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Interactive comment on "Technical Note: Sampling and processing of mesocosm sediment trap material for quantitative biogeochemical analysis" *by* T. Boxhammer et al.

Anonymous Referee #2

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"Technical note: Sampling and processing of mesocosm sediment trap material for quantitative biogeochemical analysis"

Boxhammer, T., Bach, L. T., Czerny, J., Riebsell, U.

The authors present a technical note on a new method for sampling of settling material from mesocosms as well as how to best process the collected material. This note is part of the special issue "Effects of rising CO2 on a Baltic Sea plankton community: ecological and biogeochemical impacts". The manuscript is well written and clearly presented and would be of value to the scientific community working with mesocosms. There are a few issues that are not addressed in the manuscript and would be impor-

tant to address when studying settling particles within mesocosms. The first and most obvious issue is how the authors have dealt with growth on the sides of the mesocosms? This is not mentioned in the manuscript and may have a large impact on the estimates of export from the mesocosms. Were the sides cleaned or the material left to grow – did the authors estimate that growth at the end of the experiments or were any measures taken to avoid such growth? Have the authors made any tests of the oxygen consumption of material captured in the collection cylinders at the bottom of the mesocosm? Would this material go anoxic before sampling on either daily or every second day? Anoxic conditions could have important implications for the biogeochemical measurements of the settled material, e.g. a build-up of CO2 could cause dissolution of calcium carbonate and other nutrient cycling could take place (e.g. anoxic steps of the nitrogen cycle). Some more specific points would be very useful to include in the manuscript and are also mentioned in the specific comments. These are issues of particles sticking to the sides of the funnel as they slide down to the collecting cylinder at the bottom of the mesocosm and if the flow rate of the water in the silicone tube connecting the collecting cylinder to the sample bottle is high enough to ensure collection of particles with high sinking velocities (see specific comments). Finally, why did you decide not to poison the sampled material during the sedimentation and centrifugation procedure, would this not have limited further degradation and allowed for longer sedimentation periods?

I find that the manuscript will be a valuable contribution to the special issue and would suggest it publishable after revisions, see the specific comments below.

Specific comments:

Page 2, Line 4: With “these two processes”, I guess you refer to particle flux and particle formation, maybe write the processes out to avoid confusion.

Page 4, Line 18-19: Often marine snow and other aggregates are very ‘sticky’ and adhere to surfaces, did you test if they aggregates did slide down the funnel surface.

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Page 4, Line 20: Do you mean the tip or the bottom of the collecting cylinder?

Page 4, Line 23: How long was the tube that connected the bottom of the mesocosm to the Schott Duran glass bottle? The KOSMOS mesocosms vary in depth between 15 and 25 m, is that including the funnel below the mesocosm? If the tube was 25 m plus a bit extra so likely around 30 m long? This means that there was around 3 L of seawater in the tube itself. In addition, the collecting cylinder contained 3.1 L of seawater, which means that the total water volume in the tube and in the collecting cylinder made up 6.1 L while the Duran Schott bottle only collected 5 L of water. Was this enough to ensure that all aggregates were collected?

Page 5, Line 1-3: Have you calculated or measured the water flow in the tube? This needs to be more than the settling velocity of the collected aggregates. These can potentially sink with several hundred meters per day. Some ballasted aggregates and fecal pellets have quite high sinking velocities (e.g. Bruland and Silver 1981, Iversen and Robert 2015, Ploug et al. 2008), though most are likely around 100 m d-1. Did you calculate what your theoretical flow rate was and have you considered if any potential boundary effects potentially would make you lose some particles?

Page 5, Line 3-5: Did you typically discard the volume contained within the silicon tube before sampling, e.g. 3 L for a 30 m long tube?

Page 5, Line 7-10: It seems unlikely that the integrity of the particles were preserved during the sampling. First you collected the particles in the collection cylinder where they would land on top of each other after rolling down the sides of the funnel. Already here you have changed their size and structure. Thereafter they are pumped up a long tube and finally flushed into a Duran Schott bottle. Even if this is gently done, it will still affect the aggregates, especially marine snow, fecal pellets might survive the procedure. However, it is not important for your study to preserve the size, shape, and structure of the aggregates, since you are interested in chemical analysis, so I would suggest to remove this sentence from the manuscript.

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Page 5, Line 11-14: There would be several issues by using the particles collected in this way to measure the particle sinking velocity and microbial respiration rates if you assume that the particles are the same as those formed and settling within the mesocosm. This would need some direct comparisons of aggregates collected within the mesocosm to the ones collected with the method described in this manuscript. However, as long as we are aware of the differences and changes made to the particles collected here, there is still much valuable information to be made from measurements of the particles collected here, as long as they are well characterized at each sampling point in terms of composition and type for instance.

Page 5, Line 15: When did you decide that you had collected all the aggregates? 1-4 L of particle suspensions seems rather low for the 1 L, but maybe you stopped when no more particles were observed after a certain time or a certain water volume?

Page 5, Line 16-18: Consider to point out that this subsampling is not the one used to do the biogeochemical parameters, but subsampling for other measurements and that you are keeping this low in order to be able to have reliable chemical measurements from the total flux of particles. Did you measure the precise volume of the 'pre-subsamples'?

Page 6, Line 4: Why do you use the term total particulate carbon? Was this because you did not remove inorganic carbon (calcium carbonate) with hydrochloric acid?

Page 6, Line 9-10: How did you know that the copepods were alive if they were on the filter? Did you do this step immediately after filtration or after freezing?

Page 6, Line 22: Did you calculate what the slowest sinking velocity would be for the settling particles reaching the bottom of the bottle? If the bottle was 20 cm tall, then particles sinking with velocities slower than 2.4 m d-1 would not make it from the top of the bottle to the bottom within 2 hours, assuming that the bottle was full. Try to calculate this and see what the slowest velocity would be, some single phytoplankton cells settle with around 1 m d-1. This might give you an idea of what the carbon sources for the

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supernatant could be.

Page 9, Line 27-29: Do you think the improved concentration efficiency of the FeCl3 in comparison to the passive settling and the centrifugation was due to loss of CaCO3 from both the sediment and the supernatant?

Page 12, Line 8: Change to "...macroscopic structures were visible after...".

Section 2.4: How did you do the quantitative measurements of the chemical parameters of the grounded material? Did you weigh the total mass of all the grounded material before taking subsamples from it?

Page 12, Line 9: Change to: "...diatom frustules became detectable...".

Page 12, Line 15-18: It still remains to show that particles are not stuck to the sides of the funnel when they are sliding down inside the mesocosm. In addition, it would be good to estimate the flow rate of the water within the tube leading from the bottom of the mesocosm to the collection Duran Schott bottle at the water surface and test if there are shear or boundary effects affecting the transport of particles through the silicon tube. Finally, was the water volume collected in the Duran bottle enough to sample all the particles in the collecting cylinder at the bottom of the mesocosm?

Page 13, Line 17-18: Would the simplest method to use in the field not be the passive settling? It seems that a longer settling period would increase the efficiency of collecting the settling material at the bottom of the bottle?

Page 13, Line 25-26: Do you have a reference or some tests showing that the precipitation of phosphate to particulate phosphorous is negligible?

Page 14, Line 3: For me it seems that there are many issues with the addition of FeCl3 to the sediment sample? Decrease of pH, precipitation of phosphate, addition of iron, and interference with spectrophotometric analysis?!? Would this method not be best to avoid?!?

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Section 3.3: It would be more informative if you could show some comparisons to measurements done on both non-ground and ground samples? For instance from cultures or other test samples? Are you likely to lose any material during the grinding process?

References:

Bruland, K. W., and M. W. Silver. 1981. Sinking Rates of Fecal Pellets From Gelatinous Zooplankton (Salps, Pteropods, Doliolids). Marine biology , Heidelberg 63: 295-300.

Iversen, M. H., and M. L. Robert. 2015. Ballasting effects of smectite on aggregate formation and export from a natural plankton community. Mar. Chem. 175: 18-27.

Ploug, H., M. H. Iversen, and G. Fischer. 2008. Ballast, sinking velocity, and apparent diffusivity within marine snow and zooplankton fecal pellets: Implications for substrate turnover by attached bacteria. Limnol. Oceanogr. 53: 1878-1886.

Interactive comment on Biogeosciences Discuss., 12, 18693, 2015.