

Interactive comment on “Carbon Flux Explorer Optical Assessment of C, N and P Fluxes” by Hannah L. Bourne et al.

Anonymous Referee #2

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SUMMARY

In this manuscript the authors address a current critical research field aiming at better estimating the Biological Carbon Pump (BCP) in the ocean by the use of autonomous in situ floats. These devices allow particle flux observations at very high spatio-temporal resolutions essential to capture the rapid ecological changes responsible in a large part for the BCP efficiency variations. In particular, this study targets a calibration between a proxy of particle concentrations in the water column, the volume-attenuance (VA) measured with a Lagrangian float-deployed imaging sediment trap, the Carbon Flux Explorer (CFE), and particle bulk chemical composition in Particulate Organic Carbon (POC), Particulate Nitrogen (PN) and Particulate Phosphorus (PP) measured on the same particles previously imaged and collected with a novel particle sampler

C1

added to the CFE (the whole instrument being named CFE-Cal). The ultimate goal of this calibration is an accurate estimation of element fluxes directly from particle imaging which thus would offer large potential in term of flux data collection which are still today and despite intensive efforts poorly spatially and temporally resolved. After detailing thoroughly the material and methods employed for particle imaging, collection and analyses the authors present results from 15 deployments of the CFE-Cal which lasted 18 to 24 hours near 150m depth in four different locations in the California Current system selected for their contrasting primary productivity features. Results show good correlations between particle content in C and N (but not P) and VA, promising perspectives of using this autonomous in situ imaging to estimate the fluxes of these elements. Each result is discussed (Results and Discussion grouped in the same section) and focus is put on results not meeting authors expectations or not agreeing with the literature. For the results that deviate from expectations, the authors suggest possible explanations from either material malfunctions or the characteristics inherent to the different environment sampled.

GENERAL COMMENTS AND RECOMMENDATIONS

This manuscript is well-written and leaves the reader with the general impression of a solid piece of work. Each section is correctly articulated and information are in general presented where they are expected. Overall, the figures and table shown are clear and deliver well the message intended. The objective tackled here is with no doubt one of the main current and future challenges in BCP research studies (converting particle flux from in situ imaging to biogeochemical fluxes) and I am always pleased to read about work that try to push further our methods to measure these complex and very dynamic ecological processes that drive the BCP with technical innovations. Even if not realising a major advance in the field and presenting results that could be argued, especially in term of potential bias, dataset size and finding significance – this study is worthy being published in BG because it is an attempt to a step forward and will certainly interest the research community working on ocean particle fluxes. However,

C2

even if I acknowledge the work done, its quality and how it is presented I have some concerns about this manuscript that lie mostly on a lack of details about the limitations of the method employed, that also reflect in the results, discussion and the general conclusion made by the authors.

To obtain a good conversion from particle images to POC, PN and PP content, two key parameters have to be carefully considered: (1) the conversion from particle 2D images obtained by the CFE to their 3D volume (detailed in Bishop et al., 2016). Briefly, in Bishop et al. (2016), aggregate (including those of phytodetrital and fecal origin) volume was inferred from cross sectional area converted to equivalent circular diameter and then to volume using an empirical relationship between aggregate thickness and their equivalent circular diameter reported in Bishop et al. (1978); (2) The conversion from particle volume to their chemical content. For that Bishop et al. (2016) used a published value for aggregate dry-weight density (0.087 g cm³; Bishop et al., 1978), and an estimated fraction of organic matter of 60 % in total dry weight. Finally, Bishop et al. (2016) uses an OM:C ratio of 1.88 to convert the estimated OM weight to POC.

The authors highlighted clearly the problem of using these literature-based calibration factors as they are often applicable only in the limited spatio-temporal context of their formulation. But I hardly understand the aim of the present calibration if it is not to finally succeed at reconstructing the flux from images alone and from a large range of environment and ecosystem structures. From the way it is currently presented, the manuscript suggests that the authors are trying to establish a library of relationships between VA and C, N, P contents. If it is the case it should be clearly stated. I would have found very interesting to see in the discussion and conclusion sections some perspectives on how to improve the calibration presented here. In particular, the combined acquisition of particle images and measurements of POC, PN and PP done here offers the great opportunity of estimating the quality of a traditional flux reconstruction (i.e. by inferring its value from the images using published volume to organic contents conversion factors as done before) by comparing it to the real values measured here (as

C3

explored in Estapa et al., 2017). I assume that the final goal is to estimate the POC flux from images alone and much work is yet to be done by the community to understand how to translate small differences in image detection to potentially large differences in chemical contents. A comparison between the calibration method developed here and other methods that try to convert images to elemental fluxes should have been made. The use of Polyacrylamide or Cryogels sediment traps to collect particles and then use image analysis and published values of organic content to convert the images to fluxes is a very close approach to this study. The major advance that the present work could have brought is by extending the use of particle images to push further the estimation of their organic content from the image analysis. It is a bit disappointing to finally realise that this study has the great potential of presenting both the images and the "true" values of their content usable to further our understanding of observed discrepancies but that unfortunately this opportunity was not seized by the authors.

Also, the limited number of results obtained due to device malfunction or inherent to the properties of the particle flux collected (i.e. presence of swimmers), or the corrections of POC, PN and PP values obtained from the CFE004 dives due to a discrepancy between images and sampling, should have led the authors to much more caution in their conclusion. Instead, the authors claim "strong calibrations" between the VA and POC-PN contents for a dataset on which many values have been removed or multiplied by an empirically-determined factor; in this case the 1.45 times factor representing the difference of abundance of ovoid pellets in the sampler and from the images.

Based on all these general remarks, I still recommend this article for publication in BG but after significant changes have been made to the Results, Discussion and Conclusion sections and substantial evidences provided where required. In particular, I strongly advise the authors to focus on the general issues mentioned above and summarised as follow: (1) Add to the manuscript a comparison with other techniques of image conversion to biogeochemical fluxes (e.g. gel sediment trap analyses). (2) Use the dataset presented to explore further the known discrepancies between image anal-

C4

ysis and inferred organic content. The authors could investigate if a reconstruction of the fluxes measured by the sampler here would be feasible by using the corresponding images and by applying various volume to chemical content relationships to the different particle types identified (e.g. different relationships for fecal pellets, marine snow, etc.). New insights informing on why we struggle at inferring the flux from images would certainly increase significantly the impact that this manuscript will have on the research community. (3) Depending on the modifications made after (1) and (2), moderate if needed the stated significance of the results and discuss it more objectively and into details. Especially, the term "strong correlation" can hardly be used with such confidence knowing that the dataset has been trimmed and partly multiplied by an empirically-determined factor, and that authors seem themselves unsure about potential contaminations of their samples.

Additionally, below are more detailed comments on the manuscript including technical and typographical corrections that will need particular attention before publication. I advise the authors to give a special attention to the four questions/comments below marked with an asterisk (*) as their response should influence the final decision for publication.

DETAILED COMMENTS

Page 1, Line 17: please add to the R2 the size of the sample included in the fit (n) and the p-value. Same line: "...was not sensitive to environment or classes of particles sampled." I assume this statement is used as a proof of applicability of the current calibration to many different ecological contexts. But, it could also suggest that the environment where the deployments were made was not contrasted enough for this calibration.

Page 1, line 21: a space is missing between "Approximately" and "10".

Page 3, line 11: change "our 2.8 conversion factor" to "the 2.8 conversion factor obtained by Bishop et al. (2016)". I understand it is the same team but "our" would mean

C5

a factor inferred in the current study and it is not the case.

Page 4, line 9: the glass stage appears quite small and subjected to overload if a single cm-sized particle (or a few mm-sized particles) happened to enter in the trap. What is the diameter of the opening?

Page 4, line 13: The time of ~25 min seems critical. Is there a threshold at which volume attenuation can be biased by particle overload (particles accumulating over previously deposited particles on the stage)? How did the author choose this time and the time of ~1.8h mentioned below (line 14) as it seems dependent upon the amplitude of the particle flux at the time and depth of the deployment?

Page 6, line 5: do particles larger than the size limit of 3 mm can get stuck inside the openings?

Page 6, lines 7-8: I assume the CFE-Cal has not yet been used for trace metal studies then (this intended use is mentioned above in the manuscript)?

Page 7, line 3: peri slides. Please correct.

Page 7, line 16 and below: why giving results in the Material and Methods section?

* Page 7, lines 19-22: this will need clarification as it seems to be a very serious issue. How could the process blanks be higher than the samples themselves even in case of accidental collection or contamination? Over the 6 replicates of process blanks, how many were contaminated? How did the authors deal with this issue as blanks have to be subtracted from sample values? Are the negative values on Fig. 5A a result of this correction?

Page 7, lines 24-25: "... which we assume is based on sample heterogeneity". Do the authors have evidence to support this assumption?

Page 8, lines 24-25: please mention what would have been the total number of samples in case of no malfunction or swimmers and give a percentage of "fail". My point is

C6

that it is hard to estimate the robustness of the CFE-Cal without a proper estimation of its percentage of fail (how many successful dives/samples over the total number intended?).

Page 8, line 27: this is not really a measure of "collection efficiency" (only assumed) but more a measure of transfer efficiency between the imaging stage and the bottles.

Page 8, line 29: "...close...", please give a precise number.

* Page 8 line 30: again this is a very worrying result that needs more investigation as it suggests a real issue with the collection and/or transfer method employed.

* Page 9, line 1 and lines 3-4: the authors first state that they do not fully understand the issue and then claim to have addressed the problem by solving a software issue. Please bring clarification on this.

Page 9, line 12: what does this time of 2 minutes sample collection time refer to? (how is it different to the ~25 min imaging sequential time?). Is it the duration of particle transfer to the bottles?

Page 9, line 17: please add the sample size (n) and p-values for each regression fits.

Page 9, line 17-25: being "not typical of sinking particles" is certainly not a valid reason to exclude these values from the dataset. Authors are required to provide valid reasons here (e.g. why these C/N ratios would make these particles not wanted in this dataset?).

* Page 9, lines 26-32: again this is a very serious issue. If the sampler building material is potentially responsible for contaminating the samples, how can the authors be confident that not all their POC and PN values are biased by chemicals leaked from this 3D printed part?

Page 10, lines 1-4: this also seems to be pure speculation without any evidence of TEP presence in samples.

C7

Page 10, line 8: if I understand well, the objective of this calibration is to ultimately allow an estimation of biogeochemical fluxes from in situ imaging that could be applied to the largest range of particle types and flux amplitude. It seems very contradictory then to remove particles from the dataset because they are inherently different from the rest of the flux to improve the goodness of the fit. This is very troubling as it suggests that the authors don't fully comprehend their ultimate goal here.

Page 10, line 10-13: this is precisely why it seems so hard to reconstruct a biogeochemical flux from images alone. I strongly suggest that the authors use this example to illustrate the difficulty of meeting the challenge addressed here and impartially discuss their results following the approach of reconstructing the flux from its various particle types having contrasted chemical contents (see general remarks above).

Page 11, line 26: please remove "strong" as it does not seem appropriate. Provide n and p-values.

Page 11, lines 26-27: " that apply over a wide range of environments". This statement could be made with confidence only if the deployments were made in different oceanic regions, seasons and water column layers. It appears too early at this stage to claim this.

Page 11, line 31: "... insensitive to particle classes dominating export". This is not true and is directly contradicted by previous findings shown in this manuscript (see observations made by the authors about the anchovy fecal pellet flux). Please amend as required.

Page 12, lines 5-8: this is confusing and again suggests contradictory intentions of the authors. It is still unclear at this very end of the manuscript if the authors intend to establish a library of VA:element fluxes relationships for each environment and ecological settings sampled (the use of one specific slope would then be reusable to infer the biogeochemical fluxes from images taken in the corresponding region, time of the year and depth), or if they intend to find a general relationship usable in many oceanic

C8

regions, environments and ecosystem structures. In both cases, an extensive work remains to be done and it should be clearly stated.

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