

## ***Interactive comment on “Alpha and beta diversity patterns of polychaete assemblages across the nodule province of the Clarion-Clipperton Fracture Zone (Equatorial Pacific)” by Paulo Bonifácio et al.***

### **Anonymous Referee #2**

Received and published: 13 August 2019

The paper provides results of box corer-based study of polychaete macrofauna in the eastern part of the CCZ that significantly contributes to our knowledge on faunal communities in the CCZ. The study's title is appropriate but may raise expectations that the study covers the entire CCZ (see below). The work addresses relevant scientific questions of strong societal significance – and contributes knowledge that can help an informed societal decision regarding if and how to do nodule mining. The study contributes to our understanding of patterns benthic assemblages in the deep sea and how they relate to environmental drivers. At the same time it raises important questions in the context of a potential exploitation of nodules, e.g., regarding the appropriateness of the concept and location of the APEIs and the underlying regional management plan

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– and about future effort necessary to answer the questions that have to be answered before mining commences.

The scientific methods are valid and up-to-date and the experiments and calculations are adequately described. The authors acknowledge related work and clearly discriminate their data from data obtained by others. In some cases, where they discuss their data (e.g., regarding winners and losers of the ‘competition’ of meio- and macrofauna) in relation to that of others they should be more specific about the exact content of the cited data so the reader can better comprehend the authors' discussions and conclusions. The data presented are sufficient to reach conclusions part of which – and that is not criticizing the ambitious work behind this study – are pointing out gaps in knowledge. The paper is well structured and reads mostly fine – in some parts of the results the language may seem a bit repetitive (abundances and species numbers are presented with almost the same wording). The abstract provides a good summary. In the way it is currently presented, the first part of the discussion (about the relative success of meio an macrofauna in different deep-sea environments) should be either significantly reduced or more specific with a better and quantitative presentation of data on Meiofauna from the study area from other studies. I would vote for a reduction of that part as it seems to be a bit off the main focus anyway.

To me, the main shortcoming of the paper is, that the authors are very carefully when stating their conclusions and overcautious if it's about the consequences of their findings. As the work touches societal concerns and areas of strong debates the authors should address in more depth the implication of their work regarding procedures and management of nodule mining, e.g., can we decide on regional management based on the available knowledge and are the APEIs appropriate as they are? Are there any specific recommendations that can be provided, e.g., regarding the size or arrangement of mining patches? What should be the focus of future studies and what would be the expected effort needed to come to scientifically sound conclusions? The conclusions part is so far rather summarizing what has been stated already before and may be a

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good place to discuss these things. The fact, that those discussions are rather limited in the current version of the manuscript is the reason for my general proposition that the manuscript should undergo a major revision before publication.

Below some more detailed comments

#### MAJOR ISSUES – IMPLICATIONS FOR MINING / MINING REGULATION

Regarding the main shortcoming of the paper mentioned above I urge the authors to significantly extend the discussion of their results towards the implications of their work with regard to nodule mining and its regulation. This could be distributed in several parts of the discussion as well as in a separate section in the discussion or in the conclusions. This, of course, has to be done with some caution to not extend beyond the scope of the study and has to take into account that this is a scientific publication and not a policy paper. However, it is clear that the motivation of the study – and certainly of societies providing the funding for mining-related investigations these days – is to provide the basis for scientifically sound procedures and decisions regarding deep-sea mineral exploration and exploitation. This should be better reflected in the text. This includes recommendations regarding the management and regulations - where the data of the study allow this - but also specific requests for future investigations where the results reveal significant gaps. Below I am providing some examples where I think the discussion needs to move beyond where it currently terminates.

Page 3, line 15/16 'The distribution of APEIs at the periphery of the CCFZ thus deviates from an optimal design.' Page 11, line 20-23 'The biogeochemical settings as well as the biological patterns of the three size groups of the benthic fauna thus converge to conclude that the structure and functioning of the benthic ecosystem in APEI#3 is not representative of any of the four exploration contract areas included in this study.' Page 12, line 26-29 'The influence of the fracture zones on the dispersal of the abyssal fauna remains to be better understood as the Clarion and Clipperton fractures may act as a barrier for species with low dispersal abilities such as infaunal brooders. If so, the

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representativeness of seven out of the nine APEIs, which are partly lying beyond the fractures, may be questionable.' If these statements hold true, the concept of APEIs and the regional management plan as a whole don't seem to be appropriate. What is the advice of the authors to overcome this problem? What do we know about the other APEIs and how their environmental conditions and faunal communities compare to license areas? What would be an optimal APEI layout and how would you – from the results of your study – address the question whether an area is suited as APEI or not. Can we use some easily measured sedimentological (grainsize?) or biogeochemical measurement to assess the probability that an APEI will host similar faunal communities than a specific license area? Should the assessment of correlations of habitat characteristics and fauna in APEIs be a focus of future studies? If you consider the lack of knowledge potentially only a few years before exploitation commences: Should the ISA setup a scheme by which contractors carry out or fund baseline studies in the APEIs? One consideration that lead to the APEIs' current position outside the area covered by license areas was to allow for very large areas. In light of the fact that, according to the current planning, only part of the license areas will be used for nodule extraction and the seemingly low species' ranges: do we need APEIs to conserve biodiversity or would the areas inside the patch of license areas, that are not mined do the job? Or do we need APEIs somewhere else, e.g., smaller ones between license areas?

Page 13, line 1-5 'However, based on the best knowledge we have, our study suggests that [...] nodule mining would affect each year an area that is equivalent to the average geographic range of a polychaete species.' Spatial ranges – especially if they are indeed that small – are highly relevant. Can we use polychaetes as key species here or would we need to have similar data also for other size classes and other groups of macrofauna? What data are available already? What are the implications of these results for mining operations and their regulation? Do we need more research to understand whether the estimated spatial range is really true or just mirrors the inappropriate sampling effort available scientific knowledge is based upon? Or do we

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'know enough' and could provide specific suggestions as for how to spatially arrange mined patches? Taking this further: if we take the precautionary approach seriously: if we have an average species range of 20km (and, for some of the species obviously a smaller one) wouldn't we need to restrict the mining operations by contractors including secondary impacts by the plume to that size until it is proven, that the high turnover of beta diversity is an artifact of undersampling?

Page 15, line 27/28 'The assessment of potential risks and scales of biodiversity loss thus requires an appropriate inventory of species richness in the CCFZ.' While the conclusions are basically a summary up to this point, this is where the discussion in implications starts: How should this goal should be achieved? How much of this work is, according to the knowledge of the authors, already done by baseline work of the different contractors and just needs metaanalysis of the pooled contractor's data (e.g., by an independent scientific consortium)? Or are samples and data lacking and more dedicated sampling campaigns needed? What effort would this take? Or - if that is too hard to estimate, how would you control if enough data are available (based on rarefaction curves? Based on biodiversity descriptors merging?)? If you compare this to what is found in the ISA regulations and guidelines: how does that compare? What about the key-species concept? Could that become appropriated once the necessary knowledge was obtained or do you think we always have to cope with the full complexity when we want to address environmental impacts of deep-sea operations (assessment of the risks prior to operations, assessment of impacts happening during operations).

#### OTHER MAJOR ISSUES

Page line 3-27 The discussion of meiofauna in nodule areas comes as a bit of a surprise in the context of this paper, that does only provide data on macrofauna. If you want to leave this so prominent and detailed, you should first state what the data show. Does the Pape et al. study provides data from the same station so a quantitative comparison to other deep sea areas is possible? > In this case I would suggest to provide that quantitative information here. Otherwise consider reducing the discussion or move

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it to a less prominent part of the discussion. Maybe you could also connect it more to the paper, e.g. as an argument for focusing environmental impact studies on macrofauna because they seem more relevant in terms of biomass and ecosystem function as in typical abyssal areas.

Connected to this: Page 10, line 16 'The contribution of meiofauna to benthic biomass generally increases along a bathymetric gradient [ . . ] which suggests the occurrence of a dynamic equilibrium between meiofauna and macrofauna at abyssal depths.' Again - this is very detailed for a study that does not focus on size class comparisons. I assume the Sibuet paper focuses on non-nodule areas? I understand you want to put forward that macrofauna is particularly important in the CCZ / in nodule areas as they – different to what was previously reported - show but a relative increase as compared to Meiofauna (i.e., neither do they show a relative decrease at depth as compared to meiofauna nor do they scale with meiofauna). This really would need a quantitative basis, i.e., a comparison of macrofauna abundances (better biomass) to meiofauna abundances at your study sites relative to other areas. > consider adding more quantitative information

Connected to this: Page 10, line 23 'Due to its small size, meiofauna is likely more efficient at exploiting the low level of food input, but this interstitial fauna may also be more sensitive to high nodule coverage because its ambit is largely limited to superficial sediments.' Do you mean, that the meiofauna is restricted to the top layer where the available sediment volume is limited by the presence of nodules? I think you dont show the data but I assume that also for polychaetes the top sediment layer is the one where most individuals are found. In any way you could strengthen this idea by comparing your abundance vs. depth relationship with that of meiofauna in nodule areas. > please comment, explain and consider including this information in the manuscript

Page 14, line 7/8 'Overall, the combination of high local diversity, unsaturated rarefaction curves, high levels of cryptic diversity and high rates of species turnover suggest that polychaete diversity in the CCFZ is large and vastly under-sampled.' It needs a

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discussion of the most appropriate technologies (sampling gear, analysis) and the expected effort it needs to raise our knowledge to a level appropriate to decide on mining (yes or no, spatial organization of operations and protected areas), and allow for scientifically sound impact assessment and management. Maybe in this context it should also be discussed, if (and why!) the authors believe, that polychaetes may serve as a model group for baseline and impact assessments. Or is this just the 'pet group' of the authors and any other group should be similarly addressed before taking decisions? > please extend discussions to include these points.

#### MINOR ISSUES

Page 1, line 1-3 Including the metaanalysis performed the study indeed addresses the entire CCZ. However, the stations of this study are all rather in the eastern part. > rephrase the title to not raise false expectations, e.g., by replacing 'across the nodule province of the Clarion-Clipperton Fracture Zone' with add 'across the nodule province of the eastern Clarion-Clipperton Fracture Zone'

Page 1, line 17 '...the SO239 cruise aimed at improving species inventories...'. Was this really the subject of the cruise as a whole or of this expedition? > consider rephrasing

Page 2, line 6 'Only about 1 % of abyssal plains have been explored to date'. In this context of this paper I would restrict the use of the term 'explore' / 'exploration' to deep-sea mining-related activities > consider rewording

Page 2, line 7 'In particular' seems to connect to the previous sentence but in fact does not. > consider remove

Page 2, line 9 '...mainly manganese and iron,...' > I would also mention copper, nickel and cobalt right away here - than you don't have to repeat that in line 13/14.

Page 2, line 15/16 '...the International Seabed Authority [...] is in charge of protecting fauna against any pollution or other hazards...' Pollution is not the main concern in

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the context of nodule mining and expected impacts related to this study. > I would rephrase. Maybe just refer to harm (i.e., 'protecting fauna against harm')?

Page 2, line 25 I don't understand what is meant with 'scaling issue'. Is this referring to the uncertainties connected to effects of the full scale, long-term operations with large plumes as compared to single experimental tracks? > please rephrase / be more specific

Page 2, line 27 > replace 'the high diversity' by 'a high diversity'

Page 3, line 23 'test the hypotheses that support spatial conservation planning in the CCFZ'. I don't think that these hypotheses (that the authors think would serve as guidance or that form the basis of the current regional management plan are explicitly stated somewhere in the publication. > consider being more specific here or state them elsewhere in the paper

Page 4, line 1/2 > replace '...were located between 4000 and 5000 m depth...' by 'had water depths between 4000 and 5000m'

Page 4, line 12/13 '...all nodules picked up from the sediment surface, washed and individually measured and weighed...' It should be mentioned already here that the water that was used for washing the nodules was sieved after washing. Have the nodules themselves been inspected for small polychaetes, e.g, living in tubes attached to the nodules? > rephrase and make sure to mention somewhere in the paper, if the data also include nodule-associated polychaetes

Page 4, line 18 '...The sieve residues from the overlying water and the washed nodules were combined with all layers for the community analysis...' Was the material combined (i.e., before analysis) or the data? > specify in the text

Page 4, line 20/21 '...(see Section 2.3 DNA extraction, amplification, sequencing, and alignment)' No need to refer to a section that follows directly > remove

Page 4, line 24/25 '...and 1600 bp of 18S genes...' ? Are 18S data really used in this

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study (I could not find it later on)? If not: restrict M&M to 16S and COI or discuss why that approach was not successful or not included in the analyses.

Page 5, line 13 'To separate closely related species...' [...] observed between intraspecific and interspecific variations' What does 'closely related species' mean? Specimen that could not be discriminated based on morphology? > specify Page 5, line 13-17 To separate closely related species [...] observed between intraspecific and interspecific variations' This section is describing the principle not what actually was done. This does not fully qualify for a Materials and Methods part > Move to another part of the study (introduction?) or rephrase.

Page 5, line 25 '...to calculate nodule density...' Is nodule mean size or size distribution also considered in this study? If not, why was this not included as a parameter that may shape communities? > explain, consider adding explanation to the paper

Page 5, line 25-29 'Particulate organic carbon flux (POC, mg C m<sup>-2</sup> d<sup>-1</sup>) at the seafloor for our study areas [...] applying the Suess algorithm (POC at the seafloor as a function of the net primary production scaled by depth; Suess, 1980; Table 2).' How do POC fluxes estimated with different methods compare where they overlap (i.e. in the study area?) > consider adding that information to the paper.

Page 6, line 1 '2.6 Regional-scale data' Also the Ocean Productivity-based POC fluxes in the previous section refer to the regional scale > choose another headline, e.g., 'Regional scale polychaete community data'

Page 6, line 6 > add references for ES163 and bootstrap

Page 6, line 20/21 'Spearman correlations were sought between biotic and abiotic variables, using data from the SO239 cruise in the CCFZ and data compiled from the literature.' The data used for these correlations should match the data sources listed in section 2.5 > to avoid confusion I suggest to just refer to section 2.5. here. If the 'biotic and abiotic variables' include data not mentioned in section 2.5 add them there.

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Page 6, line 23 to Page 7, line 14 Also in this section it should be described what has been done while a description of how the methods work does not seem appropriate for the M&M section (e.g., 'Low values of m give a high weight to dominant species, high values of m give a high weight to rare species:'). > Rephrase, possibly move parts to other sections

Page 7, line 21 '...tended to decrease from east to west with high spatial variation' 1. the main axes does not seem to go strictly longitudinal > replace 'east to west' by 'southeast to northwest' 2. 'high spatial variation' would make more sense in a study design, that follows a clear geographical transect. > consider rephrasing, e.g., 'high variability between neighboring areas'

Page 7, line 21 'The mean abundance in each study area tended to decrease from east to west with high spatial variation...' 'with high spatial variation' would make more sense in a study design, that follows a clear geographical transect with similar lateral distance between sampling points. > consider rephrasing, e.g., 'high variability between neighboring areas'

Page 8, line 2 'The relative contributions of trophic guilds also varied among the areas...' Is there an explanation found somewhere, how trophic guilds were determined? > If not, add description and references to M&M.

Page 8, line 6 'Off the 1223 polychaetes, 1118 specimens belonging to 78 possible genera within 40 families were identified down to Morphospecies...' What are 'possible genera'? > consider rewording, e.g., '...possibly belonging to 78 genera...'

Page 8, line 6/7 '1118 specimens [...] were identified down to morphospecies (see Section Data availability)' Not sure why you refer to that section here. > please provide explanation and consider including it in the text.

Page 8, line 14 'The mean number of species tended to decrease from east to west with high spatial variation...' see comment above (regarding Page 7, line 21, second

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comment)

Page 10, line 13/14 'Both processes [i.e., increased friction and sediment deposition / biodeposition rates] may stabilize sediments and increase organic carbon supply as tube lawns do' I don't see the connection to sediment stability. > please explain better what your idea is here

Page 10, line 15/16 'The divergent response of meiofauna to the presence of nodules further suggests some sort of competition between meiofauna and macrofauna.' I can see that - if nodules increase food supply but meiofauna abundances are relatively small, meiofauna may be unable to make full use of the additional food. What I don't understand is why the reason does need to involve competition with macrofauna (see also my major comment on the meiofauna discussion above). > please provide explanation and consider including it in the text.

Page 11, line 34/35 'No significant correlation was however found between alpha diversity and productivity, neither at the NE Pacific scale nor at the scale of the whole CCFZ.' Do the authors have a hypothesis why this can be the case? Could it be related to the fact that most of the tested areas lie within more or less similar mesotrophic conditions and that this 'biased' data set is not fully appropriate to address this question? > please consider discussing the reason for the missing significant correlation of diversity and productivity on larger scales.

Page 12, line 5/6 'The fact that the APEI#3 lies mostly north of the Clarion Fracture Zone may however also contribute to its dissimilarity with the areas located in the CCFZ per se.' This statement reads quite vague as the idea of geographical barriers is not mentioned and elaborated before the next section > please consider adding (see next section) after the statement.

Page 12, line 12/13 '...characterized by a peak and through ...' Typo > change 'through' to 'trough'

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Page 12, line 24-26 'However, species identification was based on morphology only, although cryptic species are common among scavenging amphipods, even in abyssal lineages (Melo, 2004; Havermans et al., 2013)' Another reason is, of course, that scavenging amphipods are typically highly motile. > consider adding mobility as an argument why scavenging amphipode distribution is not limited by fracture zones.

Page 13, line 5 'In other words, nodule mining would affect each year an area that is equivalent to the average geographic range of a polychaete species.' This sounds like one mining operation would lead to the extinction of one polychaete ('only' - as some may argue). > consider removing 'a', i.e., write 'equivalent to the average geographic range of polychaete species...'

Page 13, line 27/28 '...suggesting that such extreme environmental conditions...' I don't share the view that the deep sea is per se an extreme environment. > replace 'such extreme' with 'the specific' or explain what specifically is considered extreme

Page 14, line 1/2 'This highlights a shortcoming of COI-based barcoding because success rates for COI sequencing are generally low...' ? Are current molecular approaches appropriate if only a relatively small proportion could be identified based on 16S and COI and even less with both? Where is the problem and can it be overcome? If there are new promising methods that base on other regions of the genome: how can we safeguard comparability of the full data set including new and older data?

Page 15, line 2-4 'The latter estimate assumes that we have sampled 0.1 % of the polychaete species in the CCFZ and that these species have narrow geographical ranges about the size of a yearly mined area.' If I understand right, this refers to the expected annual area exploited as part of one mining operation – not the total annually mined area > replace 'a yearly mined area' with 'the area that will presumably be mined in one year by a single mining operation'.

Page 31, Fig. 3 Irrespective of the fact that the variables are provided in the diagonal panels I would prefer if to the side of the plot the variables would be indicated like in

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<https://images.app.goo.gl/oFQRE6xD7fvFwxJR6>

Page 32, Fig. 4 ‘...in relation to the 2002–2018 average particulate organic carbon (POC) concentration at the seafloor along the CCFZ. The background map shows average POC flux at the seafloor during the 2002–2018 period.’ How can the maps show relations to POC concentration and flux at the same time? > consider rephrasing the caption. The caption should also state that this shows / includes data from published studies and refer to section 2.6

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Interactive comment on Biogeosciences Discuss., <https://doi.org/10.5194/bg-2019-255>, 2019.