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Biogeosciences Discussions

Interactive comment on "Impacts of exotic mangrove forests and mangrove deforestation on carbon remineralization and ecosystem functioning in marine sediments" *by* A. K. Sweetman et al.

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We would like to thank Drs. M. Huxham and E.Kristensen for their constructive reviews.

Please note that in the original ms. the bacterial data for site KBR was incorrect. We have amended the bacterial biomass data and bacterial C-uptake data in figures 4 & 8 and table 3. Our conclusions still hold for sites PHM, PHR, PHC and KBC, and all data with the exception of the bacterial biomass and C-uptake data-set for KBR. Bacterial biomass and C-uptake rates in sediments from the KBR site were significantly greater

than in the KBC study and this adds extra support for our overall conclusion, which is that mangrove invasion and removal continues to impact benthic ecosystem functioning for at least 6yrs after removal of above sediment mangrove biomass. We have modified the discussion and abstract accordingly.

Please find our comments to the reviews by Drs. M. Huxham and E. Kristensen below:

Responses to comments from Dr. M. Huxham:

Reviewer comment # 1 re. study sites: We have adjusted the methods section 2.1 to better describe why the control sites were un-colonized by mangroves. This has been done by stating that 'Despite being suitable habitats for mangrove colonization, both control sites were free of mangroves because of active seedling removal programs in the State of Hawaii.'

Reviewer comment # 2 re. incubations: The presented SOC rates are neither from dark or light incubations. They are mean rates ($\pm$ SE, n = 3) calculated from average SOC rates measured from each algae-amended chamber over each 48-hr experiment. We have now clarified this and modified methods section 2.3 by stating in the last sentence that 'The presented SOC data are mean rates ($\pm$ SE, n = 3) calculated from average SOC rates measured from each algae-amended chamber over each 48-hr experiment.' . Reviewer comment # 3 re. number of chambers: Regarding table 2, n = 2 refers to cases in which fauna were enumerated from 3 chambers, but a replicate sample from 1 chamber was lost prior to IRMS analysis, so that only samples from 2 replicate chambers existed for biomass estimates. As such, a range was calculated for biomass estimates of a given taxon from 2 replicate chambers. With regards to table 3, a total of 3 chambers were amended by adding labeled algae. However, despite the fact that PLFA samples were sampled from all amended cores at the end of each experiment (i.e. PLFA samples = 3), some samples often possessed too little sediment for a suitable quantity of FA to be extracted, which meant that PLFA samples could only be analysed from 2 chambers in a number of cases (e.g. the PHM and KBR

sites). In the case of DIC, sometimes bubbles existed in some samples, which meant that DIC production rates could not be calculated for 1 chamber, so a DIC production mean and range was calculated from only 2 chambers (i.e. for the PHR and KBR experiments). Regarding table 4, in a number of cases only 2 replicate chambers experiments contained a specific taxon for IRMS analysis. Therefore, a mean $\pm$ range was calculated as the taxon was not available in the third chamber so a mean C-uptake rate $\pm$SE could not be calculated. We have modified the legends to tables 2,3,4 as well as appropriate figure legends to clarify this.

Reviewer comment # 4 re. normalised data: In order to explain the rate normalization procedure, we have adjusted the end of methods section 2.4, at the request of the reviewer, so that it now reads 'Nevertheless, because different amounts of algal-C were added to the KBC study compared to all the other experiments as previously stated, total DIC efflux, daily DIC production from added algal-C and C-uptakes rates by fauna and bacteria have been normalized by the amount of algal-C added (g C m-2). As such, all DIC production and C-uptake rates are given in units of mg g C m-2 d-1.'

Reviewer comment # 5 re. normalized macrofaunal C-uptake. All the rates originally presented in table 4 were normalized to algal-C added, but not to biomass. At the request of the reviewer, we have added in 5 new columns with macrofaunal C-uptake data normalized to both algal-C added and faunal biomass. We have also discussed this data in results section 3.5 by stating 'When C-uptake rates were normalized to biomass, corophiid amphipods were responsible for the highest C-uptake rates measured (Table 4). Interestingly, mean C-uptake rates (normalized to biomass) of spionids and oligochaetes fluctuated as a function of sampling site with spionid and oligochaete C-uptake rates being higher in both control experiments relative to the PHR and KBR studies (Table 4).' Furthermore, we have discussed this data in the discussion, where we state that 'In terms of C-uptake rates normalized to biomass, the apparent increase in response to labile C by similarly related taxa (e.g. oligochaetes, spionids, see Table 4) in the control studies, relative to the removal sites, may have resulted from lower

faunal densities in control sediments (see Table 2) stimulating deposit feeding as seen in previous studies (Miller and Jumars, 1986, Wheatcroft et al. 1998)...'

Technical corrections have all been done. For example, 'densities' have been changes to 'abundances', 'Muxham' has been changed to 'Huxham'. Total C was calculated as in Middelburg et al. 2000 and this has been added into the legend for Table 1. Furthermore, 'unknown taxa' have been clarified as 'unidentified taxa (i.e. poorly preserved fauna)' in results section 3.3. Please note that the unknown taxa abundances in table 2 have been re-calculated to not include animal fragments. As such, the abundances are significantly less than in the original table 2. Legends to figures and tables have also been amended at the request of the reviewer.

Responses to comments from Dr. Erik Kristensen:

Overall reviewer comment re. the use of surface deposited 13C labeled labile algal-C to quantify ecosystem functioning. We have modified methods section 2.2 by stating that 'A non-axenic clone of the green alga Chlorella spp. (Chlorophyta), initially sampled off the coast of Hawaii, was used as a food source in our experiments. Whilst this labile C-source is physically and biochemically distinct from relatively refractory mangrove material, labile algal-C is known to continuously enter and support mangrove ecosystems through natural processes including phytodetritus deposition and benthic microalgal production (Bouillon et al. 2008, Oakes et al. 2010). This type of addition is therefore not entirely artificial and allowed us to realistically trace the fate of labile algal-C in mangrove type environments and therefore, quantify various aspects of ecosystem functioning in the sites studied.' Furthermore, we have modified the manuscript and now emphasize that the introduction of algae was to the surface of the sediment, so this portion of the experiment is primarily addressing near surface labile carbon cycling. This can be seen in the introduction where we have written 'Because of the relatively short duration of each experiment (48-hrs) and the addition of algal-C to the sediment surface, our experiments primarily addressed near- surface processes, and tested the following hypotheses:...' We have also stated in section 4 that 'There are limitations

associated with this case-study as a result of the short duration of each experiment (i.e. only 48-hrs) and algal-C only being added to the sediment surface. These limitations therefore only allowed clarification of differences in near-surface processes in the different sediments studied. However, the experiments also revealed dramatic differences between mangrove, mangrove removal and control sites in the depth distribution of labeled C-uptake by fauna and bacteria. Therefore, our results collectively show that major aspects of ecosystem function in sediments from an invasive mangrove forest can differ from those in un-invaded habitats, as well highlight that ecosystem functioning in sediments from mangrove removal sites can differ substantially from those in control sites 2 - 6 yrs after above-sediment mangrove removal.'

Reviewer comment # 1 re. the use of macrofauna as a term. The criticism that we are not working with macrofauna is not justified in our opinion. Five hundred micron sieves are a widely accepted cut-off for macrofauna (e.g., Demououlos 2004, Demopoulos et al. 2007, Demopoulos and Smith, 2010) because a 1 mm sieve misses many of the "macrofaunal taxa" (including many polychaetes) in habitats with finer sediments, including mangroves. Furthermore, we identify all the major taxa in our abundance and carbon uptake data (Tables 2 and 4) and the "meiofaunal taxa" (e.g. ostracods, nematodes) contribute little to the community abundance or C-uptake. Spionid, sabellid and cirratulid polychaetes, or amphipods (all "macrofaunal taxa") account for most of the abundance and uptake. Thus, using the term "macrofauna" is fully justified, and the reader can choose to remove the "meiofaunal taxa", should they so desire.

Reviewer comment # 2 re. manuscript title – Most of the structure and function measurements are not dependent on the labeled algae introduction (e.g., root biomass, oxygen consumption, macrofaunal and bacterial biomass), so these measurements do not suffer for a near-surface focus, as suggested by the reviewer. Thus, we are not in favor of altering the title.

Reviewer comment # 3 re. Table 1. The sentence on sediment characteristics has been moved to lines 223-224 of the revised ms.

Reviewer comment # 4 re. whether sediment cores were collected from permanently inundated locations. We have revised the ms. and now state in lines 153-155 that 'Subtidal sediments from all Pearl Harbor sites were collected from approximately 15-cm water depth at low tide, and those collected at the PHM site were sampled between prop roots at a distance of approximately 0.5-m from individual emergent roots.' However, we highlight that cores from Kaneohe Bay were from an exposed location by stating in lines 155-157 that 'Intertidal sediment cores from both Kaneohe Bay sites were collected along a 15-m long transect line at an identical tidal elevation above the low tide mark at low tide.' The line 'ensuring that all cores were subjected to the same degree of air exposure' has been deleted.

Reviewer comment #5 re. delta 13C signature of algae. We enriched the algae in 13C by growing the algae in an artificial medium modified by replacing 25% of 12C bicarbonate with 25 % NaH13CO3. This is a well-known, often-used procedure, and the method used to label Chlorella is described in methods section 2.2.

Reviewer comment #6 re. how much Chlorella was added to each chamber. To enhance clarity, we have amended the methods section 2.2 and have inserted the sentence 'Approximately 1.8 g algal-C m-2 was added to each experimental chamber except to cores collected at the KBC site, where $\sim$1.6 g algal-C m-2 was added' between lines 178-180. Furthermore, lines 281-286 have been modified by stating that 'The amount of algal C added to each experiment contributed only 0.2 – 2 % to the C-standing stock in the top 5-cm of sediment (85 - 929 g C m-2, based on data in Table 1). Nevertheless, because different amounts of algal-C were added to the KBC study compared to all the other experiments as previously stated, total DIC efflux, daily DIC production from added algal-C and C-uptakes rates by fauna and bacteria have been normalized by the amount of algal-C added (g C m-2). As such, all DIC production and C-uptake rates are given in units of mg g C m-2 d-1.'

Reviewer comment # 7 re. inclusion of total DIC efflux data. We have now modified table 3 and now include mean total DIC efflux data ($\pm$ 1SE) for each site. Furthermore,

this data is discussed in methods section 3.5 in line 356-358 where we have written 'Between 71 and 90 % of the processed algal-C was found in the DIC pool after 48-hrs (Table 3), and the mean production rate of DIC from algal-C accounted for between 4 – 12 % of the mean total DIC efflux rate in all studies, assuming a molar mass weight for C of 12 (Table 3).' This data is also discussed between lines 416-424 in the form 'Nevertheless, production of DIC from the added phytodetritus accounted for between 10.59 $\pm$ 1.49 (range, n = 2) to 10.91 $\pm$ 0.26 (SE, n = 3) mmol C m-2 d-1 in the mangrove and removal site experiments and 7.67 $\pm$ 0.71 (SE, n = 3) to 12.25 $\pm$ 0.28 (SE, n = 3) mmol C m-2 d-1 in the controls, based on a molar mass weight for C of 12. Assuming a respiratory quotient between DIC and O2 of 1 (Middelburg et al. 2005), this transfer of algal-C into respired DIC corresponded to approximately 10 to 17 % of the SOC (or 4 - 12 % of the mean total DIC efflux) in the mangrove and removal site experiments compared to 18 to 30 % of the SOC (or 3 – 8 % of the mean total DIC efflux) in the PHC and KBC site studies. These results highlight the very labile nature of the added algal C-source and suggest that the measured SOC and total DIC efflux values may be higher than background values without algal addition.'

Reviewer comment # 8 re. POC not being measured and SMB used as a proxy for POC content. We measured POC and SMB content in sediments from the unamended chamber and found that they correlated very closely (r = 0.967, P = 0.007, n = 5). This therefore allowed us to use SMB as a proxy for POC content. This is stated in lines 221-223.

Reviewer comment # 9 re. dissimilarities in tidal elevation – We have amended 153-157 to clarify what we mean by dissimilarities in tidal elevation. However, we have highlighted this further between lines 289-294 by stating that 'Because of seasonal differences in temperature when the Pearl Harbor and Kaneohe Bay sites were sampled, as well as potential artefacts from dissimilarities in tidal elevation affecting comparisons between locations (e.g. subtidal vs. intertidal characteristics and processes quantified using sediments from Pearl Harbor and Kaneohe Bay sites, respectively), differences

among variables between Pearl Harbor sites were analysed separately from those in Kaneohe Bay using a one-way ANOVA test followed by a Tukey post-hoc test.'

Reviewer comment # 10 re. high abundance of macrofauna – See response to reviewer comment # 1.

Reviewer comment # 11 re. total DIC efflux data – See response to reviewer comment # 7.

Reviewer comment # 12 re. transfer of algal-C to bacteria at the sediment surface – We have now inserted the text 'The addition of labile algal-C to the sediment surface in each experiment was, in all likelihood, one of the main factors driving the high algal-C uptake rates by bacteria at 0-2cm in each experiment.' between lines 454-456 as suggested by the reviewer.

All corrections have been made. Reviewer comment re. reference with no author – No authors were found on the report so authorship could not be quoted. We have decided to keep figures 7 and 8 A, B, and D. Whilst these do show the same data as seen in table 3, post-hoc test results can be depicted much easier on the graphs, and the data in these graphs can be easily compared to C-uptake rate versus sediment depth data in figures 7 and 8C and E.

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