

Manuscript: egosphere-2023-2801 - response to reviewers and editor

Dear Jamie,

Thank you for providing an opportunity to respond to the detailed comments by two reviewers. Here, we provide our response to all the interactive comments (already available online), including the action that we took in the revised manuscript. The original reviewer comments are provided in black and italics; our responses are in red.

Best wishes,

[Mariem Saavedra-Pellitero](#) and co-authors

HANDLING EDITOR

Dear Authors,

I think that some of the confusion between what reviewer 1 thought you presented (ie a classical validation of in situ PIC versus satellite PIC), in comparison to your intended aims (a correlation analysis) is in part due to the some of your phrasing (eg within the abstract and conclusions). For example, in the abstract you use phrase 'validation', which i don't think your work provides. You also have the statement 'strong peaks in satellite-derived PIC south of the Polar Front, which do not show up in the coccolithophore data' - i am not sure that you can say this, as by your own comments you are performing a correlation analysis of two different quantities that you expect to co-vary. So you can comment on whether or not they covary, but you can not imply that one is more correct than the other.

To address these points, in your revised manuscript, please be clearer with your phrasing (eg abstract, conclusions and throughout the paper), and please only state conclusions that have evidence to support them. e.g. if you have no evidence for bubbles or ice or opal particles in your samples, then you cannot draw a conclusion that this is the reason for any perceived differences between your samples and the PIC satellite data. Similarly you can not really state that there are 'errors' in the PIC data as you are not validating the PIC data. Therefore in your discussion, its good to identify possibilities for other signals that may appear in the satellite PIC data based on the literatures (and a similar discussion for your own data would be useful), but here you are hypothesising with no way to conclude the overall answer (ie whether or not they were opal particles or not). To help, I would also suggest focussing your abstract on what you did, what you have achieved and the results (so keep the abstract evidence based) and I suggest avoiding making statements about 'validation' when you work is focussed on correlations. Most of this just about tightening up the phrasing in your narrative and I think this will help strengthen your paper further.

I hope that these comments are useful and I look forward to seeing your revised manuscript.

*best wishes
Jamie*

We thank the handling editor for those points raised. In the revised version of the manuscript we included potential errors and we were very careful with our phrasing. The comments from both reviewers were key in this regard. As you can see in the tracked revised version, the manuscript has notably changed and 'evolved'. We are rather pleased with the new version and we considered that it has improved thanks to all the suggestions provided.

REVIEWER #1

Synopsis: This paper evaluates the coccolithophores in 13 water samples from a transect south of New Zealand to Antarctica (Dec. 2004 to Jan. 2005) plus 19 more water samples from a transect at the western end of Drake Passage (Feb.-March 2016). They made SEM images of the water samples, then calculated the PIC concentration of each sample using the morphometric properties of the coccoliths. Then they used level-3 satellite data to estimate PIC concentrations. These satellite data were binned to 4km-resolution and a mix of temporal binning (daily, 8-day average and one-month time-averaged satellite images but mostly one month binning). They then concluded that Subantarctic waters and waters north of the Polar Front, the NASA PIC algorithm worked reasonably well. South of the Polar Front, however, there were few coccolithophores yet the satellite observations still suggested elevated PIC. The authors suspect that other reflective particles than PIC were causing the PIC algorithm to be in error.

General comments

This paper deals with an interesting topic, the coccolithophores of the Southern Ocean and what appear to be false, high-concentrations of coccolithophores as derived by satellite when the coccolithophore concentrations were observed to be low in their 32 water samples. Unfortunately, the paper is lacking in some fundamental key aspects and this reviewer cannot recommend publication in its current form.

We thank Reviewer 1(R#1) for their suggestions, which will help to notably improve this manuscript. We provide our response to each comment below.

The key shortfalls are: (1) the NASA PIC algorithm measures the concentration of suspended PIC. Nowhere in the paper did the authors ever actually measure PIC analytically in order to judge the errors in the satellite approach,

Our study explores whether PIC estimates derived from coccolithophore data correlated with satellite-derived PIC trends in our area of interest. Due to the lack of in situ measurements of PIC, the aim of this study was to identify broad temporal and spatial trends rather than to provide a precise validation of satellite data against analytically measured PIC in the field. Level 3 satellite-derived PIC data was chosen for the simplicity of its access and processing, but thanks to R#1's suggestions we will now include Level 2 satellite-derived PIC data.

(2) the authors "measured" PIC using an approach based on the morphology of coccoliths and the numbers of coccoliths observed on coccolithophore cells under the SEM, calculating PIC volume. Nowhere did they use analytically-derived PIC estimates to examine the error of this SEM-based approach,

We do not attempt to analytically measure PIC in this study and do not claim to have done this. This aspect of data collection was not the focus of the scientific cruises, which mainly focussed on sediment recovery, not on plankton studies. Our aim in this study is to estimate (not measure) PIC using coccolithophore abundances and in particular biometric measurements on the dominant coccolithophore species *Emiliana huxleyi*, and to compare it with PIC obtained from satellite remote sensing, which was used as a proxy for coccolithophore carbonate surface production (e.g., Balch et al. 2005). In a revised version of the manuscript, we will include standard deviation and errors in the text and on figures for both, coccolith PIC estimates and satellite derived PIC values.

(3) the remote sensing approach used here was inadequate for estimating PIC in these Southern Ocean waters,

The purpose of our study was not to measure PIC concentrations in the Southern Ocean waters for the periods of interest but to explore whether PIC estimates derived from coccolithophore data correlated with satellite-derived PIC trends in our area of interest over the period of interest.

(4) their conclusion that the PIC algorithm does not perform well south of the Polar Front leads them to hypothesize that something else is causing the error. They hypothesized in the abstract that the reflectance signal from these waters was “due to the prevalence of small opal particles or unknown highly reflective particles (such as *Phaeocystis* aggregations) or suspended sediment “ and later in the paper, they mention other possible sources of errors such as microbubbles, floating loose ice or high concentrations of other particulate matter such as glacial flour”, which could cause errors. Unfortunately, barely any evidence was provided for these possibilities. I will elaborate on each of these points below.

We agree with R#1 that our conclusions in relation to this one part of the study are somewhat speculative. However, considering the micropalaeontological / nannofloral nature of this manuscript, we can only hypothesise what could happen south of the Polar Front by citing published scientific literature, because we do not have enough evidence for that. In the new version of the manuscript, diatom data will be added to Figures 3 and 4 as an additional line of evidence to support our conclusions.

Analytical PIC measurements

The gold-standard for measuring PIC in the ocean uses analytical measurement of either particulate Calcium (using any of several methods: particulate calcium analyses using flame photometry or optical emission spectrometry, or carbonate carbon measurements using coulometry). Any of these techniques would have been ideal to use to absolutely validate the PIC concentrations with quantifiable accuracy and precision.

This point is of course absolutely correct. However, we do not aim to absolutely validate satellite-derived PIC concentrations in this study - we are comparing broad temporal and spatial trends based on PIC estimated from coccolithophores and satellite-derived PIC.

PIC volume measurements using SEM

The technique that these authors used (Young & Ziveri, 2000), involves SEM imaging of filtered coccolithophores for the multiple morphotypes of *E. huxleyi* (the most abundant coccolithophore species in these waters). Using this laborious technique, they quantified the following morphotypes for coccolithophores: Type A, Type A over-calcified, Type B, Type B/C, Type C, Type O single layered and Type O double-layered) as well as a few other coccolithophore species (Young & Ziveri, 2000). These measurements involve (for hundreds of cells) quantifying the length of the major and minor axes of the coccoliths, number of t-elements, ray width, tube width, and a shape factor. Each of these measurements will have an error, which would compound to the overall error of the coccolith volume, hence the PIC per cell. No such error bars were ever provided for the PIC measurements of coccolith morphometrics from their 32 samples. Moreover, it was critical to provide the accuracy and precision of the PIC morphometric measurements, measured against calibrated, analytical PIC measurements. As for the influence of diatoms in the Southern waters causing error in the satellite measurements of PIC, the only evidence was shown is four SEM images that contained diatoms.

We will add standard deviations and errors (following Rigual-Hernandez et al., 2020) for coccolith measurements to provide an idea of the accuracy. We also consider different *E. huxleyi* morphotypes, which, as shown by Poulton et al. (2013), is important because it is assumed there is a ~50 % lower content of coccolith calcite for the B/C morphotype of *E. huxleyi* compared to the A morphotype (~0.015 or 0.033-0.035 pmol C coccolith [Poulton et al., 2010, 2011]). Including different morphotypes depicts a more accurate picture and it lowers the error in the estimations of coccolith calcite according to Young and Ziveri (2000). Regarding the diatom data, it has been previously published in Malinverno et al. (2016) and Cardenas et al. (2018). We will also include this data in the revised version of this manuscript.

They never presented the abundance or biomass data of diatoms present in the samples, (nor an estimate of the biogenic silica associated with them, for that matter). It would have been nice to know some of the other features of these waters, too, such as chlorophyll concentrations, POC concentrations, TSS (total suspended sediments) to tease out what the mystery particles were causing the discrepancy.

We will add diatom data and MODIS-derived chlorophyll concentration (mg m⁻³) to figures 3 and 4. The other data mentioned does not exist for these two transects unfortunately.

Remote sensing approach

The authors used questionable remote sensing techniques to derive the PIC concentration. It is not clear exactly what they did here and more information is needed to assess this. They did say that their satellite PIC estimates were based on Level 3 data, highly binned satellite data in space and time. For example, in creating Level 3 data, daily, one kilometer resolution measurements would have been aggregated and averaged into 4km pixels.

We thank R#1 for this comment. MODIS L3 PIC is created by NASA Ocean Biology Processing Group (OBPG). We downloaded daily, 8-daily and monthly MODIS L3 PIC for the area and period of interest, and extracted the value of the pixels surrounding the coordinates of the sampled location. This is described in lines 192-200 of the manuscript, however, we will edit those lines with the new data for clarity.

In addition, using data at 4km resolution could likely have included multiple overpasses over a day, especially given the high latitude. Multiple overpasses mean the radiances are associated with multiple azimuthal and nadir-viewing geometries. Why is this a problem? For algorithm development, one should always use Level 2 data. These data are recovered from a single overpass, first calculated from unprocessed instrument data (Level 0), to Level 1A data (reconstructed, unprocessed instrument counts), converted to calibrated water-leaving radiance data (Level 1B) at a given solar zenith and nadir viewing angle and those radiances are finally processed to geophysical variables as Level 2 data (e.g., PIC concentrations using the PIC algorithms at hand).

We thank the reviewer for this insightful comment and for their advice to use MODIS L2 PIC for this study. As suggested, we will now incorporate L2 satellite-derived PIC in figures 3 and 4 for our comparisons with coccolithophore-PIC estimates. We find that this has no major impact on our findings; in fact, the L2 PIC data match even better than when we were previously using L3 data.

Using already-binned, level 3 data can lead to spurious results for relating radiances to PIC due to the already-mentioned bin-averaged data, likely collected from multiple overpasses, and potentially applying an arithmetic average when the variables are actually log normal in distribution (requiring a geometric average) or vice versa. Algorithm validation should always be done with Level 2 data as per Bailey and Werdell (2006). There's too much binning that goes on to get from L2 to L3 to do an accurate comparison. It looks like, due to abundant cloudy skies in the Southern Ocean, these authors had to use monthly composites for the most part, to get the satellite PIC data to match with their ship samples. Bailey and Werdell (2006) recommend overpasses within ± 3 hours of field observations (not even ± 24 hours), in order to properly validate any ocean colour-derived variable. Here, the monthly level-3 composites would have been good to a temporal resolution of ± 15 d for the most part, hardly a "coincident" matchup! This is a long temporal difference to validate against. One can go from a phytoplankton bloom to background phytoplankton concentrations in just a few days, especially when grazers and viruses are devouring them! An error propagation calculation is essential, for all the measurements described in this paper (satellite, PIC volume), in order to assess the true errors.

As mentioned above, we will now use L2 satellite-derived PIC in the revised manuscript as suggested by R#1. We acknowledge the comments about the importance of temporal resolution and the ideal use of narrow time windows for temporal coincidence when comparing satellite-derived PIC and coccolithophore PIC estimates. However, as R#1 noted, we did not use daily satellite-derived PIC data because of the gaps in the record caused by frequent cloud cover in the Southern Ocean. This is why we used 8-day and monthly composites, as they provided more complete spatial and temporal coverage for our analysis. Even when using L2 data, there are still data gaps as shown in the following Heatmaps we have created (Figures R1 and R2). We will elaborate on this limitation in our discussion and assess how these data gaps might affect the observed trends and correlations.

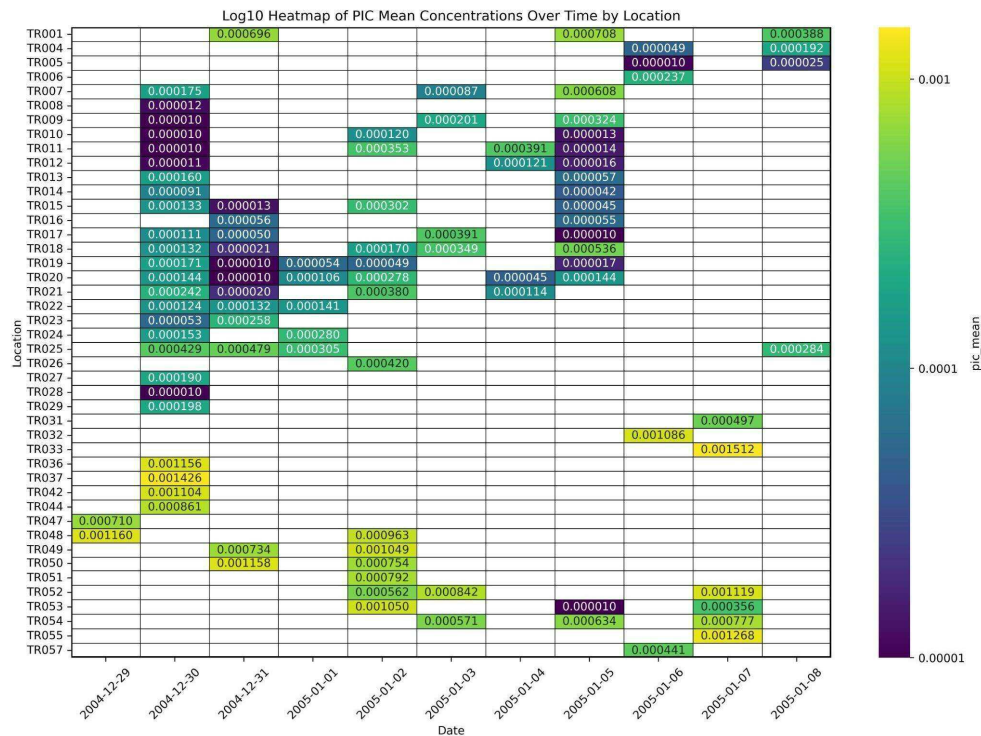


Figure R1: Heatmap showing the L2 PIC (mol m⁻³) for the New Zealand transect. Dates are shown in the X axis and stations (from North -up- to South -down-) in the Y axis. Note that the values on the labels are not logarithmic, but the colour scale is.

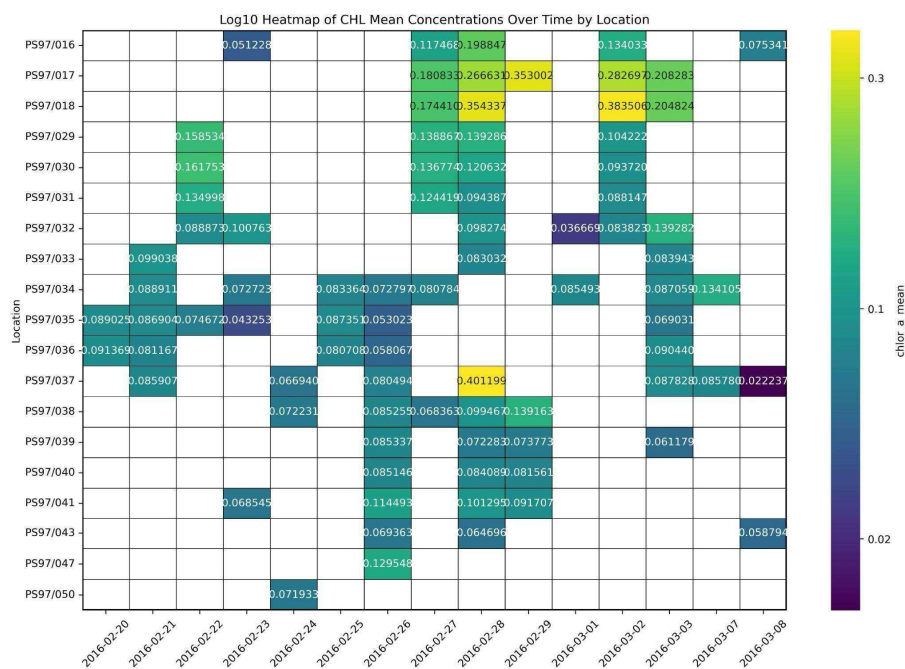


Figure R2: Heatmap showing the L2 PIC (mol m⁻³) for the Drake Passage transect. X axis shows Dates are shown in the X axis and stations (from North -up- to South -down-) in the Y axis. Note that the values on the labels are not logarithmic, but the colour scale is.

Causes for PIC error in the satellite approach south of the Polar Front

On both transects, the authors saw a drop-off in the PIC volume calculations south of the Polar Front. However, the possible reasons are numerous, such as dense diatom populations, dense *Phaeocystis* aggregations, floating ice crystals, bubbles or other suspended sediments, to name a few. Yes, they saw some diatoms in these waters south of the Polar Front (see their Fig. 9c and d) but upon close inspection, there were diatoms in the waters from north of the Polar Front, too (see their Fig. 9a and 9b). They also saw coccolithophores in their SEM from south of the Polar Front (see Fig. 9c). Nonetheless, they ultimately concluded that diatoms must be responsible for this error. Note, the SEM technique would never have visualized soft-bodied *Phaeocystis* in an identifiable form, ice crystals or bubbles, even if they were present.

It is correct that we saw some diatoms south of the Polar Front, not only in the four images included in our original submission but also in previously published studies. The (quantified) diatom data is available in Malinverno et al. (2016) for plankton samples and Cardenas et al. (2018) for surface sediment samples. We will add these data in figures 3 and 4 (but more details can be found in the original sources). We are also aware that *Phaeocystis* aggregations, ice crystals or bubbles would not be visible in our samples. We raise these as a possible explanation based on previously published work (e.g., Balch et al., 2011; Balch, 2018; Balch and Mitchell, 2023), but this is not something we are able (or aiming) to address fully in this paper.

Suspended lithogenic sediments would have been visualized under SEM, however. Finally, foraminifera can be abundant PIC sources in these Southern Ocean waters, too, contributing ~50% of the PIC in waters at 55-60oS (Trull et al., 2018) but they aptly point out that due to the optical properties of large PIC particles (they don't scatter light much per unit PIC), the satellite might not see their reflectance. But this still could be a source of error in their PIC

volume calculation (which included only coccolithophores, not foraminifera or fragments of foraminifera).

We thank R#1 for bringing up this foraminifera point. Certainly, there is PIC produced by foraminifera. However, it is well established that coccolithophores (including detached coccoliths) are responsible for the majority of PIC backscatter (Balch et al., 1996; Balch and Mitchell, 2023). This is mentioned in the introduction.

Balch, W.M., Kilpatrick, K., Holligan, P.M., Harbour, D., Fernandez, E., 1996a. The 1991 coccolithophore bloom in the central north Atlantic. II. Relating optics to coccolith concentration. *Limnol. Oceanogr.* 41, 1684–1696.

Balch, W and Mitchell, C. 2023 Remote sensing algorithms for particulate inorganic carbon (PIC) and the global cycle of PIC
Earth-Science Reviews, 239 (2023), p. 104363

This paper desperately needs an error analysis so that the authors can put error bars on the PIC estimates that appear in the figures and tables.

We agree and will add error analysis to all figures and tables in the revised version.

Specific comments:

P1 L23: Spelling “Coccolith”

We have corrected the spelling mistake

P1 L26: Which satellite product was used? What sensor?

This information is stated in line 206 of the manuscript, in section 3.4. In the new version L2 data will be used. The information is included also in the Methods section.

P2 L47: They should also cite the earlier references who were really the first ones to show this (Holligan et al., 1993; Robertson et al., 1994).

Those references will be added.

P3 L89: change word “concentrations” to “measurements”. Next sentence, change “Most” to “many” since there are a significant number of subpolar studies that enumerated free coccoliths.

Those two changes will be made

P3 L94: See also (Young et al., 2014)

This reference will be added

P4 L99: The Oliver et al paper is now published (Oliver et al., 2023) and it included non-bloom areas, too, I believe.

The citation will be updated.

P4 L104: Does aim #1 include both plated coccolithophores as well as detached coccoliths? Coccolith morphometrics were always performed on coccospheres. The number of detached coccoliths/L (plus coccospheres/L) was only considered in the estimates for the New Zealand transect for the PIC estimates (see Malinverno et al., 2015). This will be mentioned in revised sections 3.2 and 5.1). Saavedra-Pellitero et al. (2019) only considered cells/L (detached coccoliths/L numbers for the Drake Passage were not available). That is why this aim is more general and we decided to keep it as it is.

P4 L117 and occurrences henceforth in the manuscript: In the field of physical oceanography, salinity data should not be reported with units of ‰.

The units (‰) will be deleted

P5 L135: Cite also (Holligan et al., 2010)

We believe that this refers to the original L132-133, so we will add the reference there).

P5 L143: *R/V Italica* should be italicized.

We will italicise *R/V Italica* and *Polarstern*, also in the figure captions.

P5 L144: Is XX referring to the roman numeral or is this some sort of place-holder that needs to be filled in?

XX refers to the roman numeral, so this will remain unchanged.

P5 L147-154: The authors are critical of how few stations are available in NASA's remote sensing validation of the PIC algorithm (n=42; noting that these are clear-sky matchups, using L2 satellite data within +/- 3 hours of an overpass (see general comments above). It should be noted, too, there is very strong under-sampling in this study, with a total of 32 water samples from which the paper is based, and these samples are based on Level 3, binned data which should not be used in satellite validation.

We have not claimed to have performed any satellite validation in this study, we just provided the validation statistics from NASA's Ocean Biology Processing Group (OBPG) PIC validation products. We will also now use L2 satellite data, as suggested previously.

P6 L168: There are some details the authors might elaborate on. A SEM only views one side of a coccosphere. What about the coccoliths one can't see? Is this taken into consideration with the shape factors? It looks like they approximate the coccolith volume in equation 1 as a cube (L3), why not a sphere?

To estimate the number of *E. huxleyi* coccoliths per coccosphere we counted the visible ones (half coccosphere) and multiplied by two (see Table S4). This information will be added to the revised manuscript. For other species, generic numbers were taken from Young and Ziveri (2000). All this information is available in Table 1.

Equation 1 was just taken from Young and Ziveri (2000) and it has been broadly used in the literature (even in papers published in Biogeosciences, such as Rigual Hernández et al., 2020). A full discussion of this is beyond the scope of this study.

P6 L186: For those not familiar with the Young and Ziveri method, could you say what "tube" you are talking about?

L186 refers to the *relative tube width*' (an index calculated using equation 2) not the *tube width* (the latter being the tube between central area and rim or "T-elements", see figure 2 or R3 for clarity). Considering that Young et al. (2014) have shown that the degree of calcification of a coccolith appear to broadly co-vary with the *relative tube width*, we will change the *relative tube width*' for the original index (*relative tube width*; Figure R3) - which makes more sense - in the new version. We therefore modified Figures 3, 4 and 6 accordingly. We will also refer to Figure 2 at the end of that sentence, which clearly shows the measurements involved in equation 2 in different SEM pictures.

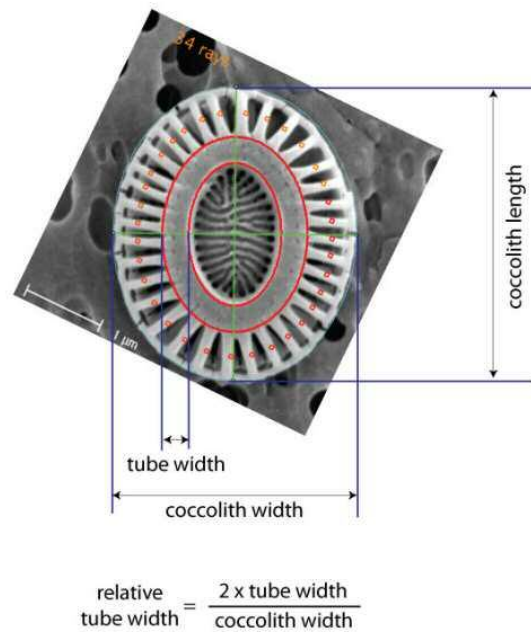


Figure R3. Morphometric parameters measured by Young et al (2014) and the equation used to calculate the *relative tube width*.

P7 L192: What fraction of your samples used daily, 8-daily or monthly PIC values? Was there a difference on how well the validation agreed when you used the shorter time scale validations?

As discussed in previous comments, we did not validate the satellite-derived PIC with in situ PIC concentration.

You should state this. I would suspect that since you show monthly images in your Figure 1, then most of your comparisons were done with monthly images. Is this correct? Your comment at line 204-205 would suggest that the number of daily match-ups was extremely low. Your table 2 lists these out but for the actual validation, if you had a daily, an 8-d and a monthly measurement, which did you use? Please explicitly state this! Using 8-d composites or monthly composites means that your validations were hardly +/- 3h (the standard for true satellite validation (Bailey & Werdell, 2006)...see general comments above)! Also, which MODIS instrument did you use? Both were aloft during these cruises.

We use monthly PIC rasters in Figure 1 maps as background. As stated in line 220, we used both weekly (8-day) and monthly satellite-derived PIC data in the study. We did not perform validation of the satellite-derived data, only compare the coccolith PIC estimates with the weekly and monthly values. The comparisons were graphed in figures 3 and 4, and will be updated with L2 data in the new version.

P7 L207-239- It is not clear what exactly the PCA analysis was for (as related to the validation of the PIC volume method). This should be stated in the lead sentence for that paragraph.

We will delete the PCA so this comment does not apply anymore.

P8 L230-“Almost all” doesn’t equal “solely”

We will delete “solely” since it was misleading.

P8 L233- What is the accuracy of the standard PIC algorithm? How does this compare to the accuracy of the morphometric approach? What is the absolute calibration? standard for PIC?

NASA Ocean Biology Processing Group (OBPG) validates PIC retrievals performed with available matchups returns mean bias of 0.30577 and mean absolute error of 4.00304 (both values calculated after applying the log10 transformation to the PIC values) (<https://oceancolor.gsfc.nasa.gov/data/reprocessing/r2022/aqua/>). This information will be included.

P8 L234- Samples probably too small (volume-wise) to adequately sample forams and certainly not pteropods, other sources of Southern Ocean PIC . See Trull et al. (Trull et al., 2018) for data on this. These can add significant PIC to the water.

R#1 is right. Unfortunately our samples are too small to check other possible sources of PIC. We added this information in the discussion.

P9 L261 I hope that in the Discussion, you compare this carbon per coccolith with other estimates (for which there are many).

We do compare our data to other estimates in the discussion.

P10 L301- And temporal scales, too!

The temporal scale will be added.

P10 L307- Yes, but those are 1:1 clear-sky ship versus satellite comparisons using analytically derived PIC concentrations. Here, the authors are using a technique with unquantified errors (coccolith-morphometrics) which are not calibrated to analytically-derived PIC. See general comments above.

We will add errors and standard deviations to our datasets.

P10 L310- Of course there are other particles! You are only seeing the diatoms and coccolithophores and maybe armored dinos in the SEM since they would preserve well on the filter. But there are huge abundances of purely organic cels like Phaeocystis, picoplankton and nanoplankton (without any sort of mineral coverings to make them visible in the SEM).

We will delete “limited”, make it clear that we are talking about our study area only, and add a reference here to emphasise that there are indeed other biogenic particles.

P11 L335-336- The attribution of light intensity and iron limitation are meaningless here since no such data were taken in this study (to know if it is even remotely feasible).

Since this is simply a case of speculating in the discussion, we will modify this statement.

P11 L351-356- You desperately need an error analysis.

We will add errors and standard deviations to our datasets

You are presenting data from some 32 water samples (all surface, or some at depth?)

This has been indicated in the methods and figure captions already, but we will add it here also.

The level-3 satellite data (which originate over the top two optical depths in a weighted fashion) are averaging over large pixel sizes. Did you pick just one pixel or the center pixel and surrounding 8 pixels? Most of the matchups were done with monthly averages (which may indeed have only resulted from a single overpass given the cloudiness in this region. A lot can happen to a population in one month! The potential errors on both sides are enormous. And you have no standard (analytical PIC measurements) on which to judge any of these. See general comments.

We use the value of the pixel that corresponds to the observation location. As already mentioned in previous comments, because this study does not aim to measure in situ PIC concentration values and because there are big gaps in the satellite-derived PIC record, we

did not perform a precise temporal matchup. Instead, our approach is to compare both sources of PIC to see if their spatial and temporal trends were similar. However, will also now use L2 satellite data, as previously mentioned.

P15 L454-455- I'm sorry, but this is not a very definitive comparison, with no absolute standard analytical measurement.

We will tone this statement down.

P15 L458- Define "good". Nowhere do you ever plot one versus the other, nor present statistics.

A plot with the correlation coefficient will be added to the supplementary material.

P15 L460- How do you know that the morphometric algorithm is also not biased, especially when there is no comparison to gold-standard analytical PIC measurements.

This is correct, we are not able to judge if the algorithm is not biased. We will make this more obvious in the conclusions.

P15 L467- But no where did you present the actual abundance of diatoms. What about the organic particles that you can't see with the SEM?

We will add diatom abundance to Figures 3 and 4 and mention the potential influence of other biogenic particles in the text.

P15 L472- See Trull (Trull et al., 2018) for elsewhere in the Southern Ocean. They show varying influence of other larger calcifiers!

We are grateful for the reference provided. We were just referring to the potential contribution of coccoliths to the PIC in the previous version of the manuscript, but it is true that larger calcifiers and fragments of larger carbonate-forming organisms could be important. This will be mentioned in the updated version.

P15 L485- Regarding future research, you never defined the accuracy of the morphometric method, nor discussed the accuracy of the satellite method.

The morphometric method of Young and Ziveri (2000) was assumed to be the most advantageous for our data set, since both the taxonomic analysis and the morphometric measurements of the coccoliths had been performed using a SEM. This is undoubtedly the most accurate approach for identifying coccoliths to a morphotype level, as well as for measuring the coccoliths length, allowing us to produce a consistent coccolith biogeochemical dataset without other errors linked to the preparation of new samples and to the coccolith-calcite calibration, both required for the optical/birefringence analysis, which would represent the alternative for the calculation of the coccolithophorid PIC.

NASA Ocean Biology Processing Group (OBPG) validates L2 PIC retrievals performed with available matchups returns mean bias of 0.30577 and mean absolute error of 4.00304 (both values calculated after applying the log10 transformation to the PIC values) (<https://oceancolor.gsfc.nasa.gov/data/reprocessing/r2022/aqua/>).

We will edit the text so that we are able to better define accuracy as stated above in the revised version.

Fig. 1 legend- What do big dots and little dots mean. Are the big dots the actual samples that you estimated coccolith morphometrics?

This information will be added to the caption: "Large dots indicate samples in which biometries on *Emiliania huxleyi* were performed, and small dots where coccolithophore census were available".

Fig. 3 legend- Are all these measurements from the surface? 3m depth? I don't recall seeing

this.

The information was provided in Sections 3.1.1 and 3.1.2 but will also be added to the figure 3 and 4 captions.

Please add error bars to the data when possible.

I'm not sure these colors will necessarily be visible to a color-blind person. Please use correct color pallet. I'm not sure "electric blue" is a good color descriptor to a color-blind person. What are the units of "tube width"? The triangle and diamond symbols are so small that one can't discern them.

We will add one standard deviation to our PIC estimates as in Rigual-Hernandez et al (2020).

Symbols will be enlarged, colours will be modified and figures checked here:

<https://www.color-blindness.com/coblis-color-blindness-simulator/>

The caption will be modified accordingly

Relative tube width' has no units. See Young et al. (2014) for further information.

Fig. 4 legend- Same comments as for Fig. 3

Symbols were enlarged, colours were modified and figures were checked here:

<https://www.color-blindness.com/coblis-color-blindness-simulator/>

The caption was modified accordingly

Fig. 5 - I'm confused for panel (C) since it shows 38 samples just for panel C (New Zealand transect) and 16 samples for the Drake Passage transect. These don't match your original numbers?

This was not very clear previously, but the initial counts for the New Zealand Transect were performed in light microscope (58 samples), from those, a subset of 38 was analysed in SEM to distinguish the different morphotypes, and from those we picked 13 samples for the biometrics. This will be explained in the updated figure caption.

In the case of the Drake Passage, 17 were included, but two of the sampling points are really close, so it looks like there are 16. In 2 of the samples there were no coccolithophores, that is why there are fewer samples. This will be mentioned now in the figure caption.

We will also reduce the width of the bars, so all the sample locations are more visible

In doing this we discovered that we made a mistake calculating coccolith PIC (due to *Calcidiscus leptoporus* average length being 5.7 instead of 5), so all the figures will be updated (3, 4 and 5), as well as Table 1.

Fig. 6- panel b- confusion again over the numbers of samples in the New Zealand transect (here showing 13 samples) and Drake Passage transect (here showing 15 samples).

Symbols in these figures are tiny! Make them bigger.

Some of the samples offshore Chile are really close to each other, and that is why it looks like there are fewer samples.

Fig. 7- I do not understand how this PCA figure advances the goal of the paper. Eliminate? We will delete this.

Fig. 8- Not sure a color-blind person will be able to interpret the colors in this. Can you add error bars to this given a thorough error analysis? What is "C-*Calcita*" referring to? PIC?

This figure has been modified in order to address these points raised.

The legend will better specify what is what in the new version.

Fig. 9- I see diatoms in all four SEMs. Did you quantify them in any way? I see plated coccolithophores and coccoliths in panel c (from south of the Polar Front). What does this mean for the hypothesis?

This information will be added to figure 3 in the revised version.

Detached coccoliths/L and coccospheres/L were both considered when estimating PIC for that transect. We will make this clear in the revised manuscript.

Summary

In summary, there are so many unquantified errors in these measurements (for both coccolith morphometrics and satellite PIC) that this reviewer cannot recommend publication of this paper in its current form. If it is to be published, it will require a major revision.

We will add error assessments to our datasets as suggested by R#1. We hope this addresses these overall concerns.

References

Bailey, S. W., & Werdell, P. J. (2006), A multi-sensor approach for the on-orbit validation of ocean color satellite data products, *Rem. Sens. Environ.*, 102, 12-23.

This reference will be added.

Holligan, P. M., Charalampopoulou, A., & Hutson, R. (2010), Seasonal distributions of the coccolithophore, *Emiliana huxleyi*, and of particulate inorganic carbon in surface waters of the Scotia Sea, *Journal of Marine Systems*, 82(4), 195-205.

This reference was already included in the previous version

Holligan, P. M., et al. (1993), A biogeochemical study of the coccolithophore, *Emiliana huxleyi*, in the north Atlantic, *Global Biogeochem. Cycles*, 7(4), 879-900.

This reference will be added

Oliver, H., McGillicuddy, J., D. .J. , Krumhardt, K. M., Long, M. C., Bates, N. R., Bowler, B. C., Drapeau, D. T., & Balch, W. M. (2023), Environmental drivers of coccolithophore growth in the Pacific sector of the Southern Ocean *Global Biogeochem. Cycles*, 37(11), <https://doi.org/10.1029/2023GB007751>.

This reference was already included in the previous version, but the paper was not yet accepted at the time of submission. The correct reference and citations will be updated.

Robertson, J. E., Robinson, C., Turner, D. R., Holligan, P., Watson, A. J., Boyd, P., Fernandez, E., & Finch, M. (1994), The impact of a coccolithophore bloom on oceanic carbon uptake in the northeast Atlantic during summer 1991, *Deep-Sea Research Part I*, 41(2), 297-314, doi:10.1016/0967-0637(94)90005-1.

This reference will be added

Trull, T. W., Passmore, A., Davies, D. M., Smit, T., Berry, K., & Tilbrook, B. (2018), Distribution of planktonic biogenic carbonate organisms in the Southern Ocean south of Australia: A baseline for ocean acidification impact assessment, *Biogeosciences*, 15(1), 31-49, doi:10.5194/bg-15-31-2018.

This reference was already included in the previous version

Young, J. R., Poulton, A. J., & Tyrrell, T. (2014), Morphology of *Emiliana huxleyi* coccoliths on the northwestern European shelf - Is there an influence of carbonate chemistry?, *Biogeosciences*, 11(17), 4771-4782, doi:10.5194/bg-11-4771-2014.

This reference was already included in the previous version

Young, J. R., & Ziveri, P. (2000), Calculation of coccolith volume and its use in calibration of carbonate flux estimates, *Deep-Sea Research Part II: Topical Studies in Oceanography*, 47(9-11), 1679-1700, doi:10.1016/S0967-0645(00)00003-5.

This reference was already included in the previous version

REVIEWER #2

Summary evaluation and general comments

I think the study is valuable and, after some major revision, should be published. One overall issue is with the way they focus the study in the Title, Abstract and Intro goes into the weakest result where they do not succeed in resolving things so well. They clearly consider the greatest importance to be the comparison of the in-situ and satellite data, but I think they end up raising more questions in this sense than they succeed in resolving. A major deficiency is the lack of direct discrete measures of PIC by chemical analysis with to which to compare microscopy-based and satellite estimates of PIC. Without such data, they really can't complete a proper comparison. I recognize that it is not always trivial to get or process such samples, and I see some other points of value.

The strongest findings are that

- a) coccolithophores are not contributing significantly to the apparent high PIC estimated by satellite in the Antarctic Zone, but are important further north;*
- b) they strengthen what we know of the biogeographic patterns in the high latitude southern hemisphere, picking two very important but quite distinct part of the circumpolar circulation;*
- c) there are uncertainties and disagreements between methods for estimating coccolith-PIC using SEM and or polarized microscopy and they provide information that allows evaluating that disagreement.*

The first of those three points, that the AZ satellite signal assigned to PIC is not from coccolithophores, is both one of the most important results, but it is also the only part where they do a strong comparison between satellite and microscope-based approaches. In the rest, they do not complete the comparative analysis with a statistical comparison of the different approaches.

Consequently, my recommendation is that the authors should be encouraged to submit a major revision, and I suggest to them to change their focus to the points where they generate robust results that are nevertheless valuable.

We thank Reviewer 2 (R#2) for their helpful suggestions that will lead to a much improved manuscript. We provide our response to each point below.

Major specific comments:

Lines 101-106 and later (generally). This study seems to combine previously published data with new analysis, specifically with new analysis that relies on samples which were analyzed more deeply. It would help to cite that or those previous studies from these transects in this paragraph. I think early in the Methods there should be a first table defining where the samples come from, including noting in which previous studies they have been used in.

A new table will be added that includes this information. Also, we will highlight the already published datasets in figures 3 and 4.

Lines 191-206: 3.3 Satellite-derived PIC data

I think it is interesting to compare in situ data to the output distributed from the NASA group, but then I am not sure what exactly they did. Did they compare a single pixel to each discrete sampling?

Bailey and Wedell (2006) outline the method used by NASA Ocean Biology Processing Group (OBPG) for validating operational ocean colour missions. According to the authors, they would have compared the mean of a 5x5 window, centred on the in situ location. The in situ data utilised was extracted from SeaBASS and the Aerosol Robotic Network - Ocean Color (AERONET-OC). Validation results for MODIS-Aqua PIC are available on NASA's Ocean Color Portal

(https://seabass.gsfc.nasa.gov/search/?search_type=Perform%20Validation%20Search&val_sata=1&val_products=9).

Beyond showing later that there is a region of very high satellite signal with low or absent coccolithophores, I suspect this sampling is not really sufficient to do a true comparison of the satellite vs discrete measures.

R#2 is right to point out that the sampling is not sufficient to do a true comparison, particularly because the discrete measurements do not provide analytically-measured concentrations of PIC. Additionally, the satellite-derived PIC data is scarce due to cloud cover. See also reply to Reviewer 1 (R#1) as well as figures R1 and R2 in that response.

Section 4.1.

Lines 233-237: "Weekly and monthly MODIS-derived PIC at the sampling locations consistently overestimate PIC values... with respect to in-situ values calculated from coccolith mass" and "there is a relatively good agreement in the latitudinal satellite and coccolith-PIC trends in the SAZ and PFZ" and again at lines 292-293: "In the studied transects, the calculated coccolith-PIC and the satellite-derived PIC trends show quite good agreement in the SAZ and PFZ,"

This is a possibly highly subjective, evaluation of correspondence has been made. What is "good agreement"? Sometimes when coccolith-estimated PIC is low, satellite-estimated PIC is also low, and when coccolith-estimated PIC is higher, satellite-estimated PIC is also higher, but, even in the SAZ and PFZ they do not track precisely. I do not think they could track precisely even if all PIC was from coccoliths and the estimations were completely faithful, because one is a very discreet estimate at a single sample in a single Niskin bottle, @and the satellite value is an average over at least many days. So they can't correlate perfectly. Nevertheless, to make the statement it would be nice to see a direct analysis of the correlation between coccolith-PIC (calculated) and satellite-estimated PIC. That is, I'd like to see figure, combining both datasets, plotting one against the other.

This point was also raised by R#1. This analysis will now be included in the supplementary material.

Also, the phrasing could be more careful in lines 233-244, because "consistently overestimate PIC values" is only correct if one knows that most PIC comes from coccolithophores instead of other sources, which might not be true. Also, it is not clear if there is a way of knowing the error on the coccolithophore PIC estimation.

We will reword these sentences and add information about potential errors relating to other sources of PIC as well as standard deviation in the text, figures and tables. This was also raised by R#1.

There are several reasons why the estimates could not agree. Here are just some principal possibilities:

Perhaps the satellite estimates in fact are right, but a lot of coccolith PIC is in the form of coccolith fragments that are too fragmented to be recognized and counted, so microscopy estimates would be lower than the “true” value

Perhaps there is another PIC producer (although that possibility was implicitly considered in the Intro)

Perhaps the assumptions in the coccolith-PIC calculations are actually wrong (e.g., the shape factor is different than estimated, or the number of coccoliths per cell assumed is wearing)

In the Discussion, the authors do touch on some of these and other possibilities, but I would like to suggest they might try a modest reorganization so as to make it clear to go one by one through the possibilities to consider.

We are very grateful for those suggestions. We will reorganise the discussion to incorporate these points.

Line 270: “The relative tube width’ (an index for calcification; Young et al., 2014), calculated using equation 2, varies from...”

We will amend (now relative tube width).

How do you deal with the issue that the “overcalcification” is variable? The tube width in the “overcalcified” forms tends to be very irregular, and sometimes the central area is covered nearly fully. This should be made explicit, perhaps with a new panel in Fig. 2 or a supplementary figure associated with the Methods section to show how this complicated situation (like the case in Fig. 6 bottom left) is dealt with.

We agree that the calcification of the central area is variable, just as all measured parameters of the placoliths are highly variable. “Overcalcified” specimens were in general rather rare, but as we have assumed an average value for all other parameters, we have calculated a mean ks value that is higher for this morphotype than for the other morphotypes (see Table 1).

This data is available in the supplementary material submitted to Biogeosciences (Tables S1 and S2). It is also stored in Pangaea: <https://doi.pangaea.de/10.1594/PANGAEA.964672> and <https://doi.pangaea.de/10.1594/PANGAEA.964674> -but still under moratorium-.

We did not add any extra panel or supplementary figure, because the other morphotypes are more dominant (see cells/L in figures 3 and 4).

I have some concerns about the PCA and how it is discussed.

First, the PCA does not seem to be very successful at separating the coccoliths based on morphometric parameters. Except for the overcalcified forms, which separate only based on tube width (needs to be clearer how that is really measured, as discussed above), the other forms overlap a lot. In fact, component 2 in the PCA, which correlates mostly with other morphometric characters quantified, does not appear to separate the morphotypes at all.

Some of the characters that have been used for separating types A and B are not analyzed, and might be quite difficult to analyze automatically (such as central area type, or whether the distal shield elements or straight or not). I think it is definitely valid to do and report the PCA, but it is important to discuss the fact that this analysis, using the current state-of-the-art quantification, seems to fail at what are visually quite striking differences is very notable. This suggests that perhaps it is time to try new approaches to distinguishing these types.

Also following a comment from R#1, we will remove the PCA section from the revised version. We will incorporate the other very interesting points raised here in the discussion of the revised manuscript.

Second, the issue also makes me wonder if we have independent estimates of how well the approaches of trying to estimate coccolith PIC by morphometry or by polarized microscopy work when comparing different morphotypes. It seems that, while there are many studies that directly quantify PIC in culture (either as acid-labile particulate carbon or as particulate calcium), it is hard to find any that also count the number of coccoliths to measure PIC/coccolith. It might be worth highlighting this need. I should mention, and not parenthetically, that I appreciated a lot the analysis they showed in Fig. 8, which goes directly to this point. I think their data suggests we are still lacking precision in the way we measure coccolith PIC, and I would hope that, while their study might not resolve it, at least it helps identify the problem.

Good points, and we will elaborate further on this topic in the new version of the manuscript.

Third, in lines 346-347 they say: “The PCA performed on the E. huxleyi morphometric dataset shows that those heavily calcified type A coccospheres occupy a relatively restricted ecological niche offshore of Chile” The PCA is only based on morphometric characters, regardless of the location or oceanographic conditions in which they are found. How can such a PCA, which does not include any environmental information, indicate anything about whether the ecological niche of one form is restricted or not?

Good point. We know that just because previous published papers dealt with the different morphotypes and the environmental preferences (add citations here), but they were not included here. That is why we decided to (initially) focus on the PIC and leave the PCA out of the main story. Ultimately, and in response to a R#1 comment too, we have decided to remove the PCA aspect.

For Figure 1, and the corresponding description in the Methods (lines 108-122), I could not tell if the white lines correspond to average positions of the fronts or to the positions the fronts occupied at the time of the study.

We will include a modified figure that uses a colour blind friendly palette and make clear that they are average positions (based on Orsi and Harris, 2019).

Minor technical and comments and corrections

Abstract: Eliminate page breaks within abstract

It will be done

Line 84: Perhaps reserve “concentration” for chemicals (such as PIC) and “abundance” for cell or coccolith numbers/volume. It’s not strictly necessary, but it might help to be clearer

It will be done

Line 88-89 : “ available coccolithophore concentrations” maybe clearer “available measurements of coccolithophore abundances”

It will be done

Line 98: “have targeted areas of coccolithophore bloom”. Should be “blooms” (plural)

It will be done

172-173: “and modified for *E. huxleyi* according to the degree of calcification obtained for each morphotype (see Table 1)” How is this modification of KS according to calcification performed? Is there a reference?

The different shape factors used were based on the identified morphotype following Young and Ziveri (2000): $ks = 0.02$ for morphotypes A and B/C and $ks = 0.04$ for morphotype A overcalcified. The shape factor for morphotype O ($ks = 0.015$) was introduced by Poulton et al. (2011) in a plankton study along the Patagonian Shelf for a morphotype with a central area described as an “open or thin plate” which the authors called type B/C but that we identified as morphotype O based on the published images and description of Hagino et al. (2011).

The dataset linked to this answer is available in the supplementary material submitted to Biogeosciences (Tables S1 and S2). It is also stored in Pangaea:

<https://doi.pangaea.de/10.1594/PANGAEA.964672> and

<https://doi.pangaea.de/10.1594/PANGAEA.964674> -but still under moratorium-.

The reference that compiles that information is Vollmar et al. (2022) and it has been included in the new version.

Vollmar, N. M., Baumann, K.-H., Saavedra-Pellitero, M., and Hernández-Almeida, I.: Distribution of coccoliths in surface sediments across the Drake Passage and calcification of *Emiliania huxleyi* morphotypes, *Biogeosciences*, 19, 585–612, <https://doi.org/10.5194/bg-19-585-2022>, 2022.

See also our response to R#1 for further details.

Line 178: “Measurements of the distal shield diameters of *Calcidiscus leptoporus* the second most abundant species” Need a comma after “*leptoporus*”

It will be done

Line 265-266: “Note that this data is shown in Figures 3 and 4, but the coccolith-PIC was calculated in this work using equation 1 and the average lengths mentioned in Table 1 “ Not clear.

All the data is shown in Figure 3c and 4 c, but to estimate the coccolith PIC, the average length was considered. The sentence will be modified to make this clearer.

Line 270: We observed that some coccoliths are clearly overcalcified (see Figure 5 for an example),...” Do you mean Fig. 6? Fig. 5 doesn’t have any photos so can’t see that observation.

Yes, that is a mistake. We meant Figure 6. Thanks for spotting it.