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## Interactive comment on "Simultaneous quantification of in situ infaunal activity and pore-water metal concentrations: establishment of benthic ecosystem process-function relations" by L. R. Teal et al.

## L. R. Teal et al.

lorna.teal@wur.nl

Received and published: 10 October 2012

## Anonymous Referee #2

In this study Teal et al. investigated sediment reworking rates and the distribution porewater metals in-situ in bioturbated sediments. They use state of the art techniques (f-SPI and DGT) to measure these parameters simultaneously. The time-lapse approach of fluorescence particle profile imaging allowed them to get insights in the temporal dynamics of particle reworking rates and link these to the distribution of Fe and Mn in

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the pore-water. The manuscript is well written and structured. I like the study, and I agree that such in-situ measurements are crucial to relate findings from laboratory experiments to the real world. In this context the present study is a very valid contribution to research focusing on effects of bioturbation on biogeochemical processes in benthic systems and fits into the scope of Biogeosciences. While the experimental study is sound, I have some major problems in the way how this study is introduced and discussed. While a lot of text is spend on describing that the interplay between organisms and the environment is complex (which results in text constructions like "context dependent effects of inter- and intra- specific behaviour and integrating species-environment interactions") there are hardly any concrete hypotheses that focus on the core of the study (e.g., why is trace metal cycling tackled other than that there are techniques to do it).

REPLY: The potential for sediment porewater associated trace metals to enter the water column will be augmented by infaunal activity, but the interaction between infaunal behavior and metal flux has received little attention. The variation of infaunal bioturbation over temporal and spatial scales is as an important determinant in whether a sediment is considered a source or sink for trace metals, but has largely been ignored in previous work. We accept however (as stated in response to reviewer #1) that we did not explain well enough in the introduction why we chose to study Fe/Mn alongside bioturbation and have added some additional text to address this shortcoming, hopefully clarifying better the objectives.

The fact that particle and pore-water mixing take place on very different time scales is true (e.g., Berg et al. MEPS 2001) but given the time-integrated and space-integrated approach of DGT used here, I do not see that this study yields new insights into the coupling between particle movement and fluid flow. In fact, the combination of high temporal imaging of "slow" particle reworking and time-integrated and horizontal averaged measurements of "fast" solute transport somehow contradicts the aim of the study, i.e., measurements at appropriate spatial and temporal scales.

REPLY: The referee highlights a key finding of our study, that in order to relate bioturbation to metal flux it will be necessary to measure fluxes at higher temporal resolution than is currently practiced. Here, we integrate over a number of days and make an attempt to account for the variability within a sediment community in order to detect underlying general patterns that we can quantitatively link. We are thus not attempting to couple particle movement and fluid flow, but link an ecosystem process (particle mixing) with a measure of function. Our bioturbation data represents >15,000x more information relative to a standard bioturbation incubation that resolves only the final time point at cm scales. This allows us to understand in much more detail what type of process in terms of particle mixing we are dealing with across the different deployment and therefore which faunal processes are occurring during the time-integrated DGT profile accumulation. Due to the high variability in the different bioturbation profiles seen, in fact our study highlights that longer deployments would be necessary to really achieve replication of the true bioturbation going on (e.g. including infrequent deep mixing events). However, longer deployments of DGT are likely to lead to problems with Fe/Mn supply. All these are difficulties we wish to highlight and although we cannot yet provide a solution to all, we are confident that our study represents a step-change in the amount of information quantified and that our findings highlight the inappropriateness of relating faunal-metal profiles using current methodology.

Recent studies by Stahl et al. (2012) and Zhu and Aller (2012) indicate the very heterogeneous (and to some extent the dynamic) distribution of porewater metals in bioturbated sediments and their findings would be helpful in discussing the results of this study.

REPLY: We thank the referee for highlighting these contributions, and have incorporated them into our interpretation in the discussion. Indeed, the heterogeneous nature of the sediment is why we increased the level of replication in our study in an attempt to still be able to detect consistent underlying patterns.

Specifically, I would like to see a much more detailed discussion of how bioturbation, C4610

including bioirrigation, may affect the distribution of Fe and Mn, under which conditions are they dissolved in the pore-water (and accumulate in DGT) or participate as particles, to what extent is their mobilization affected by oxygen, nitrate or sulfide etc. The study by Volkenborn et al. (2012) indicates the oscillatory nature of bioturbated sediment, so what would be the impact on e.g. Fe and Mn distributions, redox state and transport?

REPLY: We agree that we lacked some details on the mentioned aspects and have addressed this with additions to the intro/methods/discussion.

The authors state that they investigate trace metal cycling as a proxy of ecosystem functioning, but without a more detailed discussion of the underlying processes that may have contributed to the observed and modeled Fe and Mn profiles this study investigates pore-water metal distribution and not on their cycling. Consequently, I have strong objections is using Fe and Mn distributions as an example of "ecosystem functioning".

REPLY: Firstly, the referee objects to the use of metal cycling as an example of ecosystem functioning, but does not expand on why. Infaunal bioturbation (or microbial transformation) is firmly viewed as an ecosystem process that mediates trace metal concentrations, hence the latter is referred to as an ecosystem function. Multiple ecosystem functions (say, for example, nutrient and trace metal concentrations) collectively contribute to water quality which, in turn, would be referred to as an ecosystem service. This is standard terminology within the ecological literature and we are confident that are usage is correct. With regard to using Fe/Mn profiles and referring to cycling, our study cannot provide, and was not designed to, identify the detailed mechanistic processes the referee eludes to. Whilst we could provide opinion on the likely mechanisms, this would be nothing more than conjecture as we are not in a position to irrefutably test whether or not such conclusions are valid. We opted to avoid such controversy by focusing our study on whether we could find agreement between faunal activity and trace metal profiles. It should also be noted that we did not model Fe and

Mn distributions as the reviewer is suggesting, rather we used statistical approaches to describe the profiles that were quantified, which requires no assumptions about the chemistry. Additional text and reworking of some of the introduction and discussion is used to address these issues. We have adjusted the terminology not to refer to "cycling" but metal reduction and remobilisation, which is more accurate. Other additions to the text help to explain better how Fe/Mn profiles can be used as a measure of ecosystem function.

Indeed, bioturbation is complex and involves ecological, physiological, biogeochemical and physical aspects. To test if the use of "multiple technologies provide additional insights" is trivial, and I strongly disagree that "multiple technologies are seldom used together". I think that especially research on bioturbation has some very nice examples of interdisciplinary approaches and methodological integration.

REPLY: With respect, we strongly disagree with the referees assertion that the use of multiple technologies is trivial, routinely used together and provide few novel insights. We agree that there are several approaches that are available, but they are very rarely used together and particularly in situ studies are scarce. The vast majority of bioturbation studies are restricted to single methodology within the remit of the study. Where multiple methods have been used, as the referee eludes, there have been some significant advances made, which is exactly our position. Still some of these (such as Stahl et al. which the reviewer cites) remain in the methodological literature and are not applied to ecological questions. We have made sure to add the key references where multiple technologies have been used previously into the text as appropriate.

The introduction to some extent fails to introduce the core of this study. The same introduction could be used to introduce any other study as it largely dwells in very broad aspects on benthic ecosystem functioning and its complexity. E.g., what is the contribution of trace metal dynamics to organic mineralization processes? How is metal redox cycling affected by the presence of other chemical species? What are the spatial and temporal scales on which we need to tackle sediment reworking and bioirrigation?

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REPLY: The referee comments reflect a view point that desires a different focus for our contribution, which does not mean that our present focus is inappropriate or incorrect. Our focus was not on the details of the chemistry but on trying to establish a more quantitative link between the biology and the chemistry. We have, nevertheless, provided some additional text to incorporate more on the specifics of trace metal cycling and be more specific on our objectives.

Results: It would be useful to describe the study site a bit more in detail with respect to the sediment characteristics. Are there any data on grain size, organic content, permeability, topographic features (ripples, mounds etc)? Is it a diffusion or advective dominated system? If such data do not exist, at least a statement about the general character of the site (muddy or sandy) would be useful. The authors state several times that the outcome of bioturbation is context dependent. Sediment type is one, if not the most important context. So it would be useful to have some idea about the setting at the site.

REPLY: We have added a description of the sediment characteristics as suggested based on a recent paper from the same site and the evidence from the diver photos on topography. It should be noted that all the variables the reviewer lists will not differ much between the deployments, so the "context" setting of our measurements can be considered the same.

On the other hand information about changes in water depth due to tides is largely unnecessary. I guess the authors mention these changes in water depth because they discuss tidal flushing as a mechanism that may have affected pore water metal distributions. Pore-water advection in permeable sediment is driven by pressure gradients. Can the slow raise and fall in water level really affect pore-water flow? How strong are tidal currents close to the seafloor? What about wave action? Again, more information about the study site character would be useful.

REPLY: We included information on changes in tidal height because species behavior

is known to change with the tidal cycle (see refs provided) and it is important that our study included the full tidal cycle and integrated across these effects. We do not think that these changes will affect pore-water flow, and have not suggested that they do. Due to the sheltered nature of the Loch wave action can also be considered minimal at the seabed.

Table1: If there was 1 individual in one of the six 100 cm2 cores this would correspond to an average abundance of 16.6 ind m-2. So how is it possible a minimum abundance on 5.31 was found for several species? Please clarify.

REPLY: We thank the reviewer for picking up on this inaccuracy. The original plan was to use cores but due to technical problems we switched over to a grab. This had not been adjusted in the methods, but leads to a different sampled area per grab and should now be consistent with the numbers in the table.

Also, on page 8550 Line 26 the authors state that Amphiura and Terebellidae were consistently found across samples. Being present in three out of six samples is not consistent.

REPLY: We agree that this statement does not reflect well what is shown in the table and have reformulated the paragraph.

Page 8551 Line 8: does this not also, and more importantly, suggest spatial differences in species composition? And thus a problem of insufficient replication, especially if the aim is to derive implications for processes on larger spatial scales?

REPLY: The deployments all take place within a 50m radius and we do not expect differences in communities at this spatial scale at the study site. All species are mobile and particularly the epifauna (which cannot be sampled well with benthic sampling methods) is likely to be in constant movement and therefore appear in some deployments but not in others. We argue this has more to do with the time of the deployment rather than the absence of these species at the site of one deployment compared to another.

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Rather than spatial effects we would argue that given a longer deployment time, more species would "pass by" the SPI and the deployments would become more similar in terms of the bioturbation activity observed. Our study in fact represents a significant increase in the level of replication compared to other in situ bioturbation studies and our findings incorporate variance and error within the profiles, something that is seldom achieved in studies of a similar nature. Whilst replication can always be increased, the argument that we have insufficient replication is misguided.

Page 8552 Line 2: construction of a deep burrow does not really sound appropriate, maybe better 2 cm deep mixing (just as on page 8551 line 15)?

REPLY: The observations of mixing between the two deployments differ. In sequence one the deep mixing is caused by a sudden injection of luminophores to 2cm depth by what looks like the "walking" motion of the crab over the sediments surface, injecting luminophores to depth with its legs. In sequence 3 it is actually a crab "digging a big hole" in the surface sediment... we have tried to reflect these different mechanisms in the text but would refer reviewer and reader to the image sequences (now available online, link added). We reworded to "burrowing activity" rather than burrow construction.

Discussion: Page 8556 L 15-20 To link particle and pore water mixing, DGT is not really appropriate, because it time-integrates and horizontally averages pore-water metal distributions.

REPLY: This assertion would be correct if we were examining specific instantaneous relationships between particle movement and pore water concentration at a specific (porewater space) location. However, our focus was at a much broader scale and because particle mixing occurs at a slower rate than porewater mixing, it is necessary to use a technique that time-integrates. As we were able to integrate bioturbation over similar time scales using time-lapse imaging, the two measurements were spatially and temporally matched, but the value of the time-lapse sequence allows us to better interpret the bioturbation mechanisms encountered. Despite this attempt at time-integration

we still encounter considerable variability and this where other processes not measured come into play, as discussed.

Insights from microsensors and planar optode studies should be discussed (Wenzhoefer and Glud 2004; Timmermann et al. 2006; Stahl et al. 2012; Zhu and Aller 2012; Volkenborn et al. 2012). While DGT is an elegant way to characterize pore-water solute profiles in-situ, it does not allow characterizing the spatial-temporal dynamics which are characteristic for bioturbated sediments. In a strict sense, the present study is an example of measurements on an inappropriate spatio-temporal scale (Page 8544 L 23ff) to link fluid flow and particle movement. A broader discussion of ways to tackle these time scales would be useful.

REPLY: We have incorporated some of the references (not all, due to a rather extensive reference list already) that the referee suggests where generic arguments can be made, but we have not extended the Discussion to reviewing other technologies as that was not the focus of our manuscript (there are also many such reviews in the literature comparing the merits, or otherwise, of alternative technologies; e.g. Santos et al. 2012 recently identified 12 drivers of porewater advection in permeable shelf-sediments and concluded that no single technique can be used to study advective flow in permeable sediments). For this reason, and as the referee suggests, we will use a variety of techniques in the field. The point the referee makes is in line with our argument that multiple approaches must be used, whilst our conclusions indicate that other technologies capable of much finer resolution may be more appropriate for resolving highly spatio-temporal changes in biogeochemistry. The key discussion is in fact at which spatial/temporal scales does we need to measure processes to address the specific questions asked. In our case where our focus is on broader scale ecological questions and links between biology and function, we need some integration to smooth out some of the variability whilst incorporating enough resolution to describe and interpret correctly the underlying processes.

Overall I like the study, the in-situ approach, and the methodology, but from my point C4616

of view this paper could gain a lot if the introduction would be more focused towards the aim of this study and expand on metal biogeochemistry. The discussion of results should include some of the shortcomings of time-integrated approaches to study pore water dynamics and integrate some of the recent literature on the dynamics and heterogeneity of pore water chemistry in the presence of bioturbating organisms.

Interactive comment on Biogeosciences Discuss., 9, 8541, 2012.