Review file for "Decay experiments on shrimps provide insight into the fossilization potential of arthropod appendages" by Nora Corthésy, Gaëtan J.-M. Potin, Alexandre Torchet, Ian Quintas, Yu Wu, Sinéad Lynch, Allison C. Daley, Farid Saleh, published in *Open Palaeontology*.

This file contains the editorial and reviewer comments for the two rounds of review for this manuscript. Confidential comments to the editor and marked up/tracked changes documents are not included.

Round 1

Reviewer #1: Jane Reeves

In this paper, Corthésy *et al.* perform experiments on shrimp in freshwater and saltwater conditions to investigate decay patterns, reporting slower decay in salt water. The authors also find evidence to demonstrate that setae are a taphonomically robust character and use this result to tackle taxonomic questions in the literature. This paper presents some compelling findings that add to our overall understanding of the mechanics behind the patterns of preservation in the fossil record, something of interest to the field at large. It also includes details that would be of interest to arthropod workers more specifically. However, there is some over-interpretation in places, and I find that the data do not clearly support the main finding that shrimp decay faster in freshwater than in salt.

My last point arises from either an issue with the experimental design or the interpretation of the results, depending on your viewpoint. For the experiment, the shrimp species are only placed in one environment, and, with the design as it is, I would argue that this experiment is actually comparing the differences between species, not the salinity of the water, which is still an important finding. However, as there is no control of either the species or environmental variable, I don't see how it is possible to conclusively untangle if salinity or species-driven differences drive decay pattern variation (especially with the extra factor of size variation between both shrimps). As such, I don't think it's appropriate to say that decay/disarticulation happens faster in salt water because, with these data, the idea that species variation, or even an interaction of both factors, is the cause can't be rejected. I appreciate the animals are very similar and related shrimp, so the results are probably accurate. Still, I have an element of doubt about the salinity finding throughout the work, which unfortunately undermines the results and their subsequent applications. I acknowledge that the authors discuss this issue in the manuscript, but it does not fix this underlying problem for me.

While there is a case for requesting more lab work to collect data for both shrimp in both conditions or even with a different species more tolerant to a range of salinities, I don't think it is totally necessary. It may be something the authors consider for future projects. Ultimately, the question of the impact of salinity difference is only one aspect of this work, and even without it, the authors present an impactful piece of work. Quantifying differing decay patterns in marine and freshwater shrimp is highly useful for understanding the fossil record, as shown in the discussion considering the differences of preservation seen in fresh and saltwater fossil caridean. Similarly, identifying taphonomically robust characters is vital for the taxonomic determination of fossils.

I think adjusting and re-focusing the text is a more suitable approach. I suggest removing the comparison and analysis of salinity as an influencing factor to place more emphasis on the variation between the two species and morphological findings. I don't think this would be a substantial task; large sections of the work would be unaffected by this change, including the majority of the discussion, which contains some excellent examples that demonstrate how important this work is and could be in the future. I have also attached an annotated version of the work with additional specific comments, primarily around expansion of the experimental protocols, which are suitable (aside from the comparison of conditions concern) but would benefit from some additional detail.

If these changes were made, I would be very happy to support its publication

Reviewer #2: Silvina Slagter

This paper presents an exciting and well-documented dataset, incorporating a detailed taphonomic scale that includes breakage of appendages, detachment of appendages, and detachment of setae.

A few suggestions to further enhance the manuscript:

- As stated in the discussion, factors such as bacterial activity, pH, and oxygen concentrations can influence decay following the death of an organism. I suggest describing these parameters in the Methods section for the experiments performed.
- Please include a citation of the experiments by Slagter et al. (2024; DOI: 10.1111/gbi.12615) in the discussion when mentioning authigenic mineralization.
- It would be helpful to clarify why the experiments were terminated after 10 days. Was there complete decay, or were no further changes observed?
- How was the temperature maintained during the experiments? For instance, was a water bath used?
- In Fig. 4, no green boxplot (detachment of setae) is visible. It would be useful to mention this in the figure caption.

Editorial decision

Dear Farid Saleh

We have reached a decision regarding your submission, "Decay experiments on shrimps provide insight into the fossilization potential of arthropod appendages", to Open Palaeontology.

Our decision is to: Resubmit for Review.

Thank you for your submission and for your patience while we collected the reviews.

The reviewers have provided constructive comments on your submission. These reviews are available in the Review Files section of our submission platform in a file called **"Saleh Reviewer Comments.docx"** and are also appended to the bottom of this email.

Both reviewers are very excited by the manuscript and believe it is an important and timely contribution. However, both reviewers also feel that the manuscript requires revisions before it can be published. Their concerns fall into two main areas.

First, Reviewer #1 expresses concern about overinterpretation of the results due to limitations of the experimental setup. I agree with the reviewer that further experiments are not necessary, but the text should be modified as the reviewer suggests to focus less on the salinity and more on the specific and morphological findings. Note that Reviewer #1 has also supplied a marked-up copy of the manuscript as "Review_Arthropod+Appendage+v4.docx" with further inline comments.

The second concern is the replicability of the study. Reviewer #2 believes that much more detail about the experimental setup should be included within the manuscript to ensure that it is replicable, including information about ambient temperature and other abiotic variables. I agree that supplying these details would make for a stronger paper and would also make it easier for other researchers to followup on this study in the future.

Given these comments, we request that you make revisions to your manuscript and resubmit it for another round of review. When you submit a revised version of your manuscript, please provide:

- a revised editable version of the manuscript with tracked changes,
- a 'clean' version of the manuscript (with no tracked changes) as a pdf, and
- a point by point explanation of how you have addressed the reviewers' comments.

Please remember that we operate a transparent peer review process at *Open Palaeontology*. This means that we will publish unblinded peer review reports along with any accepted articles. Double-blinded reviews will be unblinded on acceptance and the review reports published as per our open review policy.

You will be able to resubmit you work for further consideration by logging in to the OJS platform or from this URL: https://www.openpalaeo.org/workflow/index/7073/3

We encourage preprinting of your work where possible. If you have not already done so but would like to post a preprint on the *Open Palaeontology* site or elsewhere we encourage you to do so, but please let us know.

Thank you again for your submission to *Open Palaeontology*. If you have any queries, please do not hesitate to contact us.

Best wishes

William Gearty

Response 1

Reviewer #1: Jane Reeves

In this paper, Corthésy *et al.* perform experiments on shrimp in freshwater and saltwater conditions to investigate decay patterns, reporting slower decay in saltwater. The authors also find evidence to demonstrate that setae are a taphonomically robust character and use this result to tackle taxonomic questions in the literature. This paper presents some compelling findings that add to our overall understanding of the mechanics behind the patterns of preservation in the fossil record, something of interest to the field at large. It also includes details that would be of interest to arthropod workers more specifically.

We would like to thank Reviewer 1 for their comments that helped us improve the manuscript.

However, there is some over-interpretation in places, and I find that the data do not clearly support the main finding that shrimp decay faster in freshwater than in salt. My last point arises from either an issue with the experimental design or the interpretation of the results, depending on your viewpoint. For the experiment, the shrimp species are only placed in one environment, and, with the design as it is, I would argue that this experiment is actually comparing the differences between species, not the salinity of the water, which is still an important finding. However, as there is no control of either the species or environmental variable, I don't see how it is possible to conclusively untangle if salinity or species-driven differences drive decay pattern variation (especially with the extra factor of size variation between both shrimps). As such, I don't think it's appropriate to say that decay/disarticulation happens faster in salt water because, with these data, the idea that species variation, or even an interaction of both factors, is the cause can't be rejected. I appreciate the animals are very similar and related shrimp, so the results are probably accurate. Still, I have an element of doubt about the salinity finding throughout the work, which unfortunately undermines the results and their subsequent applications. I acknowledge that the authors discuss this issue in the manuscript, but it does not fix this underlying problem for me. While there is a case for requesting more lab work to collect data for both shrimp in both conditions or even with a different species more tolerant to a range of salinities, I don't think it is totally necessary. It may be something the authors consider for future projects. Ultimately, the question of the impact of salinity difference is only one aspect of this work, and even without it, the authors present an impactful piece of work. Quantifying differing decay patterns in marine and freshwater shrimp is highly useful for understanding the fossil record, as shown in the discussion considering the differences of preservation seen in fresh and saltwater fossil caridean. Similarly, identifying taphonomically robust characters is vital for the taxonomic determination of fossils. I think adjusting and re-focusing the text is a more suitable approach. I suggest removing the comparison and analysis of salinity as an influencing factor to place more emphasis on the variation between the two species and morphological findings. I don't think this would be a substantial task; large sections of the work would be unaffected by this change, including the majority of the discussion, which contains some excellent examples that demonstrate how important this work is and could be in the future.

We agree with Reviewer 1 that the experiments were not specifically designed to address the impact of salt on degradation. It is therefore more important to emphasize that the observed differences can be explained by both the nature of the organic matter and possibly other environmental conditions. In response to Reviewer 1's recommendation, we have revised the discussion by reorganizing the text and adding elements of caution regarding the interpretation and significance of the results. The revised text now reads as follows: "The slower decay of saltwater shrimps compared to freshwater shrimps cannot be attributed to the different environmental conditions in which they

were placed, even though salt is known to be a natural preservative and can act as a conservation agent at high concentrations (Wijnker et al., 2006). It is even considered a catalyzer for the replication of non-biomineralized morphologies in authigenic minerals (Corthésy et al., 2025). The role of increased salinity in the fine-scale preservation of arthropod bodies is evident in sites like the Jurassic Solnhofen Lagerstätte in Germany, which preserved a large number of arthropods with their appendages attached, particularly shrimps (Pazinato et al., 2021; Winkler, 2014). Numerous taphonomic models and experiments developed to explain preservation in Solnhofen attribute the high degree of articulation in fossils to elevated salinity levels (Gäb et al., 2020; Gerschermann et al., 2021). However, salt is also known to increase the degradation rate of proteins under normal salinities (e.g., Corthésy et al., 2025). As such, the relationship between salinity and decay rates is not linear. For these reasons, and because the experiments were not designed to investigate the role of salinity in the decay process, we attribute the differences in decay rates between freshwater and marine shrimps to variations in their organic composition rather than to differences in salinity. For instance, considering that calcium is more limited in freshwater compared to saltwater, it is possible that freshwater shrimp appendages contain fewer biominerals and are, therefore, more prone to decay and disarticulation than marine shrimp appendages. Additionally, the thickness of the freshwater shrimp cuticle may be lower than that of marine shrimps, making the latter more resistant to degradation. Other factors, such as the quantity of polysaccharides in the cuticle, may also influence decay, as this material is not easily recyclable by bacteria (Corthésy et al., 2024). It is also worth noting that factors other than salinity and the nature of the organic matter also influence decay rates. These include pH, oxygen concentration among other parameters (Allison, 1988; Clements et al., 2017, 2022; Corthésy et al., 2024; Fraga & Vega, 2025; Hancy & Antcliffe, 2020; Mähler et al., 2023)."

I have also attached an annotated version of the work with additional specific comments, primarily around expansion of the experimental protocols, which are suitable (aside from the comparison of conditions concern) but would benefit from some additional detail.

These were considered.

If these changes were made, I would be very happy to support its publication

Reviewer #2: Silvina Slagter

This paper presents an exciting and well-documented dataset, incorporating a detailed taphonomic scale that includes breakage of appendages, detachment of appendages, and detachment of setae.

We would like to thank Reviewer 2 for their comments that helped us improve the manuscript.

A few suggestions to further enhance the manuscript:

- As stated in the discussion, factors such as bacterial activity, pH, and oxygen concentrations can influence decay following the death of an organism. I suggest describing these parameters in the Methods section for the experiments performed.

  The following text was added in the material and methods section: "*N. davidi* individuals were placed in sterilized polyester boxes of dimensions 6.5 x 4.5 x 2.3 cm, containing fully oxygenated reverse osmosis deionized freshwater and *P. varians* individuals were placed in boxes containing artificial seawater [fully oxygenated reverse osmosis deionized water with a salinity of 1.024 psu using Aquarium Systems Reef Crystals (Code 216029; www.aquariumsystems.eu)]. All boxes were filled with water to ~80% of their height. The

use of reverse osmosis water, which is mostly free of bacteria and has a neutral pH of 7, implies that most bacteria in the experiment originated from the shrimps themselves."

- Please include a citation of the experiments by Slagter et al. (2024; DOI: 10.1111/gbi.12615) in the discussion when mentioning authigenic mineralization.
  Done
- It would be helpful to clarify why the experiments were terminated after 10 days. Was there complete decay, or were no further changes observed?
  The following text was added for more context: "The time frame of ten days was chosen since some characters became difficult to monitor after 10 days, which is a typical timeframe for shrimp decay (Corthésy et al. 2024, 2025)."
- How was the temperature maintained during the experiments? For instance, was a water bath used?
  The air temperature in the room is highly regulated, as the lab contains both marine and freshwater aquaria.
- In Fig. 4, no green boxplot (detachment of setae) is visible. It would be useful to mention this in the figure caption.
  The following text was added to the caption: Note that no green boxplots representing setae detachment are shown in the figure, as the taphonomic scores for this character did not increase significantly enough to warrant representation in a boxplot.

References

Corthésy, N., Antcliffe, J.B. and Saleh, F., 2025. Taxon-specific redox conditions control fossilisation pathways. Nature Communications, 16(1), p.3993.

Corthésy, N., Saleh, F., Antcliffe, J.B. and Daley, A.C., 2025. Kaolinite induces rapid authigenic mineralisation in unburied shrimps. Communications Earth & Environment, 6(1), p.4.

Round 2

Reviewer #1: Jane Reeves

I thank the authors for their considered responses to my previous review. The edits they have made have addressed my comments well. The shift to focusing more on the differences between taxa, and the addition of a more conservative consideration of the impact of the environment, has fully addressed the main concern I had about overinterpretation of the results and experimental design. This is a well done, concise, piece of research that expands our knowledge of the complexities of the fossil record, which is vital for the palaeontological community at large, and provides important specific findings for arthropod workers. As such, it is of interest to a wide audience and I am very happy to support its publication.
Recommendation: Accept Submission

Reviewer #2: Silvina Slagter

This manuscript provides a valuable experimental baseline for evaluating the taphonomic stability of arthropod appendages and interpreting preservation patterns in the fossil record. The observation that detachment of setae exhibits strong resistance to decay in fresh and saltwater environments is particularly compelling.

However, the abstract suggests comparing freshwater and marine arthropod preservation, yet most fossil examples cited—and all radiodonts illustrated—are from marine environments. No direct evidence exists that any of the taxa discussed (e.g., Anomalocaris, Houcaris, Hurdia) lived in or were preserved in freshwater settings. While relevant to marine taphonomy, these examples do not adequately support conclusions about freshwater preservation potential. To address freshwater versus marine contrasts more effectively, it would be helpful to integrate examples of extinct freshwater arthropods, such as fossil carideans, in the discussion, or clearly frame the fossil discussion as marine-only. This clarification would strengthen the manuscript's logic and relevance to the stated aims. This study is well-executed and engaging, but I recommend some refinement.

A few suggestions to further enhance the manuscript:
In the methods section, I would replace "salinity of 1024 sg" with the water having a specific gravity (SG) of 1.024

Please also correct in the 2nd paragraph of methods "salinity of 1024 psu" and replace it with SG (note density vs. specific gravity)

In the discussion, replace catalyser by catalyst

Recommendation: Revisions Required

Editorial decision

Dear Farid Saleh

We have reached a decision regarding your submission, "Decay experiments on shrimps provide insight into the fossilization potential of arthropod appendages", to *Open Palaeontology*.

Our decision is to: Request Revisions.

Thank you for the work that you put into your revised submission and for your patience while we collected the reviews. The reviewer comments are available on the *Open Palaeontology* publication platform and are appended to the bottom of this email.

Reviewer #1 is now happy with the state of the manuscript, but Reviewer #2 has some lingering minor concerns (see below). Please address these remaining issues, especially the concern about representation of freshwater fossil examples in the discussion, and resubmit a revised version of your manuscript. I do not anticipate another round of review with be required.

When you submit a revised version of your manuscript, please provide:

- a revised editable version of the manuscript with tracked changes,
- a 'clean' version of the manuscript (with no tracked changes) **as a pdf with line numbers,**
- a point by point explanation of how you have addressed the reviewers' comments

Please remember that we operate a transparent peer review process at *Open Palaeontology*. This means that we will publish unblinded peer review reports along with any accepted articles.

You will be able to resubmit you work for further consideration by logging in to the OJS platform or from this URL: https://www.openpalaeo.org/workflow/index/7073/3

We encourage preprinting of your work where possible. If you have not already done so but would like to post a preprint of your article we encourage you to do so, but please let us know.

Thank you again for your submission to *Open Palaeontology*. If you have any queries, please do not hesitate to contact us.

Best wishes

William Gearty

Response 2

Reviewer #1: Jane Reeves

I thank the authors for their considered responses to my previous review. The edits they have made have addressed my comments well. The shift to focusing more on the differences between taxa, and the addition of a more conservative consideration of the impact of the environment, has fully addressed the main concern I had about overinterpretation of the results and experimental design. This is a well done, concise, piece of research that expands our knowledge of the complexities of the fossil record, which is vital for the palaeontological community at large, and provides important specific findings for arthropod workers. As such, it is of interest to a wide audience and I am very happy to support its publication.

Recommendation: Accept Submission

We would like to thank Reviewer 1 for their comments that helped us improve the manuscript.

Reviewer #2: Silvina Slagter

This manuscript provides a valuable experimental baseline for evaluating the taphonomic stability of arthropod appendages and interpreting preservation patterns in the fossil record. The observation that detachment of setae exhibits strong resistance to decay in fresh and saltwater environments is particularly compelling.

We would like to thank Reviewer 1 for their comments that helped us improve the manuscript.

However, the abstract suggests comparing freshwater and marine arthropod preservation, yet most fossil examples cited—and all radiodonts illustrated—are from marine environments. No direct evidence exists that any of the taxa discussed (e.g., Anomalocaris, Houcaris, Hurdia) lived in or were preserved in freshwater settings. While relevant to marine taphonomy, these examples do not adequately support conclusions about freshwater preservation potential. To address freshwater versus marine contrasts more effectively, it would be helpful to integrate examples of extinct freshwater arthropods, such as fossil carideans, in the discussion, or clearly frame the fossil discussion as marine-only. This clarification would strengthen the manuscript's logic and relevance to the stated aims. This study is well-executed and engaging, but I recommend some refinement.

We had included a discussion on this point in the previous version of the manuscript, particularly noting that in some sites where arthropods are found, there is evidence of freshwater input, such as the deltaic environment of the Chengjiang Biota. In fact, such freshwater influxes have been linked to mass mortality events in the Chengjiang Biota, during which organic matter could decay under non-saline conditions (Saleh et al. 2022, Nature Comm). In other words, despite the dominance of radiodonts in fully marine settings, they can decay under freshwater conditions (even if only briefly). We also acknowledge that the study could be better designed if we had precise information on the salinities of palaeo-oceans; unfortunately, such data are not available. For this reason, the following text was already included in the introduction:

“More importantly, although most arthropods from the Cambrian and Ordovician inhabited fully marine environments, some lived in settings influenced by freshwater input, such as deltas (Gaines,

2014; Gaines et al., 2024; Saleh et al., 2020c, 2022b). Since freshwater input in palaeodeltas cannot be easily quantified, we use purely freshwater and marine conditions to investigate if the degradation sequences are similar or significantly different between these environments."

The focus on radiodonts is the primary aim of this paper, which was clearly stated in the previous version of the manuscript:

"We also compare the degradation of appendages to patterns observed in the fossil record, with a particular focus on radiodonts since the morphological characteristics of their appendages are often used for taxonomic and palaeoecological purposes."

The discussion also included parts on fossil carideans; however, since radiodonts are mainly marine (albeit with some exceptions such as the Chengjiang Biota), and they are the main focus of the paper, more discussion is devoted to marine organisms.

"The detachment and breakage of appendages in both freshwater and saltwater experiment explain patterns observed in the fossil record (Fig. 4A, B). Although fossil carideans often preserve appendages both in marine water (Bravi et al., 1999; Garassino and Bravi, 2003; Winkler, 2013) and in freshwater (Barros et al., 2021; de Mazancourt et al., 2022; Feldmann et al., 1981; Garassino et al., 2002), most of them do not preserve all of their appendages. […]"

A few suggestions to further enhance the manuscript:

In the methods section, I would replace "salinity of 1024 sg" with the water having a specific gravity (SG) of 1.024

Done

Please also correct in the 2nd paragraph of methods "salinity of 1024 psu" and replace it with SG (note density vs. specific gravity)

Done

In the discussion, replace catalyser by catalyst

Done

Recommendation: Revisions Required