) One additional species: I understand that for showing and assess your model the dataset of *Aulocopleura koninckii* is the largest of its kind. However, I do feel that further testing your model would be aided by showing it also works for at least one additional species. This is probably problematic for disarticulated specimens, but is there no datasets suitable to at least try? Your model seems to be quite robust once a certain amount of specimens and ontogenetic stages are represented – at least in the case of *Aulocopleura koninckii*. This is at least alluded to in text (p. 6, line 6, “the growth rate for both species” – you are only analyzing one?).

While ontogenies for many species have been described, few are accompanied by complete measurements for multiple specimens. Further, the numbers of specimens figured in support of these descriptions are not sufficient for compiling enough data to be of use here. In short, to my

knowledge, there are currently no other suitable datasets to try. However, I am currently compiling a new empirical dataset to complement the *A. koninckii* dataset. I considered using it for this study (hence the “both species” reference left over from an early version of the MS, and now effectively a typo), but it needs to be published independently first (along with a complete description of the ontogeny) before it can be used here. More importantly, preliminary comparisons suggest that the growth parameters are very similar to *A. koninckii* (see for example, Hopkins MJ. 2019. Comparison of growth rates in the trilobites *Elrathia kingii* (Meek, 1890) and *Aulacopleura koninckii* (Barrande, 1846). 11<sup>th</sup> North American Paleontological Convention Program with Abstracts. *PaleoBios* 36 (Supplement 1): 174-175). This means that it would not add substantially to the current MS.

4) Meraspid growth gradient. I understand the choice to only fit curves to juvenile stages 9-17, but it might be worth considering to test what would happen with parameters if you would somehow include earlier stages (do they contain additional information or not?)

It is possible to run the analysis to include all specimens via the “Ter.min” argument in the `par.est` function (available in Github). Because this adds so few specimens ( $N = 11$ ) to the analysis (and none appear to be outliers in some capacity), the parameter estimates change very little (they all decrease slightly), and the model output is very similar, the primary differences being a slightly larger trilobite at molt 31 (29.2 mm instead of 27.5 mm with a slightly smaller cephalon relative to the trunk). I have added this information in the new Supplementary Figure 2.

5) Table 2: in my opinion it would also be worthwhile to tabulate the differences in variance in this or an additional table for the model and empirical data.

This is a good suggestion; it provides some framework for comparing the empirical values to the model values. I have added standard deviation and range to the second column in Table 2. In the process I discovered that I miscalculated the total body size for M9 and M14 (but not the individual sclerites). This only affected Table 2 and not any of the graphical output or discussion of the results.

6) Limit total number of moulds: you mention  $N < 31$ , but before you mention 20 for the holaspis stage (so maybe explicit add maximum to the  $N$  to avoid confusion with the median  $N$ )

With “ $N < 31$ ”, the Reviewer is referring to modifying the terminal number of molts (p. 11 or the revised MS). The earlier reference to  $N = 20$  for the holaspis stage is the terminal number of tergites used in the parameter estimation, not the total number of molts. The confusion here could have arisen simply from the fact that I neglected to italicize the “ $N$ ” on page 11, which would have indicated it was a model parameter (this is now fixed). However, this question has also drawn my attention to my (unfortunate) decision to use “ $N$ ” for a model parameter specifying the total number of molts (= the total number of time-steps in the model), while also using “ $N$ ” as a short-hand for sample size or number in other contexts. In order to further clarify, I have changed the model parameter to “*Molt*”, in the MS as well as in the R scripts posted on Github. I have also added text clarifying the use of “*Molt*” here vs in reference to a meraspid stage (e.g., “MO”, which represents the meraspid stage with one articulation (and zero thoracic tergites),

conveniently consistent with what is represented at time-step 0 of the model). See also new Figures 1 and 2.

7) Figure 3: I feel it might be worth to directly compare total body length both based on the model results with those measured in the empirical dataset.

This is a good suggestion, and I have added boxplots showing the variation in the empirical dataset for all meraspid and for holaspid specimens with 20 thoracic tergites. The direct comparison between the model output and the empirical dataset provides further support for statements already made about canalization, while also suggesting slowing growth rates late in the life. Additional discussion was added to the MS (p. 11), and the figure caption was modified.

These and additional comments can be found in the annotated pdf.

In response to additional comments in PDF:

Typo on page 11 of the original MS is now fixed

Regarding the comment on page 14 (p. 16 in revised MS) that comparisons of growth have been made in other groups (ammonites) from different environments: this is certainly true, which is why I specify this only for trilobites here. I should also add, however, that there has been documentation of intraspecific variation and geographic variation in trilobites, but nothing has focused explicitly on growth rates that I'm aware of. I have edited the sentence to reflect this.

Regarding the suggestion to show examples of meraspid vs holaspid individuals in figure 1B: the AMNH has some holaspid specimens of *Aulacopleura koninckii* in the collection, but no meraspid specimens. I have included an image of one of the holaspids (in the new Supplementary Figure 1) so that the reader can see what this trilobite looked like, and refer the reader to another paper where there are multiple good images of *A. koninckii* at all stages (p. 6 of revised MS).

Looking forward to seeing these implemented.

Kenneth De Baets

Suggested references:

Ackerly, S. C., 1989, Kinematics of Accretionary Shell Growth, with Examples from Brachiopods and Molluscs: *Paleobiology*, v. 15, no. 2, p. 147-164.

Okamoto, T., 1988a, Developmental Regulation and Morphological Saltation in the Heteromorph Ammonite *Nipponites*: *Paleobiology*, v. 14, no. 3, p. 272-286.

Okamoto, T., 1988b, Analysis of heteromorph ammonoids by differential geometry: *Palaeontology*, v. 31, no. 1, p. 35-52.

Urdu, S., 2015, Theoretical Modelling of the Molluscan Shell: What has been Learned From the Comparison Among Molluscan Taxa?, *Ammonoid Paleobiology: From anatomy to ecology*, Springer, p. 207-251.

Urdu, S., Goudemand, N., Bucher, H., and Chirat, R., 2010a, Allometries and the morphogenesis of the molluscan shell: A quantitative and theoretical model: *Journal of Experimental Zoology Part B: Molecular and Developmental Evolution*, v. 314 B, no. 4, p. 280-302.

Urdu, S., 2010b, Growth-dependent phenotypic variation of molluscan shells: Implications for allometric data interpretation: *Journal of Experimental Zoology Part B: Molecular and Developmental Evolution*, v. 314 B, no. 4, p. 303-326.

[Download the review \(PDF file\)](#)

*Reviewed by Lukas Laibl, 2019-11-30 18:20*

This manuscript describes and discuss a generative mode of trilobite growth, with respect to its segmentation and size changes, based on the Silurian species *Aulacpleura koninckii*. The results of the manuscript are, however, crucial to our understanding of trilobite body patterning and evolution, in particular to developmental modifications resulting in different trilobite morphotypes. The model generated here shows that nine (or twelve) parameters are needed to model trilobite growth and segmentation. This is important for several reasons. Firstly, it shows that strikingly different morphotypes of the trilobite body can result from changing only a few of these parameters and thus suggesting only simple developmental modifications might be responsible for the evolution of different morphotypes. Secondly, it shows that the trilobite growth is regulated by an array of independent factors and we should be careful when interpreting subjectively intuitive relationships in trilobite ontogeny (i.e. saying that big juveniles must result into big adults and small juveniles into small adults). Moreover, the methods and results have the potential to be useful for generating similar models for other hemianamorphic arthropods. For these reasons, I believe that the manuscript is worth a publication in a high profile journal as it is of interest to a broad readership.

Despite the high quality of the manuscript, I have two minor comments that I believe should be addressed prior to publication:

1) There are few simplifications of the trilobite development which I understand are here in order to generate a simple model. I, however, believe they should be discussed a bit more in the text. These comprise, for example, the statement that one tergite is released in one molt during meraspid period; ignoring largely the protaspid growth; or just briefly mentioning the variability in the holaspid segment no. of the model species.

As suggested, I have added discussion about variation in trilobite development that is not implemented in this model (p. 15 of revised MS). In addition, this comment inspired an additional figure showing the range in total body size if the model is run with the number of terminal segments varying from 17 to 22, as is seen in *A. koninckii*. This is described on page 10 and shown in the new Figure 5.

2) I understand this is a manuscript describing and discussing mostly the particular model. Some of the morphologies generated by changing relative timing of growth gradient or no. of tregites (figs. 4-7) are resembling morphologies of other trilobites, such as illaenids, phacopids, olenellids, etc. I think, that it is a bit pity not mentioning this a bit more in the manuscript in order to stress the importance of the presented model for trilobite evolution.

This is a good suggestion; indeed I have shown pictures of actual trilobite specimens for comparison to the model output when I give presentation on the model! I have added some discussion in the last section of the manuscript (page 16), and some images in the new Supplementary Figure 1.

Sincerely,

Lukas Laibl