

Munich, October 15th, 2022

Dear Recommender,

We thank you very much for providing us this opportunity to submit a new version of our manuscript. We would like to thank the three reviewers for their extensive and detailed feedbacks. We have answered all their questions, taken into account most of their comments and emended accordingly the concerned sections. The changes are detailed in the present document and in the manuscript with track-changes.

In contrast, we provided detailed arguments when we disagreed with the reviewers. Accordingly, these elements remained unchanged in the revised version. We hope that you will find our rationale acceptable.

Independently, we have also corrected a few minor mistakes and typos. Significant edits are listed below:

- Table 1 has been edited to provide sample sizes for each method (microwear and hypoplasia) along with the rhinocerotid assemblage at each locality;
- Tables 2 and 3 and the associated text (Results section) have been modified to separate the effects by microwear facets (grinding and shearing).

Our specific answers to each reviewer are inserted below, in the text of their reviews, next page.

We hope that this revised version will now meet the standards required for a recommendation by PCI Paleontology.

Yours sincerely,

Manon Hullot and co-authors



Dr. Manon Hullot,  
Post-doctoral Fellow

## Response to the reviewers

### Reviewer #1 – Antigone Uzunidis

*Hullot et al. submitted a very interesting paper entitled "Spatio-temporal diversity of dietary preferences and stress sensibilities of early and middle Miocene Rhinocerotidae from Eurasia: impact of climate changes" Since very few study focus on the paleoecology of Miocene Rhino, this paper is a great contribution that should be published.*

*In general, this paper is very rich, well written with supplementary material that provide a lot of data to better understand the study.*

→ Thank you!

*However some points should be improve either to facilitate the understanding of the study or to moderate some affirmations :*

*- In the method, the authors explain which data they choose to investigate hypoplasia. However they never explain the reason behind this selection. Hypoplasia remains quite a mysterious phenomenon and I am wondering for example what information can be extrapolate from the distance between the defect and the collar of the teeth. Could the author detail more their reasoning?*

→ You are right, we detailed that in another paper but forgot to explain it again here. As hypoplasia is not a topic commonly analyzed by the paleontological community, everything should be thoroughly explained. Accordingly, we added a few explanations about that in the manuscript, as detailed below:

*"In parallel, qualitative data (tooth locus affected, position of the defect on the crown, and severity) and caliper measurements (distance of the defect from enamel-dentine junction, width if applicable) were taken (details in Supplementary S3). This data may give insights into the age at which the defect occurred (locus affected, position on the crown, distance to enamel-dentine junction), and to duration and intensity of the stress (width of the defect), which can in turn help us propose causes (e.g., birth for a defect near the base on a D4)."*

*- In the discussion, "Interactions with co-occurring herbivores" part. I find the author quite rush in their affirmation. They seem to forgot that they are studying palimpsest with a very high-resolution method. There is a discrepancy between the precision of the method and the context in which it is applied which should be taken into account and reported in the text.*

→ Absolutely, we may have been a little too enthusiastic here and leading to some over-interpretation. We revised this part and nuanced a little bit more when it was necessary.

*"Interestingly, no rhinocerotid specimens studied here seemingly favored fruits, although they might have included some in their diet (*B. brachypus* from Simorre) or consumed some seasonally, which might not be detected by DMTA."*

*"might have favored grasses (Van der Made, 2003; Xafis et al., 2020), a resource not dominant in the diet of the sampled rhinocerotids neither"*

*These two points in particular seem to me very important to consider. Otherwise, I have pointed out in the text (see attachment) some minor errors/changes/suggestions that the authors may or may not take into account.*

*I hope these few comments will be helpful for the authors.*

→ Thank you very much for this constructive and pertinent feedback!

### Reviewer #2 – Christophe Mallet

*In this paper, the authors reconstruct and examine the dietary preferences and the sensibility to stress of almost 20 species of Eurasian rhinoceroses dating from the Early and Middle Miocene. Rhinoceroses were a flourishing clade during all the Cenozoic, and particularly the Miocene, and their abundance in fossil sites*

*allows precise reconstructions of their paleodiets and their variation through time and space, in relation with climatic changes. Using dental microwear texture analyses and enamel hypoplasia on a sample of 1,400 teeth coming from nine localities in Europe and Asia, authors identify different diet relations between rhino species co-occurring in a same site. While clear niche partitioning appears in sites dating from the MN2 (beginning of Early Miocene) and MN6 to 8 (middle and end of Middle Miocene), a niche overlapping is observed for sites dating from the MN4 to 6 (end of the Early Miocene and beginning of the Middle Miocene). They also observe the absence of purely grazing and/or frugivore rhinos, as well as different prevalence of enamel hypoplasia depending on sites and species. Authors notably underline the low prevalence of stress markers in elasmotheriines, a group showing the emergence of hypsodonty along its evolution. All these results highlight the specific and ecological richness of rhinos during the Miocene, the diversity of their diets, and their variation of adaptability in relation to climate changes.*

*This paper is very well-written, clear and easy to follow, relying on a broad dataset and robust results statistically relevant.*

→ Thank you!

*Only some minor flaws can be detected along the text (see below and in the annotated version) but these small problems can be fixed easily.*

*The Abstract is very clear and concise, with well-exposed problematic and results. Only the end is a little bit frustrating, because it opens on the question of elasmotheriines and their peculiarities without giving hints regarding the origin of it.*

→ You are right, we added a precision to give some hints about that and make it less frustrating.

*“The very large hippo-like *Brachypotherium brachypus* was one of the most affected species at all concerned localities (but Sansan), whereas early-diverging elasmotheriines were very little affected, suggesting an influence of phylogeny and/or diet in stress susceptibility.”*

*The Introduction is short and concise but very clear. All the important references related to the study are provided and support well the interest of the study. I feel like it is lacking a clearly stated problematic for the paper at the very end of the introduction, that would justify the existence of the present study. Hypotheses regarding the expected results can also be a significant improvement for this part, as well as some details regarding the technics used in the study, only mentioned at the very end but without details. Those details are given elsewhere in Material & Methods section but understanding how and why these methods may help to resolve the problematic would significantly improve the strength of the Introduction.*

→ We agree and we added all of that at the end of the introduction, resulting, indeed, in a great improvement.

*“With this article, we wanted to assess if the great diversity of Miocene Rhinocerotidae was associated to an ecological disparity and to explore paleoecological differences associated with climate changes between the studied species, region, and periods. To do so, we focused on the rhinocerotids from nine localities, covering wide temporal and geographical ranges (from MN2 to MN7/8 and from southwestern France to Pakistan). We coupled dental microwear texture analysis (short-term diet proxy) and enamel hypoplasia (i.e., enamel defect resulting from a stress stopping tooth development) to infer dietary preferences and stress sensibility. Eventually we compared the results of both approaches, which has the potential to detect stressful shifts in the diet (competition, punctual shortage event) or accentuated susceptibility due to a specialized diet, and put them into phylogenetic and environmental contexts.”*

*The Material & Methods part is also very clear, even for a non-specialist of these methods like me. All the code, the raw data and the detailed results are provided as Supplementary Data, which constitutes an excellent point to ensure the replicability and the relevance of the results. For what I saw in the raw code and raw data, no major flaw seems to be present.*

→ Good to know that everything is clear to non-specialists as well!

*The Results are clearly presented, focusing on relevant and essential points among the multitude of analysed data. Figures 4, 5 and 6 are particularly well-presented and informative. The summary for each locality is also appreciable and all the results are supported by enough details given as Supplementary Data. However, I am concerned by one point regarding the computation of the frequencies of hypoplasia. As these frequencies are computed on samples of very different sizes (a few teeth VS hundreds of teeth), the direct comparison of these values might lead to spurious results. A simple way to avoid such a problem would be*

to perform an ANOVA or a MANOVA on the frequencies to compare their variance instead of their raw values. I do not think the results will change a lot doing so, but they would be more relevant and robust statistically speaking.

→ Regarding sample size this is a very pertinent point. This is the reason we always provide counts and frequencies. Regarding statistics, ANOVA or MANOVA are indeed pertinent, which is why we used a Kruskal-Wallis test (non-parametric ANOVA) and GLMMs that do take into account sample size. We indeed found similar results/trends using GLMM and comparing raw number/frequencies.

*The Discussion is well-constructed and well-written, the part regarding the ecological niches is clear and relies directly on the results presented before. Overinterpretation of the data is avoided, especially for the sites with too little data available. There is a very good correlation between the data presented here and what we already knew in terms of climatic changes on the concerned localities. Two very interesting points might be reinforced. First, regarding the variation of diet in B. brachypus, we could expect some hypotheses trying to explain the origin of such a variation, unique among the rhinos studied here. Second, concerning the particular case of elasmotheriines, that show far less hypoplasia, the reader could also expect here stronger hypotheses concerning this fact.*

→ These are indeed two very interesting findings of this study. We completed the concerned paragraph in the section "Paleoecologic implications and changes" of the discussion

*Finally, at the end of the Discussion, I have the feeling that all these results should be replaced in a larger "big picture" regarding the evolution of paleodiets in rhinos and ungulates, and their relation with climate changes, as described in the title of the paper.*

→ We agree on that point but we chose to do that in the revised conclusion rather than in the discussion. See next point

*Finally, the Conclusion is maybe the less accomplished part of the manuscript. Although good, it tends to repeat the main results and findings of the study, while the reader might expect more opening on the interest of these results for the fields of paleodiet, climate changes, evolution of rhinos, etc. New questions or new hypotheses to explore can be expected after such a massive work and should be stated here, at least partially.*

→ The conclusion was indeed a bit dull, mostly repeating the results we obtained (which were already presented in the abstract). We modified the conclusion according to these remarks.

*Except for these minor flaws, the article in general is a strong piece of work with robust results and of great interest for the comprehension of the paleobiology of rhinos, and how it correlates with climate changes. In summary, I recommend hardly this paper for acceptance with minor revisions for PCI Paleontology.*

→ Thank you very much for this very complete, detailed and pertinent review!

### **Reviewer #3 – Matthew Muhlbachler**

*This paper includes an interesting mixture of dental microwear texture analysis and documentation of dental hypoplasias in Neogene Eurasian rhinos. The dental microwear analyses adopt established methods. Likewise, the distribution of hypoplasias are documented soundly. However, the results and discussion sections wander, generate unsupported paleodietary interpretations and make loose associations of the results with paleoenvironmental conditions. While the data themselves are analyzed with sound statistical methods, the authors take very large leaps from the statistical results regarding differences between the various samples to generalizations about diet and paleoecology.*

→ Thank you for noting the robustness of our methods. Regarding your comment on "unsupported paleodietary interpretations", we are a bit surprised as we have tried to be very careful in our interpretations especially when we had a small sample size, which has been noted by reviewers #1 and #2. This may come from a misunderstanding of our use of some terms ("soft browsing", "favoring leaves...") as dietary categories, when we only wanted to give insights into mechanical properties of the items that might have been consumed within the range of the classic categories (browser, mixed-feeder, grazer).

*The paper states that its aim is to assess "dietary preferences using dental microwear texture analysis, and stress sensibility via the study of enamel hypoplasia". (lines 65-67). However no hypothesis was provided,*

*leading to an unfocused presentation. I think the authors would be better served by generating a testable hypothesis or series of hypothesis which could then be used to give the investigation purpose and focus. What ecological paleoecological questions concerning Neogene rhinos can be addressed by these data? Answering this question may also lead to a more coherent research design. Why are dental microwear data and dental hypoplasias being presented together? Is there some hypothesis that they can both address? Presently, it seems as if microwear reveals more about diet, while hypoplasias have more to do with ecological stress. These aren't mutually exclusive phenomena. Is there some interesting question that could be answered by examining both. If not, the authors may be better off publishing these two studies separately.*

→ This pertinent point has also been raised by reviewer 2, and we developed the last paragraph of the introduction, notably adding a problematic and we explaining the link between microwear and hypoplasia.

*Throughout the DMT results and discussion sections and summarized in table 6, a variety of interpretations are made about diet and feeding. Most of these interpretations use the classic browser, mixed-feeder, grazer categories, but other ad hoc terms and special qualifications are thrown about including "folivore", "browser favoring leaves", "soft food items", "soft browsing", "browsing preferences with the inclusion of hard objects, probably fruits".*

→ All categories in table 6 (except folivore) have been defined by Scott (2012) based on extant bovids. Regarding folivory, it is based on repeated observations of extant browsing species having high values of anisotropy (deer: Berlioz et al. 2017; rhinos: Hullot et al. 2019; various ruminants: Merceron et al. 2021) and on the low probability of finding grazing species during early/middle Miocene as grasslands and grazing forms are documented only from the late Miocene onward in Europe.

All other terms used in the manuscript are synonyms of these categories or precisions based on the textural parameters (e.g., soft browsing would be a browser in the lower range of complexity for this category as opposed to browsing preferences with the inclusion of hard objects, probably fruits). These terms and precisions are often used in the microwear literature and are a way to overcome dietary categories that are purely abstract and not representative of the whole herbivore diversity.

*The statistical methods used to test for differences in the data give a false impression of scientific rigor. But the actual dietary interpretations have seemingly been made out of thin air.*

→ We find this comment a little bit harsh and almost offensive, considering the transparency effort on raw data and statistical analyses. All the interpretations have been made carefully, as noted by the other two reviewers, within the calibrated framework of extant species from Scott (2012) and the extant rhino-based dataset of Hullot et al. (2019). As this point seems not to be clear enough, we specified further in the materials and methods section that our interpretations are based on DMTA calibrated in extant species, following the works mentioned before (Scott, 2012; Hullot et al., 2019).

*How were these interpretations made? Extant rhino microwear data are discussed in a subsequent section but it is not clear how the modern data are being used to interpret diets of extinct species.*

→ We feel that our interpretations have been justified enough, both here and in the manuscript. They arose from robust statistics (ANOVA/MANOVA and GLMMs) of the textural parameters in the framework of extant species.

*What are the confidence levels of these dietary assignments? How are these dietary categories defined? How were the lines drawn between browser, mixed-feeder and grazer in terms of microwear data and diet?*

→ These are very interesting questions but we feel that they may fall beyond the scope of this manuscript. These problematics have been addressed in various other papers and cutpoints have been defined (e.g., Scott, 2012; Merceron et al., 2016; Hullot et al., 2019). We have added some elements in order to justify these statements accordingly.

*What aspects of the microwear data lead you to make conclusions about diet?*

→ As detailed in the materials and methods section, our study is based on five classical textural parameters: anisotropy, complexity, fine textural fill volume, and two heterogeneities of the complexity at different scales. Similarly to most of the DMTA studies, a greater focus was put on anisotropy and complexity as they best discriminate the traditional dietary categories (browser, mixed-feeder, grazer), while other parameters were

used to confirm or precise our interpretations (e.g., great values of HASfc have been linked with a greater diversity of food items consumed).

*What about these other special categories, such as folivore, soft browsing, and other special exceptions (e.g. browser favoring leaves, soft browsing etc)? What elements of the data reveal these? It is already challenging enough to differentiate classic browsers, mixed feeders, and grazers using microwear data and these additional diet characterizations, I suspect, are actually beyond the powers of the available dental microwear data to differentiate.*

→ We agree that microwear data can be limited sometimes. However, as detailed before these terms are not dietary categories *per se* (except folivore), but precisions and used for comparison (e.g., a sample with higher values of complexity is interpreted as consuming harder food items than one with lower values).

*In several areas, this paper makes strong conclusions about diet and other aspects of paleoecology. For example, in the conclusion section (lines 663-665):"Though, DMTA results suggested only browsers and mixed-feeders (no grazers nor frugivores) in the studied rhinocerotid sample, they unraveled clear niche partitioning through food resources at several diachronous localities..." Niche partitioning is a process by which natural selection drives competing species into different patterns of resource use or different niches. Some microwear differences found in a small number of teeth do not constitute clear evidence for niche partitioning or any other complex evolutionary and ecological process. The dental microwear differences merely suggests different diets or feeding environments. It does not reveal why those diets are different. With regard to niche partitioning, it can only be concluded from this analysis that it cannot be falsified as an explanation for the differences. Perhaps it would be helpful here, and, in fact, for all the conclusions derived in this paper, to consider what is the null hypothesis regarding ecological differentiation of these species to begin with? This is one example of numerous in the manuscript where your conclusions need substantial softening.*

→ We agree that our sample is somewhat limited, which is why we concluded on our sample and not on the whole population(s) or species. In agreement with reviewer 1 we have toned down some elements of the discussion. Niche partitioning is indeed a complex process, and we discussed several strategies besides diet. Competition and dietary shifts rather than niche partitioning are also debated in parallel. We feel that our statements are supported and already nuanced in the manuscript.

**Specific comments** (see comments in manuscript for additional comments)

*The first two sentences of the abstract seem unnecessary to me. Of course major changes are documented in most of not all geological ages. Rhinoceroses are interesting, according to who? They are abundant and diverse according to what metric?*

→ We kindly disagree with this comment, as it is a convention to start an abstract with a general catch phrase.

Rhinocerotidae are interesting in that they are particularly abundant (i.e., number of specimens), species-rich and morphologically diversified in the concerned time interval and area. As the abstract is by convention citation free, there is no specific "according to who".

*In both the abstract and the main document, proxy data are confused with diet (or other ecological parameters of interest). For example, line 22 states, "Our results suggest a clear niche partitioning based on diet at Kumbi 4..." Replace "diet" with "DMT data". The evidence for niche partitioning is not based on diet, it is based on DMT data, which is not diet. Also see comments above about niche partitioning.*

→ See the justification above.

*Likewise on lines 24-25, "All rhinocerotids studied were browsers or mixed feeders, and none had a grazing nor frugivore diet." Avoid such definitive conclusions. To be more precise, it would be better to write, "the DMT data are consistent with browsing and mixed feeding diets, but not grazing".*

→ We corrected the manuscript accordingly and added more nuanced statements.

*Line 27: If the prevalence of hypoplasias document the local conditions, those conditions need to be stated, otherwise omit this sentence.*

→ We modified the abstract to make it clearer that Sansan and Devínska Nová Ves were examples of that, and we added Béon 1 as well. Besides, the local conditions are detailed extensively in the Supplementary Data and in the Discussion, and it is difficult to detail them for all nine localities in a word-limited abstract.

*Line 90: A figure of the grinding and shearing facets sampled needs to be provided. A person who is not very familiar with rhinocerotid molars will need a figure to understand this.*

→ This figure is indeed provided in Supplementary Data as indicated in the text.

*Line 171-172: The statement about avoiding classic thresholds is contradicted in the results tables. Table 2 appears to use  $P=0.1$  as a threshold. Table 3 explicitly uses  $P=0.05$  as a threshold. A keyword search for the word "significant" is needed to be sure that it is not used in ways that contradict the statements about significance in the methods (see line 666)*

→ The statement of the ASA on p-value is about limiting the unwary use of the term “statistically significant” as a holy grail for a p-value < 0.05 or other arbitrary cutpoint. This does not mean that thresholds should be completely banned, just that scientists should be more careful about their use and the conclusions they draw. For instance, p-value = 0.048 and p-value = 0.052 should be treated with more nuances than “significant” and “not significant”. We clearly said in the manuscript that we would avoid this “as much as possible” and not get rid of it. This is why throughout the manuscript, such thresholds can be found. However, when we have used them it is clearly mentioned and explained.

Regarding the use of “significant” in the conclusions it has nothing to do with statistics, it is just a visual observation and it could have been replaced by one of the following words: “great”, “marked”, “important”. Anyway, this sentence has been removed for the revised conclusion, following the remarks of reviewer 2

### **Other points addressed based on annotated versions**

In the abstract:

We kept “moderate” and “high” as well as the values about the prevalence of hypoplasia because we thought it adds an interpretation to the raw results. The reader might not be familiar with hypoplasia and not know if 10 % is a lot or not.

In the introduction:

The local conditions at every locality are described in the Supplementary Data

In the M&M:

We used a polynomial degree 8 because it was visually the most efficient to flatten the surface. The vertical Hunter-Schreger bands (HSB) in rhinos are very marked and would artificially bias the topographic analyses if they are not removed (greater complexity and heterogeneities). To our knowledge, we are the only team using DMT to investigate rhinoceros microwear. Other scholars use 2D approaches (Rivals, Muhlbachler, Uzunidis, or Pandolfi) and thus they do not need surface levelling, or do not study rhinos microwear (Kaiser, Clauss, or Winkler). As for other taxa with vertical HSB (some tapiroids, South American Native Ungulates, ectoloph of some chalicotheroids and brontotheroids), 3D microwear has only been applied to tapirs (DeSantis et al 2020), and there is no precision on the facet sampled (maybe no HSB?).

In the results:

“great variety of microwear patterns”: the values are detailed after