Interactive comment on "Benthic communities in the deep Mediterranean Sea: exploring microbial and meiofaunal patterns in slope and basin ecosystems" *by* K. Sevastou et al.

Anonymous Referee #2

Received and published: 2 February 2013

General comments

The present study entitled "Benthic communities in the deep Mediterranean Sea: exploring microbial and meiofaunal patterns in slope and basin ecosystems" by K. Sevastou and colleagues presents analyses on diversity patterns of meiofauna and bacteria in deep-sea sediments of the Mediterranean Sea. The authors have used a large set of samples to span two different habitats, namely slope and basin areas. Their results show decreasing depth trends of meiofaunal abundances, bacterial abundances were not included. Also the number of meiofauna taxa (richness) showed a significant decrease with depth, while bacterial richness rather displayed an increasing trend with

water depth. In addition, significant differences in beta diversity were evidenced between depth ranges in the basin habitat, but not for the slope. This seems surprising, as I would expect larger differences along slopes due to stronger environmental gradients. The authors address this question in the discussion, but due to the lack of contextual data, they cannot conclusively suggest an explanation for this result. One of the main results is that depth plays an important role for meiofaunal diversity; this is something that was known before, but was here confirmed with an extensive dataset from the Mediterranean deep sea, including slope and basin samples. Little is known about the distribution patterns and driving environmental factors of deep-sea fauna, and even less about the small size classes, meiofauna and bacteria that were studied here. The present study therefore adds important information on the structuring of deep-sea meiofaunal and bacterial communities.

The Abstract could be phrased a bit more concise, with less listing of the different diversity results, but rather an overview and the most important points/findings highlighted. Due to the large dataset and the different types of analyses and comparisons, special care should be taken to structure and write the results and discussion as clearly as possible. In the Results it is ok. to list all the different results, but I think that the discussion could be improved in order to formulate/present conclusions more clearly. Maybe a summarizing sentence for the different sections would help, with the major message of that sub-section. I also miss a bit more discussion/speculation on possible reasons for the observed patterns. Maybe it would be possible to include rough extrapolations of certain environmental parameters to test or at least describe possible environmental drivers for the observed differences in the sampled areas.

In such a large sampling effort it would have been desirable to incorporate at least a few environmental parameters. As the authors themselves state, depth has been identified as an important factor in a large number of studies, however the driving factors that may be hidden in the factor depth have not been identified. Unfortunately, no environmental parameters (other than depth) were considered for the analyses in this study, so most of

the discussion on shaping factors/underlying mechanisms (related to depth or habitat type) remain very hypothetical. This study therefore remains very descriptive. However, considering the general sparsity of deep-sea studies, this study contributes important information toward a better assessment of diversity patterns in the deep sea.

Below are some more specific comments on the manuscript.

Specific comments

Introduction p. 17542, l. 11-13: rephrase: Hence, increasing the sampling effort and extending research to include more taxa and more areas of the deep oceans will help to further elucidate bathymetric trends of benthic diversity.

p. 17543, l. 9-11: First you refer to bacterial diversity estimates, but then you talk about the occurrence of bacteria in different habitats such as deep-sea sediments, soils and shallow waters (what do you want to compare?). I do not agree with the statement that bacteria found in the deep sea are similar to those living in soil or shallow water. Clear differences in the composition of taxa/the most important taxa have been shown for these different kinds of environments.

p. 17543, l. 12-14: it is unclear to me what you want to say here. "On the other hand" compared to what? Change "and" to "or", ubiquitous dispersal is the opposite of biogeographic patterns. But, yes, there is little evidence for any of those scenarios because of a lack of studies on bacterial communities in deep-sea sediments.

p. 17543, l. 26: But you didn't measure productivity or proxies of input to the deep sea. Also, I don't really see this discussed in the Discussion section.

p. 17541, l. 1 (see below) p. 17544, l. 1-7: I find the wording of "benthic parameters" to describe diversity estimates very confusing (appears throughout the text). Benthic parameters to me usually describe parameters like oxygen availability or organic carbon content. Better say what you really mean, i.e. diversity estimates/indices. Do not write bacterial species, rather types or groups. In general, the formulation of goals

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should be a bit more specific, what did you specifically want to test, what were your expectations/hypotheses.

Methods You need to give some more detail on the processing of 454 sequences (may also be added to SI). How were 454 sequences processed and cleaned? How did you deal with pyrosequencing noise? What was the number of singletons (sequences occurring only once in the dataset)? Especially richness estimates may be flawed by pyrosequencing noise.

p. 17547, l. 21: check your reference of Schloss and Handelsmann; in the reference list you cite their MiniReview in Genome Biology, but you probably mean Schloss & Handelsmann 2005, AEM 71:1501 (DOTUR)?

p. 17547, l. 7: add reference for DISTLM.

Results Is there no information on bacterial numbers?

p. 17548, l. 21: it is not clear in all instances, what you refer to when you speak about "the two habitats", make sure to remind the reader again that you mean the slope and the basin.

p. 17548, l. 22: change: ..., more pronouncedly at the basin habitat..., into: ..., and were more pronounced for the basin habitat...

p. 17550, l. 10: For nematodes you find 30% singletons (only occurring at one station). Can you put this into perspective, in comparison to other studies?

p. 17551, l. 9-10: move the second part of the sentence to the discussion (and add reference for soil comparison). Be aware that technical differences in 454 analyses may also lead to differences in richness estimates (direct comparisons may be difficult).

p. 17552, l. 21-23: rephrase "meiofaunal major taxa community". ?

Tables The results include a large number of Tables. Tables may in general be informative, but I would suggest to design/adjust the information to be a bit more visual, e.g.

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highlight dissimilarities with color schemes or produce a real heatmap instead.

Table 1: give clear references to the expeditions (ship, cruise). Table 2 and 3: give definition/description of PERMANOVA and DISTLM again. Table 2: footnote 1, "design" instead of "desing" Table 4: Think about more visual representation here, e.g. color scheme according to dissimilarities. Figure 3: plots and legends are too small to read. Try to re-arrange so that they are easier to read. Figure 4: plots and legends are also too small. Re-arrange as 2x2. Did you test for linear trends? Were the results significant (R, p value)? Indicate a trendline in the plots (visual aid). Figure 5: Explain in legend what the "Overall" stars mean (I assume that it is the richness when both habitats are combined). Maybe choose a different sign, e.g. just a bar, to indicate the overall number of taxa. Figure 6: the diamond and the triangle sign at the bottom of the figure are not displayed correctly. Figure 7: better visualize what you want to state/highlight with this figure. Include colors or shapes to separate different depth or habitat groups.

Discussion p. 17554, l. 5: "particularly", instead of "particular"

p. 17555, l. 3-10: what are further possible explanations? What were the differences between the studies?

p. 17556, l. 11-12: Zinger et al. 2011 also includes deep benthic samples from the Mediterranean.

p. 17556, l. 12-21: be careful with absolute richness estimates (possible sequencing artifacts may influence them). May technical differences play a role in the large differences obtained? Comparison with further studies?

p. 17558, l. 2: "gradients" instead of "gradient". "..., it is now also widely accepted (or hypothesized) that the high complexity of the seafloor affects..."

p. 17558, l. 21: "... the only exception being THE eastern Levantine BASIN, ..."

p. 17558, l. 24: "..., the results of THE true richness estimator Jack1..." What do you

mean with "true"?

p. 17559, l. 5: "metazoa" instead of "metazoan"

p. 17559, l. 7-8: ... and we CAN therefore NOT preclude...

p. 17559, l. 25 – p. 17560, l.1: The source-sink hypothesis refers to source and sink of populations, where bathyal populations present the source and abyssal populations the sink. Rex et al. propose an increased significance of these dynamics with increasing depth and attribute it to the exponential decrease in organic carbon flux with depth. There is of course regions in the deep sea where, e.g., depressions may act as a sediment trap accumulating organic matter. But this is not what the source-sink hypothesis relates to! Also, earlier you argue that the food limitation could be a reason for the higher richness observed (here you now say that basins may act as food sinks and therefore lead to higher biodiversity).

p. 17560, l. 17: ... ranges FROM low to moderate values... Set numbers into context, what is high and what is low, what does it mean?

p. 17562, l. 4-5: please do not say variables, but rather richness and diversity or only diversity. Meiobenthic variables could be anything.