

Supplement. Answer to reviewer #1

We thank anonymous referee #1 for the interest he showed in our manuscript and detailed comments that have helped to improve the original version of the manuscript. We have considered all his/her comments and addressed each of his/her concerns below.

R1-Cx : Referee comment, **R1-Rx**: authors response.

R1-C1: *The manuscript documents an annual record of coccolithophore production and coccolith weights/lengths at a Southern Ocean site. The topic is worthy of publication in biogeosciences, and the results will be of potential interest to the wider scientific community.*

In general, the manuscript is well written and illustrated, and does not contain any major flaws. However, see below for minor points.

Scientific points Line 38: “ : : : coccolith assemblages experienced weight and length reduction ..” !? It is not the assemblages that have reduced weight and length, it is the coccoliths

R1-R1: Corrected according to reviewer 1’ suggestion. Now it reads: “coccoliths captured by the traps experienced weight and length...”

R1-C2: *Line 89: the Southern Ocean is a small area ? – 25% of the global area looks quite significant to me*

R1-R2: The sentence referred by reviewer #1 has been rephrased in order to avoid subjective descriptions of the size of the Southern Ocean. Now it reads: “Despite the fact that the Southern Ocean accounts for about 25% of the global ocean, it contains ~40% of the global ocean inventory of anthropogenic CO₂”

R1-C3: *Line 148: why is this section in the methods ? move regional setting and oceanography to the introduction*

R1-R3: Corrected according to reviewer 1’ suggestion. In the new version of the manuscript, section “Regional setting and oceanography“ has been moved to the introduction (subsection 1.2). Section 1.1. has been titled “1.1. Background and objectives”. Subsections of Material and Methods section have been renumbered accordingly.

R1-C4: *Line 235: why unfiltered ? I am not a specialist on sediment traps but it seems odd to use unfiltered seawater. Is there not a risk of contamination ?*

R1-R4: The water used to fill the sediment trap cups was unfiltered deep seawater from > 1000m, where the particle abundance is so low that filtering is unnecessary and hard to do without adding more particles than you remove. Moreover, it is important to highlight that the risk of contamination is negligible since the particle levels in sea water are of the order of micrograms per litre while concentration in the trap cups after recovery are of the order of milligrams per litre. This point has been clarified in the new version of the manuscript. The following text has been added in lines 446-449 of the new version of the manuscript with tracked changes:

“Risk of sample contamination by the unfiltered seawater is considered negligible due to the fact that the deep water exhibits low particle abundance and also because particle concentration in sea water is of the order of µg/L while concentration in the trap cups after recovery was of the order of mg/L.”

R1-C5: *Lines 357 and 345: “coccolith particle bloom” – since coccoliths are inanimate (just pieces of calcium carbonate) I think the word ‘bloom’ is inappropriate here – use ‘The summer coccolith flux exhibited : : :’*

R1-R5: Corrected according to reviewer 1’s suggestion.

RI-C6: Line 548: you mention two factors that possibly explain the changes in calcification. Calcification (i.e. overgrowth) tends to increase with depth in the photic zone, at least in some areas of the world. So in winter it may be that the coccolithophores are sitting deeper and therefore have more calcified coccoliths than in the summer when they are closer to the surface and therefore with lightly calcified coccoliths. Of course this difference in surface vs deeper photic could be related to various parameters (light, nutrients, temperature). Do you have data/images of coccoliths from different photic depths? In Plate 1 you show lightly and heavily calcified coccoliths from the traps – but how do they relate to the surface oceans?

RI-R6: We appreciate reviewer 1's comment. Unfortunately, there is no data available of coccolith weight from different photic depths. Only samples collected by two sediment traps (that were deployed far below the photic zone) and satellite data are available for the study site. Therefore, our current data set precludes the assessment of the relationship between coccolith weight and the depths where the coccolithophore populations developed. In regard to seasonality, no relationship between the overgrowths and a particular period of the annual cycle was observed. This is now clarified in a sentence that has been included in the new version of the manuscript with tracked changes ("lines 674-675").

Minor points for correction/consideration

RI-C7: Title and elsewhere: Just a query. Is the use of Australian Sector OK? Naming the sectors after the oceans, like the Atlantic Sector, Pacific Sector and Indian Ocean Sector is fine, but I wonder whether using country names (for sectors and territories) is considered to be geopolitical.

RI-R7:

We acknowledge the point highlighted by reviewer 1. Indian sector could also be an appropriate term for referring to the study region of this research. Nonetheless, we decided to use the term "Australian sector of the Southern Ocean" in order to be consistent with the terminology of previous work along the 140°E parallel such as Findlay and Giraudeau (2000), Quéguiner (2001), Trull et al. (2001), Sedwick et al. (2008), de Salas et al. (2011), Lannuzel et al. (2011) and many others. Please find the references of the publications mentioned in the previous sentence listed below:

de Salas, M. F., Eriksen, R., Davidson, A. T., and Wright, S. W.: Protistan communities in the **Australian sector** of the Sub-Antarctic Zone during SAZ-Sense, Deep Sea Research Part II: Topical Studies in Oceanography, 58, 2135-2149, <http://dx.doi.org/10.1016/j.dsr2.2011.05.032>, 2011.

Findlay, C. S., and Giraudeau, J.: Extant calcareous nannoplankton in the **Australian Sector** of the Southern Ocean (austral summers 1994 and 1995), Marine Micropaleontology, 40, 417-439, [http://dx.doi.org/10.1016/S0377-8398\(00\)00046-3](http://dx.doi.org/10.1016/S0377-8398(00)00046-3), 2000.

Lannuzel, D., Bowie, A. R., Remenyi, T., Lam, P., Townsend, A., Ibsanmi, E., Butler, E., Wagener, T., and Schoemann, V.: Distributions of dissolved and particulate iron in the sub-Antarctic and Polar Frontal Southern Ocean (**Australian sector**), Deep Sea Research Part II: Topical Studies in Oceanography, 58, 2094-2112, <http://dx.doi.org/10.1016/j.dsr2.2011.05.027>, 2011.

Quéguiner, B.: Biogenic silica production in the **Australian sector** of the Subantarctic Zone of the Southern Ocean in late summer 1998, Journal of Geophysical Research: Oceans, 106, 31627-31636, 10.1029/2000JC000249, 2001.

Sedwick, P. N., Bowie, A. R., and Trull, T. W.: Dissolved iron in the **Australian sector** of the Southern Ocean (CLIVAR SR3 section): Meridional and seasonal trends, Deep Sea Research Part I: Oceanographic Research Papers, 55, 911-925, <http://dx.doi.org/10.1016/j.dsr.2008.03.011>, 2008.

Trull, T. W., Bray, S. G., Manganini, S. J., Honjo, S., and François, R.: Moored sediment trap measurements of carbon export in the Subantarctic and **Polar Frontal zones of the Southern Ocean, south of Australia**, Journal of Geophysical Research: Oceans, 106, 31489-31509, 10.1029/2000JC000308, 2001.

R1-C8: Line 30: Don't mix z and s verbs. For example, here you use 'characterized' and on line 151 'summarized', but on line 35 you use 'analysed' and on line 135 'fertilisation'. Furthermore, on line 236 you use 'programmed' and on line 349 'grey'. You need to be consistent, and choose between British English and US English. It looks like you are favouring the former.

R1-R8: Corrected according to reviewer 1's suggestion. The whole manuscript has been revised and corrected in order to be consistent with the use of British English (i.e. verbs "z" has been replaced by "s" when needed".

R1-C9: Line 45: coccolithophorid vs coccolithophore. Be consistent, and choose one.

R1-R9: Corrected according to reviewer 1's suggestion. In order to be consistent the word coccolithophorid has been replaced by coccolithophore in the new version of the manuscript.

R1-C10: Line 62: "...some species (but not all) of coccolithophore .." – please change to 'some species of (but not all) coccolithophores ..'

R1-R10: Corrected according to reviewer 1's suggestion.

R1-C11: Lines 71-75: needs to be rewritten, as it doesn't make sense

R1-R11: The sentence highlighted by reviewer 1 have been deleted following the suggestion of reviewer 2 (See **R2-C21**).

R1-C12: Line 79: one bracket is missing

R1-R12: Corrected according to reviewer 1's suggestion.

R1-C13: Line 102: dominantly present -> dominate

R1-R13: Coccolithophores are abundant in the subantarctic waters of the Southern Ocean, but this does not mean that they dominate the phytoplankton communities in terms of numbers or biomass. In order to be more precise, the sentence highlighted by reviewer 1 has been rewritten: "coccolithophores exhibit high concentrations in the Subantarctic Southern Ocean"

R1-C14: Line 115: spares -> sparse

R1-R14: Corrected according to reviewer 1's suggestion.

R1-C15: Line 149: ".at the north : : : at the south .." -> '..in the north : : : in the south ..'

R1-R15: Corrected according to reviewer 1's suggestion.

R1-C16: Line 203: CO2 rich -> CO2-rich

R1-R16: Corrected according to reviewer 1's suggestion.

R1-C17: Line 253: "After settling 12 hours .." -> 'After settling for 12 hours ..'

R1-R17: Corrected according to reviewer 1's suggestion.

R1-C18: Line 271: by using-> using

R1-R18: Corrected according to reviewer 1's suggestion.

R1-C19: Line 273: "...to the winter .." -> '..to winter ..'

R1-R19: Corrected according to reviewer 1's suggestion.

R1-C20: Line 285: Scanning Electron Microscopy -> scanning electron microscope (SEM)

R1-R20: Since this point has been also mentioned by reviewer 2 (See **R2-C55**), the text has been modified trying to satisfy both reviewers suggestions. The text now reads: “a Scanning Electron Microscope (SEM)”.

R1-C21: Line 298: “..using a with a Nikon ..” -> ‘ : : :using a Nikon .. ’

R1-R21: Corrected according to reviewer 1’s suggestion.

R1-C22: Line 346: “ ..should be looked with caution .. ” -> ‘should be viewed with caution ’

R1-R22: Corrected according to reviewer 1’s suggestion.

R1-C23: Line 358 (and elsewhere) : you need to insert *x* (times) between the number and the power. For example, 2.2 10 -> 2.2. *x* 10

R1-R23: Corrected according to reviewer 1’s suggestion. The manuscript has been revised and “x” has been included between the number and the power when absent.

R1-C24: Line 365: *Biogenic* -> *biogenic*

R1-R24: Corrected according to reviewer 1’s suggestion.

R1-C25: Line 370: *of the species Calcidiscus* -> *of Calcidiscus*

R1-R25: Corrected according to reviewer 1’s suggestion.

R1-C26: Figure 5 (and elsewhere): *I realise that ‘liths’ is in common use in presentations, but it is not an official term. Better to use coccoliths.*

R1-R26: Corrected according to reviewer 1’s suggestion. The word “liths” has been replaced by coccoliths in the Y-axis of Figure 5. The text has been revised although no inconsistencies were found.

R1-C27: Line 382: *tiles? I think you mean ‘elements’*

R1-R27: PREGUNTAR A Lluïsa Cross The text has been corrected following reviewer 1’s recommendation. The term tiles has been replaced by “tile-like elements”.

R1-C28: Line 384: “*Distal shield measures ranged between 2 and 4,35 ..*” -> ‘*Distal shield ranges from 2.0-4.35 ..*’ [use decimal point not comma]

R1-R28: Corrected according to reviewer 1’s suggestion.

R1-C29: Line 424: *here you use station 62 S, and before 62 S site – perhaps be consistent in usage*

R1-R29: The words site and station are used as synonyms in the text and are used alternatively in order to avoid repetition. Therefore, no changes in the usage of these words have been incorporated in the text.

R1-C30: Line 463: *genetical* -> *genetic*

R1-R30: Corrected according to reviewer 1’s suggestion.

R1-C31: Line 550: *degrees of calcification* -> *degree of calcification*

R1-R31: Corrected according to reviewer 1’s suggestion.

R1-C32: Line 555: *B/C south 50_S* -> *B/C south of 50_S*

R1-R32: Corrected according to reviewer 1’s suggestion.

R1-C33: Line 576: *light-dependant* -> *light-dependent*

R1-R33: Corrected according to reviewer 1’s suggestion.

R1-C34: Line 617: *absence accompanying in situ* -> *absence of in situ*

R1-R34: Corrected according to reviewer 1’s suggestion.

R1-C35: Line 643: *That supported* -> *That is supported*

R1-R35: Corrected according to reviewer 1's suggestion.

R1-C36: Lines 741-759: *delete, as the same as later references*

R1-R36: Corrected according to reviewer 1's suggestion.

R1-C37: Lines 773 and 816 (and elsewhere): *Deep Sea* -> *Deep-Sea*

R1-R37: Although we agree with reviewer 1 that the title of the journal is "Deep-Sea Research" all the references downloaded from their official website display the title of their own journal like "Deep Sea Research". Therefore, in order to be consistent with the references of the journal we have kept the title in all references without the "-".

R1-C38: Line 860: *emiliana huxleyi*

(*haptophyta*) ¹, *Journal of phycology*, -> *Emiliana huxleyi* 860 (*haptophyta*) ¹,
Journal of Phycology, [why is there a superscript 1 at the end of the title ?

R1-R38: Corrected according to reviewer 1's suggestion. The superscript 1 has been deleted from the title.

R1-C39: Lines 864, 867 and 884 (and elsewhere): *italicize the species name*

R1-R39: Corrected according to reviewer 1's suggestion. The references have been revised and species names have italicized.

Additional changes

- Section 3.3, line 674, of the "new version of the manuscript with tracked changes", the sentence "annual amplitude of the coccolith weight was approximately" has been replaced by "annual amplitude of the **mean** coccolith weight was approximately" in order to be clearer.
- The correlation between Biogenic silica and coccolith fluxes at 2000 m showed in line XX of the first version of the manuscript was the correlation coefficient ($r = 0.86$), not the coefficient of determination ($R^2 = 0.74$). In the new version of the manuscript the coefficient of determination is shown.
- The coccolith length values presented the results listed in section 3.3 of the first version of the manuscript (lines 405-409) corresponded to an earlier version of the data set.. The correct values have been included in the new version of the paper. Please note that the seasonal trend remains identical (only there was is a slight variation of the absolute values). Please also note that the calculations made in the discussion regarding the relationship between size and weight in the first paragraph of Section 4.3 are correct. The coccolith length data at 3700 plotted in figure 6 also corresponded to the older version of the dataset. This has been corrected in the new version of the manuscript. Please note that the seasonal trend remains identical.
- Plate I: In the first version of the manuscript the we skipped the letter "d" listing the photos of Plate I. In the new version of the manuscript, this typo has been fixed.

Supplement. Answer to reviewer #2

R2-Cx : Referee comment, **R2-Rx**: authors response.

R2-C1: General comments.

The calcification of coccolithophores in the high latitude regions is a growing concern as it will have large influence on ocean biogeochemistry and thus climate. The data provides new information on coccolithophore response to varying environmental conditions at Antarctic Zone using sediment traps samples. However, the information provided here can be expressed in a much better way. Sediment trap data from Southern Ocean is difficult to obtain and is therefore a welcome addition to knowledge and needs to be published. Here are some major and minor comments which need to be incorporated in the manuscript.

R2-R1: We sincerely thank reviewer #2 for the careful reading of our manuscript and constructive criticisms and comments that helped to improve the manuscript. The text and figures have been revised and improved accordingly. Next, we briefly summarize the main changes included in the text. A potential explanation behind the formation of secondary crystallizations observed in some of the coccoliths has been included together with the references suggested by the reviewer. Also, in the new version of the manuscript the possible influence of salinity on coccolith morphology is discussed and new references dealing with the impact of temperature in the coccoliths have been included. Moreover, several figures have been improved following reviewer 2's suggestion: Figure 1 now shows the maximum and minimum sea ice extent during the deployment period; the vertical structure of temperature of the water column in Figure 2 is now plotted with Ocean Data View; several aspects of Figure 3 have been improved following reviewer 2's comments; and a new figure has been created (Fig. 7) that shows the regression plots between coccolith weight and length. Finally, a throughout revision of the references cited in the manuscript has been performed.

R2-C2: Major comments/changes needed to be done:

1. Authors documented only abundance of coccoliths of E. huxleyi B/C morphotypes in the traps. It is also mentioned that occurrence of other coccolithophores were also documented. Though other coccoliths are in low abundance, it represents changes in the upper oceanic conditions. It is also important to plot a graph of other coccolithophores and discuss what their assemblage indicates.

R2-R2: Due to the very low abundance of other coccolithophore species in the trap samples, authors decided to focus the discussion on total coccolith fluxes because flux plots based on very small counts could be biased and therefore misleading for the reader. However, in compliance with reviewer's request the fluxes and relative contribution of *Emiliana huxleyi*, *Calcidiscus leptoporus* and *Gephyrocapsa* spp. at both sediment trap depths have been included in the new Supplementary Figure 1. Moreover, the seasonality of these species is now described in the section 3.1 of the results, as well as, discussed at the end of the first paragraph of section 4.2.

R2-C3: 2. The overcalcification of E. huxleyi is documented by few researchers in past few years. But not many papers are published on this. Authors have documented overcalcification on coccoliths retrieved from sediment traps. I assume intact coccospheres are also documented in the both the sediment traps. In this case, whether authors documented overcalcification on coccospheres of E. huxleyi? If any information

is available on living coccolithophores in this region, it should be included. It is important to document the overcalcification on E. huxleyi is a natural process and not a part of secondary calcification. So, if extant coccolithophores data is available at/around study site then it should be presented.

R2-R3: We appreciate reviewer 2's comment on the possible overcalcification of some of the coccoliths captured by the trap. As stated in section "3.2 SEM analyses", the unusual structures (mainly small spherules deposited on the coccoliths) observed on some of the coccoliths, such as that of the coccoliths shown in Plate I, e-g, is attributed to a secondary recrystallization but not overcalcification. This interpretation is based on the small spherules often present in the coccoliths, particularly on the laths, a feature consistent with a secondary recrystallization and not with overcalcification of the coccoliths during the life cycle of the coccolithophore.

Cubillos et al. (2008) undertook a comprehensive analysis of the *E. huxleyi* morphotypes in the Australian sector of the Southern Ocean along the 140°E meridian (covering the location of the sediment trap station analysed here). According to the former authors the overcalcified forms of *Emiliana huxleyi* are restricted north of the Polar Front (Subantarctic and Subtropical Zones), and therefore it is unlikely that we register these forms in our traps. Furthermore, the only overcalcified forms reported by Cubillos et al. (2008) correspond to morphotype A, which is characterized by a larger coccoliths than those observed here. Please note that Cubillos et al. (2008) paper is discussed in section 4.3 of the manuscript. Moreover, an explanation about the possible origin of the "small spherules" observed on some coccoliths has been included at the end of section 4.4. of the discussion in order to satisfy reviewer 2's request.

In regard to the documentation of coccospheres in the trap samples. The number of coccospheres found in the samples was very low due to the low abundance of coccolithophores in the study region. In order to overcome this problem, one could state that concentration used in the SEM preparations could have been increased. However, this was not possible in our particular case due to the large abundance of diatoms at this site which concentration obviously increase with concentration. Please note that the biogenic silica fluxes at the 61°S site are the arguably the highest ever reported in the world's ocean. Such high concentration of diatoms greatly hampered the finding of coccospheres under the SEM, that it is the only methodology that allows discriminating between overcalcified and "normal" coccospheres.

R2-C4: 3. Authors often compared diatom assemblage with coccoliths. Plotting a graph of total diatom assemblage vs coccoliths abundance will be useful for understanding changes in the AZ region.

R2-R4: Authors do not completely understand reviewer 2's request. Total diatom valve and total coccolith fluxes at both sediment trap depths are plotted in figure 5. Only one diatom species is mentioned in the text: *Thalassiotrix antarctica*. The seasonal succession of diatom species at the 61°S site is discussed in detail in Rigual-Hernández et al. (2015, JMS) paper that is mentioned in the text. Authors believe that plotting the fluxes and relative abundance of the all the diatom species would be out of the scope of this paper, would not contribute to the discussion and would be misleading for the reader. However, these graphs can be included in the manuscript or in a supplementary figure if the editor considers this information relevant.

R2-C5: 4. A recent study indicates polysaccharides are also responsible for overcalcification of coccolithophres. Authors need to discuss outcomes in more detail and should be cited with recent references.

R2-R5: Corrected according to reviewer 2's suggestion. A paragraph dedicated to the possible role of the *polysaccharides serving as organic scaffold* for coccolith formation has been included and new references included and discussed (Gal et al., 2016; Lee et al., 2016) (end of section 4.4)

R2-C6: 5. Authors should be consistent in framing sentences. Some sentences are too large, some are too small. Be consistent in using AZ vs AZ-S, Fe vs. iron, E. huxleyi, vs Emiliana huxleyi etc.

R2-R6: Corrected according to reviewer 2's suggestions. The whole manuscript has been revised and several sentences have been split into two when possible:

- Line 85 of the new version of the manuscript with tracked changes: a long sentence has been removed.
- Line 86-87: the sentence has been split into two.
- Line 145-146: the sentence has been split into two.
- Line 1096-1098: The introductory sentence of the conclusions section has been split into two and rephrased.

Moreover, the text has been revised for inconsistencies in the use of the terms:

- AZ vs AZ-S, Fe vs. iron, *E. huxleyi* vs *Emiliana huxleyi*.
- Fe vs. iron: The term "Fe" has been replaced by "iron" in the new version of the manuscript.
- The term "AZ" has been replaced by "AZ-S" when possible.
- *Emiliana huxleyi* has been replaced by *E. huxleyi* when possible.

R2-C7: 6. Since, both sediment traps are located in the 61degS and there is no comparison done with other sediment traps showed in the Fig. 1, it is not necessary to mention "at 61S sediment trap everywhere"

R2-R7: Corrected according reviewer 2's suggestion. The name of the sampling site has been replaced by synonyms when possible. However, the term 61°S site is still used in the new version of the manuscript when needed.

R2-C8: 7. Authors should crosscheck references very carefully. Many references listed in the text are not reflected in the reference list. Similarly, many references listed in the Reference list are not

R2-R8: Corrected according to reviewer 2's suggestion. The references listed in the text and in the reference list have been revised. Only a few errors were found and have been corrected in the new version of the manuscript.

R2-C9: mentioned in the text. Genus and species name should appear properly and in italics.

For ex. Line 860- emiliana huxleyi.

R2-R9: Corrected according to reviewer 2's suggestion. The text and references have been thoroughly revised in order to show all species and genus names in italics.

R2-C10:Hagino et al. 2011 reference- written in caps

R2-R10: Corrected according to reviewer 2' suggestion.

R2-C11:Minor comments:

Line 14-30: The information provided in the abstract can be shifted to the introduction. The abstract should start from Line 31.

R2-R11: We agree with reviewer 2 that the first lines of the abstract could also fit in the introduction. However, we believe that providing a very short rationale of the experiment and highlighting the gaps in the knowledge about the effects of a changing climate in Southern Ocean ecosystems is important to help the non-specialized reader to understand the relevance of our study, thereby potentially reaching a larger audience. Therefore, authors have decided to leave the abstract as it is in the first version of the manuscript.

R2-C12:Line 32: In the deep ocean >>> at the Antarctic Zone

R2-R12: Corrected according to reviewer 2's suggestion. Now it reads: "We report here on seasonal variations in the abundance and composition of coccolithophore assemblages collected by two moored sediment traps deployed at the Antarctic Zone south of Australia (2000 and 3700 m depth) for one year in 2001-02."

R2-C13:Line 33: ~2000>>> 2000

R2-R13: Corrected according to reviewer 2's suggestion.

R2-C14:Line 33: and 3700>>> and 3700 respectively

R2-R14: In the new version of the manuscript this sentence has been rephrased. As consequence of this change is no need to include the word "respectively".

R2-C15:Line 37: Emiliana huxleyi morphotype B/C>>>> E. huxleyi morphotype B/C

R2-R15: Corrected according to reviewer 2's suggestion.

R2-C16: Line 38: coccolith assemblage experienced weight and length reduction>>> coccoliths experienced weight and length reduction

R2-R16: This sentence had already been corrected following reviewer 1's suggestion.

R2-C17: Line 39: during the summer months>>> during summer months (December-March?)

R2-R17: The text has been modified slightly different to that suggested by reviewer 2. Now it reads: "reduction during summer (December – February)"

R2-C18: Line 40: at both sediment trap depths>>> at both sediment traps

R2-R18: Corrected according to reviewer 2's suggestion.

R2-C19: Line 41: in other southern ocean settings>>>> which settings?

R2-R19: Corrected according to reviewer 2's suggestion. Patagonian shelf and Scotia sea are mentioned between brackets in the new version of the manuscript.

R2-C20: Line 43-46: Apart from first record, significant outcomes of the study needs to be highlighted here.

R2-R20: The major findings of the investigation are summarized (and numbered) before this sentence. Moreover, in the last sentence of the abstract we clearly explained the importance of our results clearly explaining that our results provide a reference/baseline for evaluation of Southern Ocean coccolithophore responses to changing environmental conditions in the coming decades. Therefore, we believe that the main objectives and outcomes of the study are already mentioned in the text and no more extra information is required.

R2-C21: Line 71-75: May not required in the introduction

R2-R21: Corrected according to reviewer 2's suggestion. The text: "For example, diatoms can play a prominent role in export of organic matter from the surface ocean, because of their heavy siliceous frustules and capacity for aggregation and rapid sinking facilitates efficient transport of organic carbon (Buesseler, 1998; Smetacek, 1999). Nonetheless, it has also been suggested that this silica-mediated carbon export driven by diatoms may not always reach the ocean interior efficiently (Francois et al., 2002; Lam and Bishop, 2007)." has been removed from the introduction.

R2-C22: Line 76: Coccolithophores also has the potential >>>> coccolithophores has potential

R2-R22: Corrected according to reviewer 2's suggestion.

R2-C23: Line 89: 25% of ocean area is not small.

R2-R23: This sentence has been modified following the suggestion of both reviewers. Now it reads: "Despite the fact that the Southern Ocean accounts for about 25% of the global ocean, it contains ~40% of the global ocean inventory of anthropogenic CO₂"

R2-C24: Line 107: Trull et al., 2017 is not mentioned in reference list

R2-R24: We believe there must have been a misunderstanding here since Trull et al., 2017 (Biogeosciences) was mentioned in the first version of the manuscript (lines 1097-1099)

R2-C25: Line 109: Cubilos et al. 2008 or 2007? Cubilos et al. 2008 is not mentioned in the reference list

R2-R25: Corrected according reviewer 2's suggestion. Cubillos et al. 2008 does not exist. The text has been corrected, now only the citation Cubillos et al., (2007) appears in the text.

R2-C26: Line 111: Freeman and Lovenduski (2015) not mentioned in the reference list

R2-R26: Corrected according reviewer 2's suggestion. The reference Freeman and Lovenduski (2015) appears now in the text.

R2-C27: Line 116-122 and elsewhere in the introduction: very large sentence. Authors should be consistent in framing sentences. Such long sentences to be avoided in the introduction

R2-R27: Corrected according reviewer 2's suggestion. Please see **R2-R6** for more details.

R2-C28: Line 123: Trull et al., 2017 missing in the reference list

R2-R28: As mentioned in a previous comment (**R2-R24**), we believe that there must have been a misunderstanding here since Trull et al., 2017 did appear in the reference list (lines 1097-1099 of the first version of the manuscript)

R2-C29: Line 138: inferred from one-year record>>> inferred from 10 month record

R2-R29: The sentence has been rephrased and the words “one-year record” replaced by “during ten months”.

R2-C30: Line 139: SOIREE..... Elaborate when using short forms for the first time

R2-R30: Corrected according to reviewer 2’s suggestions.

R2-C31: Line 147: Regional setting and oceanography; and water carbonate chemistry, should be shifted in the introduction under a different sub-heading.

R2-R31: Corrected according to both reviewer 2 and reviewer1’s suggestion. Sections “regional setting and oceanography” is section 1.2 in the new version of the manuscript, while “Water column chemistry in the study region” is now section 1.3

R2-C32: Line 151... SAACF>>>SACCF

R2-R32: Corrected according to reviewer 2’s suggestion.

R2-C33: Line151: (SAACF)>>>(SACCF) Fig.1

R2-R33: Corrected according to reviewer 2’s suggestion. Figure 1 in now cited between brackets at the end of the first sentence of section “1.2. Regional setting and oceanography”.

R2-C34: Line 155: upper water column with nutrients (add reference). Similarly for sentences between lines 155-160 (add references)

R2-34: Before the description of the seasonal evolution of water column physical, chemical and biological properties, it is mentioned: “Trull et al. (2001b) summarized the seasonal evolution of water column properties in the study region”. Although we could refer to this cite in each following sentence, authors believe that the text should remain as is now in order to avoid repetition.

R2-C35: Line 155: Chlorophyll-a, vs/ Line 484 chl-a>>> use one style of writing

R2-R35: Corrected according reviewer 2’s suggestion. The term chl-a has been replaced by chlorophyll-a and the text has been revised for inconsistencies.

R2-C36: Line 160: in algal biomass occurs within the mixed layer (add reference)

R2-R36: Corrected according reviewer 2’s suggestion. The reference Trull et al. (2001b) has been included.

R2-C37: Line 164: Trull et al. 2001>>>> Trull et al. 2001a or Trull et al. 2001b or 2001c??

R2-R37: Trull et al. 2001b is the correct reference here. In the new version of the manuscript the correct reference is specified.

R2-C38: Line 169: Large calanoid copepodites.>>> Copepods and copepodites are different. Copepodites are immature form of copepods. What authors are trying to say? “large Calanoid copepods” or “mainly Calanoid copepodites”??

R2-R38: As highlighted by reviewer 2, copepodites are immature forms of copepods. Zeldis et al. (2001) reported that “The SOIREE site mixed-layer mesozooplankton community was dominated by copepods, with salps and pteropods absent, and euphausiids either absent or very rare (maximum 7 animals m⁻³). The copepod community was numerically dominated by large copepodites (> 1.5mm prosome)...”. Therefore, we believe that the sentence “Mesozooplankton analysis during the SOIREE experiment by Zeldis (2001) indicates that zooplankton community in the study region is dominated by copepods, mainly large calanoid copepodites.” is correct. That is the reason why the sentence has not been modified.

R2-C39: Fig. 1. Since author has mentioned that sediment trap location was away from sea ice activity, authors should draw seasonal sea ice zone or winter sea ice limit for the year 2001-2002 in Fig. 1

R2-R39: Corrected according to reviewer 2’s suggestion. The winter sea ice limit for August 2001 is now showed in Figure 1. The Figure caption has been adapted accordingly citing the source of the sea ice data represented in Fig. 1. Moreover, the database where this data was obtained is also cited in the new version of the manuscript.

R2-C40: Line 180: SACCF- Southern ACC Front>>>> SACCF- Southern Antarctic Circumpolar Current Front

R2-R40: Corrected according to reviewer 2’s suggestion.

R2-C41: Line 193: calcite saturation horizon (CSH)>>>> Calcite Saturation Horizon (CSH)

R2-R41: Corrected according to reviewer 2’s suggestion.

R2-C42: Line 194: CaCO₃ compensation depth (CCD)>>>> CaCO₃ Compensation Depth (CCD)

R2-R42: Corrected according to reviewer 2’s suggestion.

R2-C43: Fig. 2. Similar to Fig. 1 and Fig. 3, Fig. 2 should be plotted in the Ocean data view and figures should be readable. What does the dotted line indicate in Fig. 2b?

R2-R43: Corrected according to reviewer 2’s suggestion. The vertical structure of temperature of the water column has been plotted in Ocean Data view (Fig 2a of the new version of the manuscript). Due to the low number of observations of nutrient concentrations in the study regions, their representation using ODV would require a large interpolation of measurements and the resulting graph would be somewhat misleading for the reader. Therefore, authors have decided to leave the Figure representing silicate and nitrate concentration as it was in the first version of the manuscript (Fig. 2b in the new version of the manuscript).

R2-C44: Fig 2 a. legend should contain surface macronutrient concentrations?

R2-R44: Only data of the nutrient concentration in the mixed layer is available. This data is data plotted in Figure 2b. In order to be clearer, in the new version of the manuscript it is clarified that the data showed in Figure 2b is representative for the mixed layer.

R2-C45: Line 200: (Fig. 3)>>> (Fig.3a) or (Fig. 3b)

R2-R45: Corrected according to reviewer 2’s suggestions. Figure 3b is mentioned in the new version of the manuscript.

R2-C46: Fig. 3. Mark Fig. 3a and Fig 3b; Mark frontal locations, put units for color scale bar, x axis etc. Also, include sampling dots if possible. Mark 1000m sediment trap in fig 3 in different color., as it is mentioned in line 226.

R2-R46: Figure 3 has been corrected according to reviewer 2's suggestions.

R2-C47: Line 210: Elaborate when appear for the first time

R2-R47: Corrected according to reviewer 2's suggestions.

R2-C48: Line 211: Tanhua et al. 2008 is missing in the reference list

R2-R48: The reference Tanhua et al. (2008) has been replaced by CARINA group (2011) which appears now in the reference list. The new reference refers to the same data set of that used by Tanhua et al. (2008), i.e. both references are correct.

R2-C49: Line 122: Draw seasonal sea ice zone in Fig. 1

R2-R49: Corrected according to reviewer 2's suggestion. In the new version of the manuscript the Maximum Winter Sea Ice Extent and Minimum Summer Ice Extent for the study period (August 2001 and February 2002) are represented in Figure 1. The Figure caption has been rewritten accordingly and the dataset from where the sea ice data has been extracted is now cited in the text (Fetterer et al., 2017).

R2-C50: Line 225: for approximately one year>>>> for ten months

R2-R50: Strictly speaking the sampled period is 10 months and a half (317 days / 30 day per month = 10.56 months). Authors believe that it is correct to leave the text as it is now because it is specified the number of days sampled between brackets. "The 61°S mooring was equipped with three McLane Parflux time series sediment traps (Honjo and Doherty, 1988) for approximately one year (November 30, 2001 to September 29, 2002, 317 days)."

R2-C51: Line 227: highlight Fig. 3a and 3b

R2-R52: Corrected according to reviewer 2's suggestion. In the new version of the manuscript both Fig. 3a and 3b are mentioned in the sentence referred by reviewer 2.

R2-C52: Line 231: be consistent in using depths. Either use "~2000" or "2000",,,, "3800 or 3700

R2-R52: Corrected according reviewer 2's suggestion. In the new version of the introduction only "2000 m" is used (i.e. not "~2000" used once in the introduction in the first version of the manuscript. Moreover 3800 has been replaced by 3700 following the comments of the reviewer.

R2-C53: Line 283: Emiliana huxleyi>>>> E. huxleyi

R2-R53: Corrected according to reviewer 2's suggestions.

R2-C54: Line 235: why unfiltered seawater used? Won't it contaminate samples?

R2-R51: A similar question was raised by reviewer 1 (R1-C4). In the new version of the manuscript it has been clarified the reasons why unfiltered seawater was used. In the new version of the manuscript it is stated: "Risk of sample contamination by the unfiltered seawater is considered negligible due to the fact that the deep water is characterized by low particle abundance and also because particle concentration in sea water is of the order of µg/L while concentration in the trap cups after recovery was of the order of mg/L."

R2-C55: Line 285: *Scanning Electron Microscopy*>>>> *Scanning Electron Microscope (SEM)*

R2-R55: Corrected according to reviewer 2's suggestion.

R2-C56: Line 286: *decantation method outlined by*>>>> *method outlined by*

R2-R56: Corrected according to reviewer 2's suggestion.

R2-C57: Line 287: *coated in Gold*>>> *coated with/using Gold*

R2-R57: Corrected according to reviewer 2's suggestion.

R2-C58: Line 288: *please mentioned magnification range (for example 2000-7000x) used during analysis.*

R2-R58: Corrected according to reviewer 2's suggestion. The magnification used during the SEM analysis is specified in the new version of the manuscript (magnification 5000-20000x).

R2-C59: Line 288: *Emiliana huxleyi*>>>>*E. huxleyi*

R2-R59: Corrected according to reviewer 2's suggestion.

R2-C60: Line 291: *Emiliana huxleyi*>>>>*E. huxleyi*

R2-R60: Corrected according to reviewer 2's suggestion.

R2-C61: Line 293: *Emiliana huxleyi*>>>>*E. huxleyi*

R2-R61: Corrected according to reviewer 2's suggestion.

R2-C62: 293-294: *Emiliana huxleyi coccoliths into morphotypes*>>> *Emiliana huxleyi into different morphotypes*

R2-R62: Corrected according to reviewer 2's suggestion.

R2-C63: Line 298: *using a with a Nikon*>>>> *using a Nikon*

R2-R63: Corrected according to reviewer 1 and 2's suggestions.

R2-C64: Line 319: *sea surface temperature (SST) already elaborated in line 153*

R2-R64: Corrected according to reviewer 1 and 2's suggestions.

R2-C65: Line 320: *Sea Surface Temperature Analysis*>>>> *SST Analysis*

R2-R65: Corrected according to reviewer 1 and 2's suggestions.

R2-C66: Line 323: *SST variations*>>>> *Sea Surface variations*

R2-R66: This change has not been incorporated because the authors wanted to refer specifically to Sea Surface Temperatures (SST) not Sea Surface variations in general.

R2-C67: Line 326: *Photosynthetically active radiation (PAR)*>>>> *Photosynthetically Active Radiation (PAR)*

R2-R67: Corrected according to reviewer 2's suggestion.

R2-C68: Line 327: *particulate inorganic carbon (PIC)*>>>> *Particulate Inorganic Carbon (PIC)*

R2-R68: Corrected according to reviewer 2's suggestion. Moreover, in the caption of figure 4, the names Photosynthetically Active Radiation and Particulate Inorganic Carbon have been replaced by their acronyms, i.e. PAR and PIC.

R2-C69: Fig. 4. Authors have mentioned March as a late summer months in the line 158. In this case, the shaded area should also include March

R2-R69: The sentence highlighted by reviewer 2 has been corrected. In the new version of the manuscript, it reads: "By late summer-early autumn (March) SST ranges between 2 and 3 °C"

R2-C70: Line 341: particulate inorganic carbon (PIC)>>>> PIC

R2-R70: Corrected according to reviewer 2's suggestion.

R2-C71: Line 342-346- It is important..... Trull et al., 2017)>>> already mentioned before

R2-R71: Reviewer 2 is right, the fact that the satellite algorithm used to detect PIC is not reliable in Antarctic waters is mentioned in the introduction as well. Nonetheless, authors believe it is important to mention this point again in the caption of Figure 4 in order reinforce this idea and to make the reader aware that the PIC satellite data presented in that figure should be viewed with caution.

R2-C72: Line 347: particulate organic carbon (POC)>>>> POC; Calcium Carbonate (CaCO3)>>>> CaCO3

R2-R72: Corrected according to reviewer 2's suggestion.

R2-C73: Fig. 5. Please check scale bars. 2x10⁹ appeared twice on left side.; in the first figure only 10⁹ appeared. Is it 1x10⁹?. Put same scale in both figures. Mark Figure 5a and 5b.

R2-R73: Corrected according to reviewer 2's suggestion. A decimal has been included in the all the labels of axis of Figure 5 in order to avoid the number repetition highlighted by reviewer 2. Moreover, now axis in both figures have the same scale. Finally, the axis titles have been corrected as they were wrongly named in the first version of the manuscript.

R2-C74: Fig. 5. What is the reason diatom valve flux remained almost constant at 2000 and 3700 but there is an increase in diatom flux during February and March. Please explain.

R2-R74: Differences in the magnitude of fluxes between the upper and deeper trap are most likely due to small differences in the source area of the particles collected by each trap, the so-called statistical funnel (discussed in section 4.1). This statistical funnel increases with depth and therefore it is expected some variability between the fluxes captured by each trap. This is clarified in the new version of the manuscript where it is stated: "The slightly different seasonal pattern observed at both sampling depths (Fig. 5) is mainly attributed to the fact that the area of the ocean from which the particles have been produced increases with depth (Siegel and Deuser, 1997) (863-865 of the new version of the manuscript with tracked changes).

R2-C75: Line 370-372: which Calcidiscus leptoporus species? Small or intermediate? Which time of sampling month these species are documented?

R2-R75: *Calcidiscus leptoporus* coccoliths were not divided into size classes during the LM microscopy analysis. Therefore, in the new version of the manuscript it is specified “*sensu lato*” after the species name. Relative abundances of this species can be found in Table 1. Moreover, in the new version of the manuscript the fluxes and relative abundance of this species have been plotted in supplementary Figure 1, are described in the results section “3.1 Seasonal dynamics of coccolith export fluxes” and discussed in discussion section “4.2 Seasonal dynamics of the calcareous and siliceous phytoplankton fluxes”. Please also note that due to the low abundance of this species in the samples and to its similar seasonal pattern to that of *E. huxleyi*, *C. leptoporus* data does not provide any relevant contribution to the discussion. That is the main reason why initially this data was not included in the first version of the manuscript and also why the information of this species is included as supplement.

R2-C76: Line 383: distal shields partially missing, may be due to >>>> distal shield partially missing, due to

R2-R76: Corrected according to reviewer 2’s suggestion. The sentence has been rephrased to: “...partially missing, mainly due to the slender and delicate structure of the laths”.

R2-C77: Line 384: 2 to 4,35>>>> 2 to 4.35

R2-R77: Corrected according to reviewer 1 and 2’ suggestion.

R2-C78: Line 394: (2.3 and 2.1 pg/coccolith)>>>> (2.3 ±.... and 2.1±.... pg/coccolith)

R2-R78: Corrected according to reviewer 2’s suggestion. The standard deviation is now showed in all the coccolith mass values provided in section “3.3 Coccolith weight and length changes”

R2-C79: Line 396: (down to 1.6 and 1.9 pg at 2000 and 3700m, respectively)>>>> (down to 1.6±.... and 1.9±.... pg/coccolith at 2000 and 3700m)

R2-R79: Corrected according to reviewer 2’s suggestion. The standard deviation is now showed in all the coccolith mass values provided in section “3.3 Coccolith weight and length changes”

R2-C80: Line 399: Average annual coccolith weight at the 61S traps>>>>it is already mentioned that both traps are located at AZ-S, at 61S. just mentioned depths. Similarly at Line 433, at the 61S site>> study site. Similarly correct changes at line 446 and elsewhere

R2-R80: Corrected according to reviewer 2’s suggestion. The name of the station 61°S site has been replaced when possible in the text. Please note that some time specifying the station is needed, such as in line 446 of the first version of the manuscript. Here we make a comparison with data from other station 47°S site in the subantarctic zone, and therefore, specifying the name of our sampling site is needed. Please see also **R2-R7**.

R2-C81: Line 405-406: what makes E. huxleyi coccolith change their lengths in early spring to summer discuss under discussion. Please refer Bollmann et al paper.

R2-R81: Authors believe that reviewer 2 refers to Bollmann and Herrle (2007, EPSL) paper where a close relationship between the length of *E. huxleyi* coccoliths and salinity is described. We appreciate reviewer 2’s comment and in the new version of the manuscript the possible effect of Sea Surface Salinity (SSS) on the observed coccolith weight and length variability is discussed (lines 966-971 of the corrected version of the manuscript with tracked changes). Moreover, SSS data for the 61°S site was obtained

from the World Ocean Atlas and is presented in section “2.5 Satellite imagery, meteorological and oceanographic data” of the new version of the publication.

R2-C82: Line 410-412: If possible, plot graphs of correlation

R2-R82: Corrected according to reviewer 2’s suggestion. A new Figure (Fig. 7) has been included in the new version of the manuscript showing the regression plots between *E. huxleyi* coccolith weight and length.

R2-C83: Line 422: South of the Polar Front>>>> South of the PF

R2-R83: Corrected according to reviewer 2’s suggestion.

R2-C84: Line 422: include recent studies carried out in the Southern Ocean such as, Patil et al., 2017, Saavedra Pellitero et al, Malinverno et al.,

R2-R84: Corrected according to reviewer 2’s suggestion. The references suggested by reviewer 2 have been incorporated in the new version of the manuscript.

R2-C85: Line 426: Buesseler et al., 2007 reference is not in the reference list

R2-R85: Corrected according to reviewer 2’s suggestion.

R2-C86: Line 435: What author mean by “coccolith particle bloom”?

R2-R86: The term “coccolith particle bloom” has been replaced by “the period of enhanced coccolith flux” in order to be clearer.

R2-C87: Line 450: use either AZ or AZ-S

R2-R87: Corrected according to reviewer 2’s suggestion. The term AZ has been replaced by AZ-S in the new version manuscript when required.

R2-C88: Line 463: general variability they found between>>>> general variability found between

R2-R88: Corrected according to reviewer 2’s suggestion.

R2-C89: Line 463: please differentiate morphological differences between Morphotypes A, B/C, var. huxleyi and var. aurorae. E. huxleyi morphotype C is usually less calcified morphotype of E. huxleyi usually found in the AZ. What are the probable reasons for absence of morphotype C in sediment trap samples?

R2-R89: Corrected according to reviewer 2’s suggestion. A description of the other morphotype found in our study region (morphotype A) has been included in section 3.2 of the new version of the manuscript with tracked changes (lines 636-656). As stated in the first version of the manuscript (Lines 463-4365), morphotype A has been documented to be genetically different than morphotype B/C. Based on this observation, Cook et al. (2011) associated these two morphologies with two varieties defined as *E. huxleyi* var. *huxleyi* and *E. huxleyi* var. *aurorae*, respectively. The text has been rephrased in order to be clearer (see line 804 of the new version of the manuscript with tracked changes). Findlay and Giraudeau (2000, Mar Mic) and Cubillos et al. (2011) analysed samples from a transect along the 140°E meridian, where our 61°S sediment trap was deployed. Findlay and Giraudeau (2000) did report *E. huxleyi* morphotype C in their samples, Young et al. (2003) reviewed their classification of *E. huxleyi* morphotypes and revised Type C specimens from the Antarctic Ocean documented by Findlay and Giraudeau (2000) to be Type B/C. This is mentioned in Young et al. (2003), Hagino et al. (2005, Mar Mic) and

Cubillos et al. (2007). So the fact that morphotype C has not been previously reported in our study region further supports our observations.

R2-C90: Plate 1: I don't agree with all six images belonging to morphotype B/C. Plate 1b, e, looks like morphotype C. Plate 1. g is unrecognizable due to overcalcification. Authors can follow Young et al., 2003 atlas for differentiating *E. huxleyi* morphotypes.

R2-R90: Authors did follow Young et al. (2003) classification, in fact, one of the co-authors (Lluïsa Cros) co-authored of the Atlas referred to by reviewer 2. Authors believe that the different morphologies observed in the coccoliths are just variations within an *E. huxleyi* B/C population. Morphotype B/C exhibits a similar morphology to types B and C (Young et al. 2003) but it is intermediate in size. However, coccolith size was considered of limited value in discriminating morphotypes by Cubillos et al. (2007) based on the large variability in size of the coccoliths on the same coccospheres. This is now clearly explained in the paper (Lines 646-656). It is also worth noting that Young et al. (2003) revised the morphotype described as C in our study area by Findlay and Giraudeau (2000) and redefined it as Type B/C.

R2-C91: Line 508: *Silicate and/or Fe*>>>> *Silicate and/or iron*

R2-R91: Corrected according to reviewer 2's suggestion.

R2-C92: Line 522, line 535: *Tagliabue et al., 2014. Year of publication missing in the reference list*

R2-R92: We believe there has been a misunderstanding because the year of the publication (2014) is mentioned was in line 1074 of the first version of the manuscript.

R2-C93: Line 533: *the part of the *Emiliana huxleyi**>>>> *the part of the *E. huxleyi**

R2-R93: Corrected according to reviewer 2's suggestion.

R2-C94: Line 598: *Trull et al., 2001*>>>> *Trull et al., 2001a or 2001b or 2001c?*

R2-R94: Corrected according to reviewer 2's suggestion. The reference of Trull et al. 2001b is not specified in the text.

R2-C95: Line 623: *both traps (2.5 pg at 2000m to 2.6 pg at 3700)*>>>> *both traps (2.5±.... pg at 2000m to 2.6±.... at 3700m)*

R2-R95: Since the annual coccolith weights are already mentioned in the results section and later on in the discussion (section 4.5 Calcium carbonate content of *Emiliana huxleyi* coccoliths), the annual coccoliths weights have been removed here in order to avoid repetition.

R2-C96: Line 634: *(Turner, 2002; Turner, 2015)*>>>> *(Turner, 2002; 2015)*

R2-R96: Corrected according to reviewer 2's suggestion.

R2-C97: Line 655: **E. huxleyi* morphotype B/C is more weakly calcified than other morphotypes*>>>> *I am not convinced with this. *E. huxleyi* morphotype C is more weakly calcified than B/C. It can be written as >> *E. huxleyi* morphotype B/C is weakly calcified than A> if authors want to tell extent of calcification.*

R2-R97: In order to be clearer, the sentence highlighted by reviewer 2 has been rephrased. Now it reads" Since *E. huxleyi* morphotype B/C is considered to be geographically restricted to the Southern Ocean (Cubillos et al., 2007; Cook et al., 2013)

we limit the comparison of our results to studies reporting this morphotype conducted only in the Southern Ocean.“ (lines 1052-1053).

R2-C98: Line 676, 682: Salter et al. (2014) missing in the reference list

R2-R98: Corrected according to reviewer 2’s suggestion. The reference is now listed in the new version of the manuscript.

R2-C99: Line 679, 683: Rembauville et al. missing in the reference list

R2-R99: Corrected according to reviewer 2’s suggestion. The reference is now listed in the new version of the manuscript.

R2-C100: Line 698: Analysis of the sediment trap materials>>> analysis of two sediment trap material

R2-R100: This sentence has been rephrased “Analysis of the materials captured by two sediment traps deployed...”. Moreover, this sentence has been split into two following reviewer 2’s suggestion R2-C6.

1 **Coccolithophore populations and their contribution to carbonate export during an**
2 **annual cycle in the Australian sector of the Antarctic Zone**

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13 **Abstract**

14 The Southern Ocean is experiencing rapid and relentless change in its physical and
15 biogeochemical properties. The rate of warming of the Antarctic Circumpolar Current
16 exceeds that of the global ocean, and the enhanced uptake of carbon dioxide is causing
17 basin-wide ocean acidification. Observational data suggest that these changes are
18 influencing the distribution and composition of pelagic plankton communities. Long-term
19 and annual field observations on key environmental variables and organisms are a critical
20 basis for predicting changes in Southern Ocean ecosystems. These observations are
21 particularly needed, since high-latitude systems have been projected to experience the
22 most severe impacts of ocean acidification and invasions of allochthonous species.

23 Coccolithophores are the most prolific calcium carbonate producing phytoplankton
24 group, playing an important role in Southern Ocean biogeochemical cycles. Satellite
25 imagery has revealed elevated particulate inorganic carbon concentrations near the major
26 circumpolar fronts of the Southern Ocean, that can be attributed to the coccolithophore
27 *Emiliana huxleyi*. Recent studies have suggested changes during the last decades in the
28 distribution and abundance of Southern Ocean coccolithophores. However, due to limited
29 field observations, the distribution, diversity and state of coccolithophore populations in
30 the Southern Ocean remain poorly characterised.

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32 We report here on seasonal variations in the abundance and composition of
 33 coccolithophore assemblages collected by two moored sediment traps deployed at the
 34 Antarctic Zone south of Australia (2000 and 3700 m depth) for one year in 2001-02.
 35 Additionally, seasonal changes in coccolith weights of *E. huxleyi* populations were
 36 estimated using circularly polarised micrographs analysed with *C-Calcita* software. Our
 37 findings indicate that (1) coccolithophore sinking assemblages were nearly monospecific
 38 for *E. huxleyi* morphotype B/C in the Antarctic Zone waters in 2001-2002; (2) coccoliths
 39 captured by the traps experienced weight and length reduction during summer (December
 40 – February); (3) the estimated annual coccolith weight of *E. huxleyi* at both sediment traps
 41 (2.11 ± 0.96 and 2.13 ± 0.91 pg at 2000 m and 3700 m) was consistent with previous
 42 studies for morphotype B/C in other Southern Ocean settings (Scotia Sea and Patagonian
 43 shelf); (4) coccolithophores accounted for approximately 2-5% of the annual, deep-ocean
 44 CaCO_3 flux. Our results are the first annual record of coccolithophore abundance,
 45 composition and degree of calcification in the Antarctic Zone. They provide a baseline
 46 against which to monitor coccolithophore responses to changes in environmental
 47 conditions expected for this region in coming decades.

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49 **Key words:** Southern Ocean, Antarctic Zone, coccolithophores, coccolith weight,
 50 sediment traps.

51

52 **1. Introduction**

53 **1.1. Background and objectives**

54 The rapid increase in atmospheric CO_2 levels since the onset of the industrial
 55 revolution is modifying the environmental conditions of marine ecosystems in a variety
 56 of ways. The enhanced greenhouse effect, mainly driven by increased atmospheric CO_2
 57 levels, is causing ocean warming (Barnett et al., 2005), shallowing of mixed layer depths
 58 (Levitus et al., 2000) and changes in light penetration and nutrient supply (Bopp et al.,
 59 2001; Rost and Riebesell, 2004; Sarmiento et al., 2004b; Deppeler and Davidson, 2017).
 60 Moreover, the enhanced accumulation of CO_2 in the ocean is giving rise to changes in the
 61 ocean carbonate system, including reduction of carbonate ion concentrations and
 62 lowering of seawater pH. Most evidence suggests that the ability of many marine
 63 calcifying organisms to form carbonate skeletons and shells may be reduced with
 64 increasing seawater acidification including some species of (but not all) coccolithophores,

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77 corals, pteropods and foraminifera (e.g. Orr et al., 2005; Moy et al., 2009; Lombard et al.,
78 2010; Beaufort et al., 2011; Andersson and Gledhill, 2013). Since phytoplankton are
79 extremely sensitive to global environmental change (Litchman et al., 2012) all predicted
80 changes in marine environmental conditions are likely to modify the abundance,
81 composition and distribution of phytoplankton communities.

82 Changes in the relative abundances of major phytoplankton functional groups are
83 likely to influence ocean biogeochemistry and ocean carbon storage, with feedbacks to
84 the rate of climate change (e.g. Boyd and Newton, 1995; Boyd et al., 1999; Falkowski et
85 al., 2004; Cermeño et al., 2008). The precipitation and sinking of CaCO₃ by
86 coccolithophores has the potential for complex contributions to carbon cycling.
87 Carbonate precipitation removes more alkalinity than dissolved inorganic carbon from
88 surface waters, thereby acting to increase pCO₂ in surface waters (the so-called carbonate
89 counter pump, e.g. Zeebe, (2012)). On the other hand, ballasting by carbonates appears
90 to increase transfer of organic carbon to the ocean interior (Armstrong et al., 2002; Klaas
91 and Archer, 2002). On seasonal timescales the counter pump contribution dominates
92 (Boyd and Trull, 2007), but more complex interactions can occur over longer timescales
93 as a result of changing extents of carbonate dissolution in sediments, including the
94 possibility that enhanced calcite dissolution in the Southern Ocean contributed to lower
95 atmospheric CO₂ levels during glacial maxima (Archer and Maier-Reimer, 1994; Sigman
96 and Boyle, 2000; Ridgwell and Zeebe, 2005).

97 The Southern Ocean is a critical component of the Earth's ocean-climate system
98 and plays a pivotal role in the global biogeochemical cycles of carbon and nutrients
99 (Sarmiento et al., 2004a; Anderson et al., 2009). Despite the fact that the Southern Ocean
100 accounts for about 25% of the global ocean, it contains ~40% of the global ocean
101 inventory of anthropogenic CO₂ (Khatiwala et al., 2009; Takahashi et al., 2009; Frölicher
102 et al., 2015), and it exports nutrients to more northern latitudes ultimately supporting ~
103 75% of the ocean primary production north of 30°S (Sarmiento et al., 2004a). Model
104 projections suggest that the reduction in the saturation state of CaCO₃ will reach critical
105 thresholds sooner in cold, high-latitude ecosystems such as the Southern Ocean (Orr et
106 al., 2005; McNeil and Matear, 2008; Feely et al., 2009). Therefore, calcifying organisms
107 living in these regions will be the first to face the most severe impacts of ocean
108 acidification.

109 In view of the rapid changes in climate and other environmental stressors presently
110 occurring in the Southern Ocean, a major challenge facing the scientific community is to

Eliminado: For example, diatoms can play a prominent role in export of organic matter from the surface ocean, because of their heavy siliceous frustules and capacity for aggregation and rapid sinking facilitates efficient transport of organic carbon (Buesseler, 1998; Smetacek, 1999), although this silica-mediated carbon export may not reach the ocean interior efficiently (Francois et al., 2002; Lam and Bishop, 2007).

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127 predict how phytoplankton communities will reorganise in response to global change. In
 128 this regard, two main aspects of the distributions of coccolithophores are emerging.
 129 Firstly, coccolithophores exhibit high concentrations in the Subantarctic Southern Ocean,
 130 a feature termed by Balch et al. (2011) as the “Great Calcite Belt” based on satellite
 131 reflectance estimates of PIC abundances. Although importantly the PIC accumulations
 132 are significantly less than those that arise in the North Atlantic, and the satellite algorithm
 133 is not reliable in Antarctic waters, where it badly overestimates PIC abundances (Balch
 134 et al., 2016; Trull et al., 2017). Secondly, recent studies suggest that the magnitude and
 135 geographical distribution of *E. huxleyi* blooms may be experiencing significant and rapid
 136 changes. Cubillos et al. (2007) and Winter et al. (2014) postulated that *E. huxleyi* has
 137 expanded its ecological niche south of the Polar Front in the recent decades.
 138 Contrastingly, Freeman and Lovenduski (2015) suggested an overall decline in Southern
 139 Ocean PIC concentrations using satellite records between 1998 and 2014. The
 140 explanation of these contrasting results may lie in the methodologies applied. While
 141 shipboard surface water observations provide a highly detailed picture of a given
 142 ecosystem, they are very sparse, only represent a snapshot in time, and can easily miss
 143 blooms of any given species. The satellite PIC signal has the great advantage of largescale
 144 and repeated coverage, but can miss subsurface populations (e.g. Winter et al., 2014) and
 145 be mimicked by the spectral characteristics of other scattering sources. The most
 146 important among them are probably microbubbles (Zhang et al., 2002), glacial flour
 147 (Balch et al., 2011) and noncalcifying organisms such as *Phaeocystis antarctica* (Winter
 148 et al., 2014), a colonial prymnesiophyte algae very abundant in high latitude systems of
 149 the Southern Ocean (e.g. Arrigo et al., 1999; Arrigo et al., 2000). Notably the PIC
 150 algorithm performs particularly poorly in Antarctic waters (Balch et al., 2016; Trull et al.,
 151 2017)

152 For these reasons, year-round field observations of areas representative of key
 153 Southern Ocean regions are essential to determine the current state of coccolithophore
 154 communities and to develop baselines against which long-term trends can be detected.
 155 Moreover, a better understanding of coccolithophore distribution, ecology and seasonal
 156 dynamics is required to improve our interpretations of the sedimentary record and our
 157 models of biogeochemistry. Sediment traps are a direct method to collect data about
 158 calcareous and siliceous micro and nanoplankton. Traps allow the monitoring of seasonal
 159 and annual variability of plankton export, document species successions, and help to
 160 determine the specific role of microplankton species in the biological and carbonate

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170 pumps. The autonomous collection capacity of sediment traps is particularly useful in the
171 remote Southern Ocean, where inaccessibility and harsh working conditions prevent year-
172 round ship-based sampling.

173 We present here the first record of composition, abundance, and seasonality of
174 coccolithophore assemblages in the Antarctic Zone of the Southern Ocean, collected by
175 two deep ocean sediment traps deployed on a single mooring during 10 months south of
176 Australia at the site of the Southern Ocean Iron Release Experiment (SOIREE) near 61°S,
177 140°E (Boyd et al., 2000a). Moreover, we report weight and length measurements on *E.*
178 *huxleyi* coccoliths, assessing the impact of seasonally varying environmental parameters
179 on *E. huxleyi* coccoliths. That provides a baseline of coccolith dimensions for the
180 populations living in this region. All the above information is needed for monitoring
181 coccolithophore responses, if any, to changing environmental conditions in the Antarctic
182 Zone south of Australia during coming decades.

183

184 1.2 Regional setting and oceanography

185 The southern Antarctic Zone (AZ-S; Parslow et al., 2001) is delimited in the north
186 by the southern branch of the Polar Front (PF) and in the south by the southern front of
187 the Antarctic Circumpolar Current (SACCF) (Fig. 1). Trull et al. (2001b) summarised the
188 seasonal evolution of water column properties in the study region. The intense heat loss
189 of surface waters during winter decreases Sea Surface Temperature (SST) to values <
190 1°C, resulting in strong vertical convection. Winter mixing extends to depths of about
191 120 m, replenishing the upper water column with nutrients. Chlorophyll-*a* levels during
192 winter are negligible throughout the region due to the reduced solar radiation and the
193 deep, continuous vertical mixing. During summer, increasing solar radiation warms the
194 surface ocean and a seasonal thermocline forms (Fig. 2). By late summer-early autumn
195 (March) SST ranges between 2 and 3 °C. Considerable nutrient depletion associated with
196 a moderate increase in algal biomass occurs within the mixed layer (Trull et al., 2001b).
197 Nonetheless, due to the limited sampling of the study region, the timing of the summer
198 nutrient minimum is not well constrained by the available data (Trull et al., 2001b).
199 Silicate exhibits the strongest summer draw-down of all the macronutrients, reaching
200 ~30% of its winter values (Fig. 2; Trull et al., 2001b), mainly due to diatom growth and
201 subsequent biogenic silica export to the deep sea (Rigual-Hernández et al., 2015a). The
202 low algal biomass accumulation in the region is attributed to the very low iron levels (0.1-

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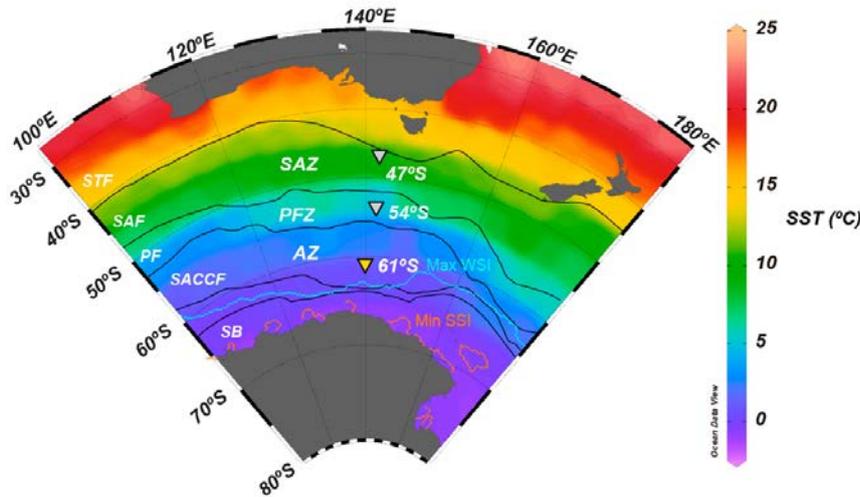
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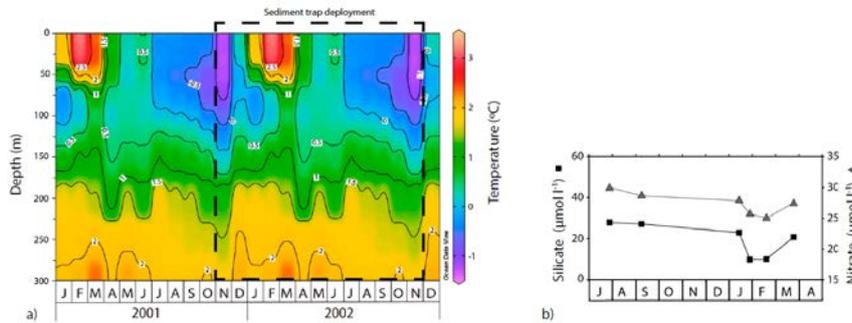
212 0.2 nM; Boyd et al., 2000a; Sohrin et al., 2000). Mesozooplankton analysis during the
 213 SOIREE experiment by Zeldis (2001) indicates that zooplankton community in the study
 214 region is dominated by copepods. Grazing pressure was low (<1% of the phytoplankton
 215 standing stock removed per day) and, therefore, is thought not to play an important role
 216 in the control of the micro-phytoplankton (primarily diatom) stocks, but nanoflagellate
 217 grazer abundances were significant and were likely to have regulated smaller



218 phytoplankton abundances (Hall and Safi, 2001).

219
 220 Figure 1. Annual mean sea surface temperature map (World Ocean Atlas; Locarnini et
 221 al., 2013) of the Australian sector of the Southern Ocean, showing the position of the
 222 main frontal and zonal systems (adapted from Orsi et al., 1995) and the location of the
 223 61°S, 54°S and 47°S sediment trap stations (inverted triangles). Abbreviations: STF -
 224 Subtropical Front, SAZ - Subantarctic Zone, SAF - Subantarctic Front, PFZ - Polar
 225 Frontal Zone, PF - Polar Front, AZ - Antarctic Zone, SACC - Southern Antarctic
 226 Circumpolar Current Front, SB - Southern Boundary, Max WSI - Maximum Winter Sea
 227 Ice Extent (August 2001) and Min SSI - Minimum Summer Sea Ice Extent (February
 228 2002) (Fetterer et al., 2017).

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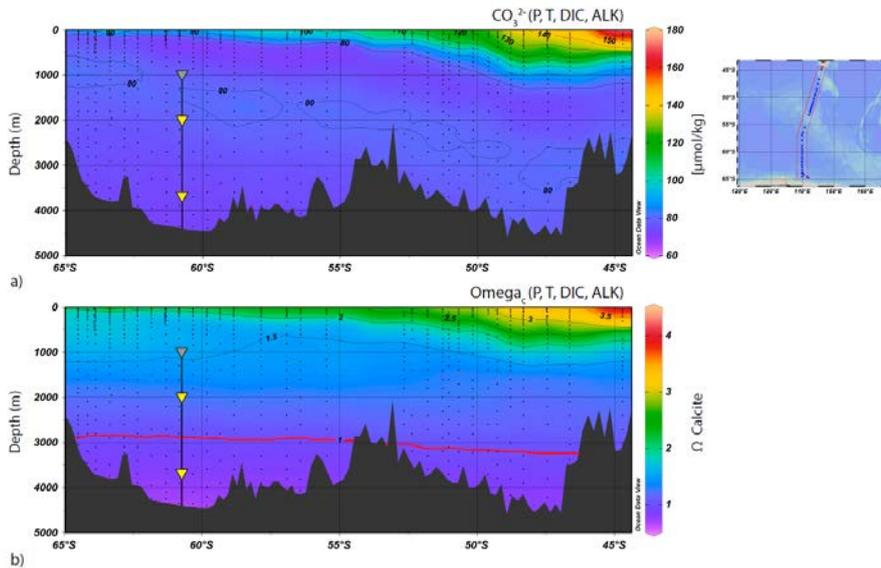
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236
 237 **Figure 2:** (a) Seasonal variation in the vertical structure of temperature (°C) between
 238 January 2001 and December 2002 for the 61°S site from the World Ocean Atlas 2009
 239 (Locarnini et al., 2010). (b) Summary of seasonal evolution of macronutrient
 240 concentrations (silicate and nitrate) in the mixed layer at the 61°S site taken from the
 241 WOCE SR3 transects between 1993 and 1996 (modified from Trull et al., 2001b) (b).

242 **1.3 Water carbonate chemistry in the study region**

243 Calcite solubility increases at higher pressures and lower temperatures, so that
 244 dissolution increases with depth in the water column. Based on downward changes in the
 245 calcite dissolution rate, two critical depth horizons can be distinguished: the Calcite
 246 Saturation Horizon (CSH) that can be defined as the depth at which the water becomes
 247 undersaturated with respect to calcite (i.e. where $\Omega_{\text{calcite}} = 1$); and the CaCO₃
 248 compensation depth (CCD), the depth at which the rate of calcite rain from the upper
 249 water column equals the dissolution rate. Figure 3 shows carbonate concentrations [CO₃²⁻
 250] and calcite saturation (Ω_{calcite}) for the WOCE SR03 2001 transect between Antarctica
 251 and Tasmania along the 140°E meridian as estimated by Bostock et al. (2011). In the AZ-
 252 S waters south of Tasmania, the CSH and CCD occur at 3000 and 3700 m, respectively
 253 (Fig. 3). Therefore, the location of sediment traps at the 61°S site allows for the
 254 assessment of dissolution changes, if any, of coccolithophore assemblages between the
 255 two critical dissolution depth horizons: the CSH and CCD. Notably, both progressive
 256 uptake of anthropogenic CO₂ and increased upwelling of naturally CO₂-rich deep waters
 257 over the past 20 years is leading to shallowing of these features (Pardo et al., 2017)



264

265 [Figure 3. Cross section of the mooring location in comparison to regional seafloor](#)
 266 [bathymetry, carbonate concentrations \$\[\text{CO}_3^{2-}\]\$ and calcite saturation \(\$\Omega_{\text{calcite}}\$ \) for WOCE](#)
 267 [transect SR03 2001 from Bostock et al. \(2011\), who calculated them from the Dissolved](#)
 268 [Inorganic Carbon \(DIC\) and alkalinity in the CARINA database \(CARINA, 2011\). The](#)
 269 [location of the transects is shown on the map on the right top. \$\Omega_{\text{calcite}} = 1\$ contour is](#)
 270 [highlighted with a red line to show the approximate depth of the CSH across the transect.](#)

271

272 2. Material and Methods

273 2.1 Sediment trap experiment

274 As part of the SAZ collaborative research program (Trull et al., 2001c), a sediment
 275 trap experiment was carried out at the 61°S site (60° 44.43'S; 139° 53.97'E) in the
 276 Australian sector of the southern Antarctic Zone within the region where the Southern
 277 Ocean Iron Release Experiment (SOIREE) was conducted (Boyd et al., 2000a). The 61°S
 278 site is characterized by weak currents with a mean eastward geostrophic surface velocity
 279 of approximately $0.03 \pm 0.02 \text{ m s}^{-1}$ (Trull et al., 2001b). The site is north of the Seasonal
 280 Sea-Ice Zone (Massom et al., 2013; Rigual-Hernández et al., 2015a) and remote from any
 281 known iceberg pathway (Gladstone et al., 2001).

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Movido hacia arriba[1]: 2.1 Regional setting and oceanography ¶

The southern Antarctic Zone (AZ-S; Parslow et al., 2001) is delimited at the north by the southern branch of the Polar Front (PF) and at the south by the southern front of the Antarctic Circumpolar Current (SAACF). Trull et al. (2001b) summarized the seasonal evolution of water column properties in the study region. The intense heat loss of surface waters during winter decreases Sea Surface Temperature (SST) to values $< 1^\circ\text{C}$, resulting in strong vertical convection. Winter mixing extends to depths of about 120 m, replenishing the upper water column with nutrients. Chlorophyll-*a* levels during winter are negligible throughout the region due to the reduced solar radiation and the deep, continuous vertical mixing. During summer, increasing solar radiation warms the surface ocean and a seasonal thermocline forms (Fig. 2). By late summer (March) SST ranges between 2 and 3°C . Considerable nutrient depletion associated with a moderate increase in algal biomass occurs within the mixed layer. Nonetheless, due to the limited sampling of the study region, the timing of the summer nutrient minimum is not well constrained by the available data (Trull et al., 2001b). Silicate exhibits the strongest summer draw-down of all the macronutrients, reaching $\sim 30\%$ of its winter values (Fig. 2; Trull et al., 2001), mainly due to diatom growth and subsequent biogenic silica export to the deep sea (Rigual-Hernández et al., 2015a). The low algal biomass accumulation in the region is attributed to the very low iron levels (0.1–0.2 nM; Boyd et al., 2000a; Sohrin et al., 2000). Mesozooplankton analysis during the SOIREE experiment by Zeldis (2001) indicates that zooplankton community in the study region is dominated by copepods, mainly large calanoid copepodites. Grazing pressure was low ($< 1\%$ of the phytoplankton standing stock removed per day) and, therefore, is thought not to play an important role in the control of the micro-phytoplankton (primarily diatom) stocks, but nanoflagellate grazer abundances were significant and were likely to have regulated smaller phytoplankton abundances (Hall and Safi, 2001). ¶

Figure 1. Annual mean sea surface temperature map (World Ocean Atlas; Locarnini et al., 2013) of the Australian sector of the Southern Ocean, showing the position of the main frontal and zonal systems (adapted from Orsi et al., 1995) and the location of the 61°S, 54°S and 47°S sediment trap stations (inverted triangles). Abbreviations: STF - Subtropical Front, <objeto>¶

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2.2 Water carbonate chemistry ¶

Calcite solubility increases at higher pressures and lower temperatures, so that dissolution increases with depth in the water column. Based on downward changes in the calcite dissolution rate, two critical depth horizons can be

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435 The mooring was equipped with three McLane Parflux time series sediment traps (Honjo
436 and Doherty, 1988) for approximately one year (November 30, 2001 to September 29,
437 2002, 317 days). The traps were located at 1000, 2000 and 3700 m below the surface in
438 a water column of 4393 m (Figures 3a and 3b). Each trap was provided with 21 cups.
439 Sampling intervals were synchronised between traps and in order to resolve the seasonal
440 flux cycle ranged from 8 days (in austral summer) to 55 days in austral winter. No samples
441 were recovered from the shallowest trap owing to equipment malfunction and, therefore,
442 only results for the 2000 and 3700 m traps are presented here. Each trap was paired with
443 an Aanderaa current meter and temperature sensors. The 250 ml collection cups were
444 filled with a buffered solution of sodium tetraborate (1 g L^{-1}), sodium chloride (5 g L^{-1}),
445 strontium chloride (0.22 g L^{-1}), and mercury chloride (3 g L^{-1}) in unfiltered, deep (> 1000
446 m) seawater from the region. [Risk of sample contamination by the unfiltered seawater is](#)
447 [considered negligible due to the fact that the deep water exhibits low particle abundance](#)
448 [and also because particle concentration in sea water is of the order of \$\mu\text{g/L}\$ while](#)
449 [concentration in the trap cups after recovery was of the order of \$\text{mg/L}\$.](#)

450 The two deeper traps completed their collection sequence as programmed,
451 providing continuous time-series for a year. Due to the low particle fluxes during the
452 winter, insufficient material remained for phytoplankton analysis of cup 1 from the 2000
453 m trap and cups 1, 2, 19, 20 and 21 from the 3700 m trap (Table 1).

455 2.2. Sample processing and coccolithophore counting procedure

456
457 The sediment trap cup contents were washed through a 1 mm sieve after recovery
458 and then divided into 10 aliquots using a rotary splitter (McLane, Inc.). A description of
459 the analytical procedures for estimation of geochemical fluxes is provided in Trull et al.
460 (2001a) and Rigual-Hernández et al. (2015a). One aliquot was used for siliceous and
461 calcareous micro- and nano-plankton analyses. Each fraction for plankton analysis was
462 refilled with distilled water to 40 ml, from which 10 ml was subsampled and buffered
463 with a solution of sodium carbonate and sodium hydrogen carbonate (pH 8) and kept
464 refrigerated for calcareous nannoplankton analysis. Samples for coccolithophore analysis
465 were prepared following the methodology of Flores and Sierro (1997). In short, 300 μl
466 were extracted with a micropipette and dropped onto a glass Petri dish previously filled
467 with a buffered solution and with a cover slip on its bottom. After settling for 12 hours,
468 the buffer solution was removed using short strips of filter paper placed at the edge of the

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475 dish. Then, the cover slip was left to dry completely and mounted on a glass slide using
476 Canada balsam. Coccoliths were identified and counted using a Nikon Eclipse 80i
477 polarised light microscope at 1000× magnification. A minimum of 400 coccoliths were
478 counted in each sample. Cocospheres occurred in much lower numbers than loose
479 coccoliths in these preparations. The coccolith counts were transformed into daily fluxes
480 of specimens m² d⁻¹ following the formula:

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$$F = \frac{N \times \frac{A}{n \times a} \times V \times S}{d \times T}$$

482

483

484 where “*F*” is the daily coccolith flux, “*N*” the number of coccoliths, “*A*” the total area of
485 a Petri dish, “*n*” the number of fields-of-view analysed, “*a*” the area of a single field of
486 view, “*V*” the dilution volume, “*S*” the split of the cup, “*d*” the number of days of
487 collection and “*T*” the aperture area of the sediment trap.

488 Since the sediment trap collection period was shorter than a full calendar year, an
489 estimate of the annual coccolith flux of the 2000 m trap was calculated. This estimate
490 takes into account the fact that the unsampled days occurred in winter when particle fluxes
491 were low, and were obtained using the flux for the last winter cup (#21 in 2002) to

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492 represent mean daily fluxes during the unobserved interval. Due to the lack of samples
493 corresponding to winter 2002 for the 3700 m sediment trap record, the annualization of
494 the coccolith fluxes for this trap was made based only on the samples with available data.

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495 Therefore, the annualised and annual flux data for the 3700 m trap presented in Table 1
496 should be used with caution.

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61_2000		Sampling period	Length	Total Mass Flux		CaCO ₃	POC		Diatoms	Coccolithophore flux	Relative abundance		
Cup	mid point	days	mg m ⁻² d ⁻¹	mg m ⁻² d ⁻¹	%	mg m ⁻² d ⁻¹	%	10 ⁶ valves m ⁻² d ⁻¹	10 ⁶ coccoliths m ⁻² d ⁻¹	<i>E. huxleyi</i>	<i>C. leptoporus</i>	Other	
1	nov. 30, 2001	8	48	14	30	0.7	1.5	-	-	-	-	-	
2	dic. 08, 2001	8	78	17	22	1.7	2.2	9	2.5	98.8	1.2	0.0	
3	dic. 16, 2001	8	326	62	19	6.9	2.1	82	2.7	98.5	1.3	0.2	
4	dic. 24, 2001	8	509	140	28	6.4	1.3	85	8.2	99.5	0.5	0.0	
5	ene. 01, 2002	8	1151	44	4	26.9	2.3	408	12.3	99.8	0.2	0.0	
6	ene. 09, 2002	8	1069	170	16	14.8	1.4	200	22.3	99.8	0.2	0.0	
7	ene. 17, 2002	8	656	60	9	11.3	1.7	159	9.2	99.3	0.7	0.0	
8	ene. 25, 2002	8	702	38	5	11.0	1.6	296	8.4	99.3	0.7	0.0	
9	feb. 02, 2002	8	666	39	6	12.0	1.8	184	5.4	98.8	1.2	0.0	
10	feb. 10, 2002	8	595	24	4	8.2	1.4	295	6.0	99.5	0.5	0.0	
11	feb. 18, 2002	8	534	20	4	6.2	1.2	149	9.8	99.0	0.5	0.5	
12	feb. 26, 2002	8	524	19	4	4.7	0.9	152	5.0	100.0	0.0	0.0	
13	mar. 06, 2002	8	586	15	3	6.9	1.2	120	6.4	99.8	0.2	0.0	
14	mar. 14, 2002	8	285	11	4	3.2	1.1	71	2.0	99.8	0.2	0.0	
15	mar. 22, 2002	8	290	7	3	3.2	1.1	66	2.0	97.6	1.0	1.5	
16	mar. 30, 2002	8	263	8	3	2.6	1.0	87	0.9	99.2	0.8	0.0	
17	abr. 08, 2002	10	264	7	3	2.2	0.8	97	1.3	98.1	1.9	0.0	
18	may. 08, 2002	50	130	5	4	1.2	1.0	47	0.8	99.8	0.2	0.0	
19	jun. 29, 2002	54	65	2	4	0.7	1.0	10	0.7	98.8	0.8	0.4	
20	ago. 22, 2002	55	56	2	4	0.8	1.5	19	0.9	99.5	0.2	0.2	
21	sep. 29, 2002	20	42	2	4	0.5	1.3	6	0.9	98.0	2.0	0.0	
Annualised values			232	17	7.4	3.3	1.4	67	2.8				
Annual flux			85 g m ⁻² y ⁻¹	6 g m ⁻² y ⁻¹		1.2 g m ⁻² y ⁻¹		24 10 ⁶ valves m ⁻² y ⁻¹	1.03 10 ¹¹ coccoliths m ⁻² y ⁻¹	99.4	0.5	0.1	

61_3700		Sampling period	Length	Total Mass Flux		CaCO ₃	POC		Diatoms	Coccolithophore flux	Relative abundance		
Cup	mid point	days	mg m ⁻² d ⁻¹	mg m ⁻² d ⁻¹	%	mg m ⁻² d ⁻¹	%	10 ⁶ valves m ⁻² d ⁻¹	10 ⁶ coccoliths m ⁻² d ⁻¹	<i>E. huxleyi</i>	<i>C. leptoporus</i>	Other	
1	nov. 30, 2001	8	38	9	23	0.4	1.1	-	-	-	-	-	
2	dic. 08, 2001	8	31	9	28	0.4	1.2	-	-	-	-	-	
3	dic. 16, 2001	8	99	29	30	1.4	1.4	4	1.3	99.0	0.7	0.2	
4	dic. 24, 2001	8	231	59	26	1.4	0.6	12	5.5	99.3	0.5	0.2	
5	ene. 01, 2002	8	873	87	10	17.3	2.0	118	11.6	99.8	0.2	0.0	
6	ene. 09, 2002	8	1157	154	13	19.8	1.7	479	15.9	100.0	0.0	0.0	
7	ene. 17, 2002	8	828	166	20	9.4	1.1	354	20.0	100.0	0.0	0.0	
8	ene. 25, 2002	8	490	34	7	6.4	1.3	169	11.0	99.8	0.2	0.0	
9	feb. 02, 2002	8	491	32	6	6.5	1.3	385	4.6	100.0	0.0	0.0	
10	feb. 10, 2002	8	419	19	4	6.0	1.4	281	4.2	99.8	0.2	0.0	
11	feb. 18, 2002	8	584	36	6	6.2	1.1	254	15.9	99.1	0.7	0.2	
12	feb. 26, 2002	8	581	31	5	5.2	0.9	238	12.2	100.0	0.0	0.0	
13	mar. 06, 2002	8	849	23	3	7.6	0.9	326	15.0	99.8	0.2	0.0	
14	mar. 14, 2002	8	369	18	5	3.3	0.9	44	6.6	99.2	0.8	0.0	
15	mar. 22, 2002	8	218	8	4	2.6	1.2	32	6.6	99.5	0.2	0.2	
16	mar. 30, 2002	8	258	10	4	2.5	1.0	43	6.8	99.3	0.7	0.0	
17	abr. 08, 2002	10	257	9	3	2.3	0.9	32	4.8	99.5	0.2	0.2	
18	may. 08, 2002	50	118	5	4	1.2	1.0	8	1.2	99.8	0.0	0.2	
19	jun. 29, 2002	54	0	0	4	0.0	1.0	-	-	-	-	-	
20	ago. 22, 2002	55	0	0	4	0.0	1.0	-	-	-	-	-	
21	sep. 29, 2002	20	0	0	4	0.0	1.0	-	-	-	-	-	
Annualised values			188	17	9	2.3	1.2	62	3.3				
Annual flux			69 g m ⁻² y ⁻¹	6 g m ⁻² y ⁻¹		0.9 g m ⁻² y ⁻¹		23 10 ⁶ valves m ⁻² y ⁻¹	1.20 10 ¹¹ coccoliths m ⁻² y ⁻¹	99.7	0.2	0.1	

501 **Table 1:** Daily export fluxes of total mass flux, calcium carbonate (CaCO₃), particulate
502 organic carbon (POC), diatom valves and coccoliths registered at the 61°S site from
503 November 2001 through October 2002. Mass fluxes listed as zero were too small to
504 measure (<1 mg).

505
506 **2.3 SEM analysis**

507 As the resolution of the light microscope is insufficient to differentiate *E. huxleyi*
508 morphotypes, the samples of the 2000 m trap record were analysed using a Scanning
509 Electron Microscope (SEM). Glass cover-slips were prepared following the method
510 outlined by Flores and Siervo (1997). The dried cover-slips were mounted on aluminium
511 stubs and coated with gold. An EVO HD25 SEM (Carl Zeiss) was used to determine the
512 morphotype of *E. huxleyi* coccoliths found in the samples (magnification 5000-20000x).
513 Due to the large abundance of diatom valves and the scarcity of coccoliths in the samples,
514 a compromise between number of identified coccoliths and time spent had to be reached.
515 Therefore, a target minimum of thirty *E. huxleyi* coccoliths per sample were identified.
516 The taxonomic concepts of Young and Westbroek (1991), Young et al. (2003), Cubillos

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527 et al. (2007) and Hagino et al. (2011) were followed to classify the *E. huxleyi* coccoliths
528 into different morphotypes.

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530 2.4 C-*Calcita* analyses

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531 The glass slides used for coccolith counts were also analysed for coccolith mass
532 and size measurements using a Nikon Eclipse LV100 POL polarised light microscope
533 equipped with circular polarization and a Nikon DS-Fi1 8-bit colour digital camera.
534 Calibration images were performed on an apical rhabdolith of the genus *Acanthoica*
535 collected by a sediment trap at the 47°S site (46°48'S, 142°6'E), located in the Australian
536 sector of the Subantarctic Zone. Camera parameters and microscope light settings were
537 maintained constant throughout the imaging session. Depending on coccolith
538 concentration, between 13-28 random fields of view per sample were photographed. The
539 images were then analysed by the image processing software *C-*Calcita** (Fuentes et al.,
540 2014). The output files for single *E. huxleyi* coccoliths were visually selected. Length and
541 weight measurements were automatically performed by *C-*Calcita** software. A total of
542 2328 coccoliths were analysed with a minimum of 50 coccoliths per sample. For more
543 methodological details see Fuentes et al. (2014).

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544 An estimated range of annual contributions of coccoliths to total CaCO₃ export
545 was calculated for the 2000 m trap record by multiplying the coccolith flux of each
546 sampling interval by the maximum and minimum standard deviations of coccolith weight
547 values measured on each sample. Then, the minimum and maximum estimates of
548 coccolith-CaCO₃ fluxes for each sampling interval (i.e. cup) were used to estimate the
549 minimum and maximum annual contribution of coccoliths to total carbonate following
550 the same procedure as for the annual coccolith fluxes.

552 2.5 Satellite imagery, meteorological and oceanographic data

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553 Weekly mean SST for the 2001-2002 interval were obtained from the NOAA
554 Optimum Interpolation SST Analysis database (Reynolds et al., 2002). Seasonal SST
555 variation range was low, with maximum SSTs of 2.94 °C observed during March 2002
556 and minimum of 0.12 °C, in early October 2002. SST variations mirrored changes in the
557 vertical structure of the water column temperature profile (Fig. 4) that displayed vertical
558 homogeneity of the water column in autumn and winter and a seasonal thermocline during
559 the austral summer (Fig. 2b).

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569 [Sea surface salinity \(SSS\) climatology for the study site was obtained from the NOAA](#)
570 [World Ocean Atlas 2005](#) (Antonov et al., 2006). [SSS exhibited very low seasonal](#)
571 [variability with values ranging between 33.7 and 33.9 psu.](#)

572 Photosynthetically [Active Radiation](#) (PAR), monthly chlorophyll-*a* concentration
573 and [Particulate Inorganic Carbon](#) (PIC) concentration estimates were obtained from
574 NASA's Giovanni program (Acker and Leptoukh, 2007) (Fig. 4) for the region: 130°E,
575 62.5°S, 150°E, 59.5°S. Chlorophyll-*a* concentration was low throughout the year (ranging
576 from 0.07 to 0.30 mg m⁻³) and in line with previous observations in the study region (Trull
577 et al., 2001b). Algal biomass responded rapidly to the solar radiation increase in
578 September 2001 and reached its highest levels in November 2001 (Fig. 4). Chlorophyll-*a*
579 concentration declined throughout the summer, reaching negligible values in autumn
580 and winter (i.e. from March to August 2002). Satellite-derived PIC concentration
581 exhibited a clear seasonal pattern similar to that of the chlorophyll-*a* with peak
582 concentrations in November (up to 0.003 mol m⁻³) and values below detection limit in
583 winter (Fig. 4).

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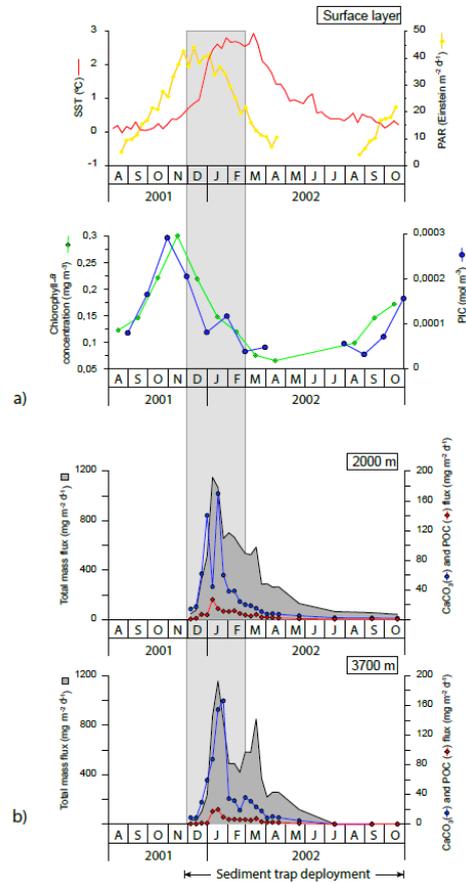
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590 **Figure 4:** (a) Satellite-derived SST (°C), PAR (Einstein m⁻² d⁻¹), chlorophyll-*a*
 591 concentration (mg m⁻³) and PIC concentration (mol m⁻³) for the period November 2001
 592 to September 2002. It is important to note that satellite PIC concentration estimates have
 593 been reported to be biased for high latitudes systems of the Southern Ocean where the
 594 satellite algorithm is thought to produce overestimates (Balch et al., 2016; Trull et al.,
 595 2017). Therefore PIC data presented here should be viewed with caution. (b) Temporal
 596 variability of the total mass, CaCO₃ and POC the < 1mm fraction at 2000 and 3700 m
 597 water depth from November 2001 through to November 2002 at the 61°S site (Rigual-
 598 Hernández et al., 2015a). Grey strips represent summer.

599

600 **3. Results**

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611 **3.1 Seasonal dynamics of coccolith export fluxes**

612 Coccolith fluxes showed a pronounced seasonal pattern at both sediment trap
613 depths, roughly following the chlorophyll-*a* dynamics in the surface layer with maximum
614 fluxes during the austral summer and minima during winter (Fig. 4 and 5). The summer
615 coccolith flux exhibited a bimodal distribution with a major peak registered in early
616 January (2.2×10^9 coccoliths $m^{-2} d^{-1}$ at 2000 m) and a secondary maximum recorded in
617 mid-February (9.8×10^8 coccoliths $m^{-2} d^{-1}$). Coccolith flux was low in autumn and winter
618 (down to $\sim 7 \times 10^7$ coccoliths $m^{-2} d^{-1}$). Coccolith fluxes in the deeper trap (3700 m)
619 followed a similar pattern to that in the 2000 m trap with a delay of about one sampling
620 interval.

621 The fluxes of all biogeochemical components were closely correlated (Table 2 in
622 Rigual-Hernández et al., 2015a). Coccolith fluxes at both traps were broadly in line with
623 biogenic particle fluxes estimated by Rigual-Hernández et al. (2015a) showing strongest
624 correlations with biogenic silica ($R^2 = 0.74$ at 2000 m and $R^2 = 0.71$ at 3700 m), followed
625 by PIC ($R^2 = 0.62$ at 2000 m and $R^2 = 0.47$ at 3700 m) and POC ($R^2 = 0.56$ at 2000 m
626 and $R^2 = 0.41$ at 3700 m).

627 Coccolithophore sinking assemblages captured by the traps were nearly
628 monospecific, with an overwhelming dominance of *E. huxleyi* that represented >99% of
629 the annual coccolith sinking assemblage at both trap depths. Background concentrations
630 of *Calcidiscus leptoporus* (*sensu lato*), *Gephyrocapsa* spp. and *Helicosphaera* spp. were
631 also registered, together representing 0.6% and 0.3% of the coccolith assemblage at 2000
632 and 3700 m, respectively, of the total annual coccolith fluxes (Table 1). The seasonal
633 changes in the coccolithophore species flux and relative abundance can be found in
634 Supplementary Figure 1. The seasonal pattern of *C. leptoporus* and *Gephyrocapsa* spp.
635 followed that of *E. huxleyi* with peak values during the summer and minima during
636 winter. The numbers of coccospheres found in the samples were negligible in both
637 sediment trap records.

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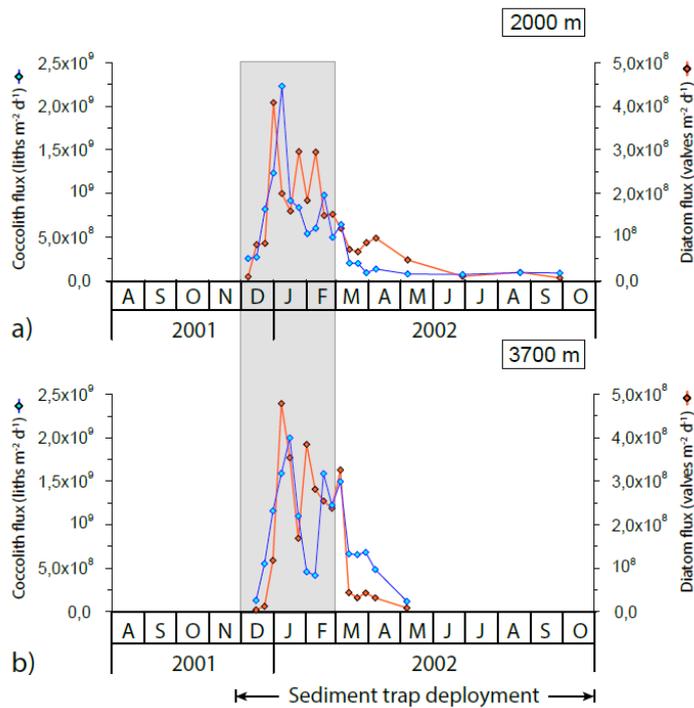
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647 **Figure 5:** Seasonal variation of total coccolith and diatom valve flux at the 2000 and 3700
 648 m sediment traps at the 61°S site. Grey strips represent summer.

649

650 **3.2 SEM analyses**

651 *Emiliana huxleyi* coccoliths correspond to morphotype B/C, having proximal
 652 shields slightly wider than the distal ones and with a central area usually filled by several
 653 (usually 5 to 11) flat, wide and thin tile-like elements (see Plate 1, image a). Distal shields
 654 of several are partially missing, most likely due to the slender and delicate structure of
 655 the laths. Distal shield measures ranged between 2 to 4.35 μm in the samples recovered
 656 from the 2000 m sediment trap. The coccoliths captured by the traps were clearly different
 657 than those of morphotype A which is the other morphotype that has been reported in the
 658 Australian sector of the Southern Ocean (Cubillos et al., 2007). Morphotype A has a
 659 central area composed of curved elements (Young et al., 2003) and its distal shields
 660 elements are often straight or concave (Cubillos et al., 2007) and more robust than those
 661 of B/C (Young et al., 2003). Since the size of the coccoliths has been reported to vary

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670 [significantly on the same coccosphere, coccolith size was not used as a discriminatory](#)
671 [feature to differentiate between morphotypes following Cubillos et al. \(2007\).](#)

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672 It is conspicuous that most of the coccoliths display a morphology which is
673 compatible with a secondary recrystallisation. Small spherules like recrystallisations are
674 present on these coccoliths, especially on the laths (Plate 1, images c-f). However, some
675 coccoliths, mostly from cup 10 (February) have no spherules covering them (Plate 1,
676 images a and b). [Aside from this sample, no relationship between the morphology of the](#)
677 [coccoliths and collection time was found.](#) These coccoliths present very thin slender laths
678 (usually from 20 to 26) and wider central areas than the coccoliths having spherules.

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680 3.3 Coccolith weight and length changes

681 Average coccolith weight at both sediment trap depths exhibited a clear seasonal
682 pattern with high values (2.28 ± 1.16 and 2.09 ± 0.80 pg/coccolith at 2000 m and 3700
683 m, respectively) at the onset of the [coccolithophore productive period](#) in early spring,
684 followed by a pronounced decrease (down to 1.65 ± 0.63 and 1.88 ± 0.63 pg at 2000 m
685 and 3700 m, respectively) in approximately January – early February. Average coccolith
686 weight followed a gradual increasing trend from approximately mid-February into winter,
687 reaching values up to 2.71 ± 1.20 pg in August 2002 at 2000 m and up to 2.43 ± 1.00 in
688 May at 3700 m, respectively. Average annual coccolith weight was 2.11 ± 0.96 and 2.13
689 ± 0.91 pg at 2000 and 3700 m, respectively. The annual amplitude of [the mean coccolith](#)
690 [weight](#) was approximately 1 pg at 2000 m and 0.5 pg at 3700 m. The lower annual
691 amplitude exhibited by the coccolith assemblages captured at the 3700 m trap is attributed
692 to the lower sampling duration at that depth over the winter season.

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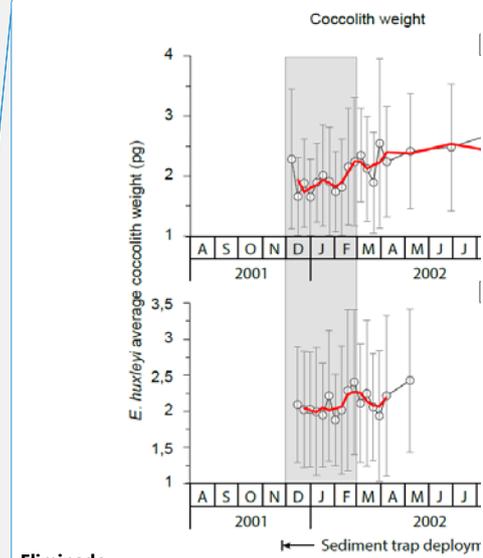
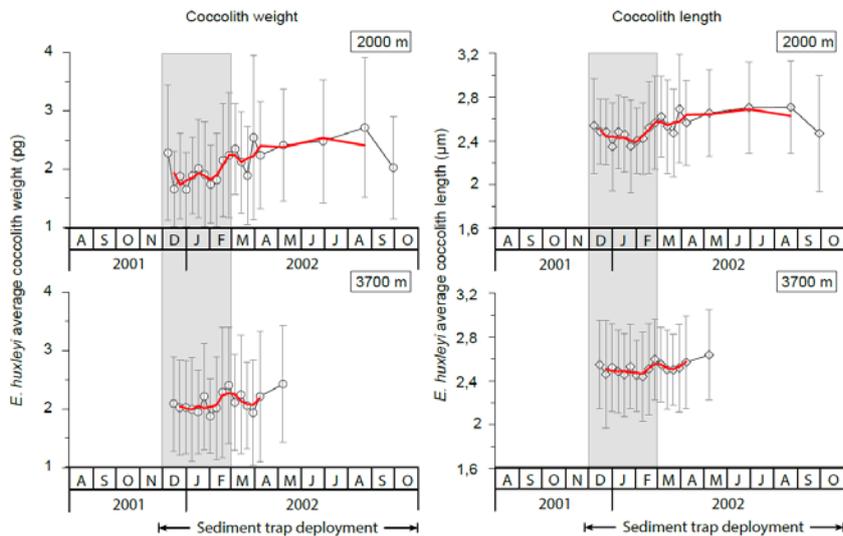
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693 Mean coccolith length was greatest in early spring 2001 (2.54 ± 0.44 and $2.55 \pm$
694 0.40 μm at 2000 and 3700 m, respectively), followed by a decrease in early summer
695 (down to 2.35 ± 0.43 and 2.44 ± 0.41 μm at 2000 and 3700 m, respectively) (Fig. 6). From
696 late February coccolith length increased again reaching the highest values of the record
697 in winter 2002 (up to 2.71 ± 0.42 and 2.64 ± 0.41 μm at 2000 and 3700 m, respectively).

698 Seasonal variations of coccolith length and weight exhibited a strong correlation
699 at both depths ($R^2 = 0.84$, $n = 20$ at 2000 m; $R^2 = 0.61$, $n = 16$ at 3700m), indicating a
700 clear, dependable relationship between the two variables (Fig. 7).

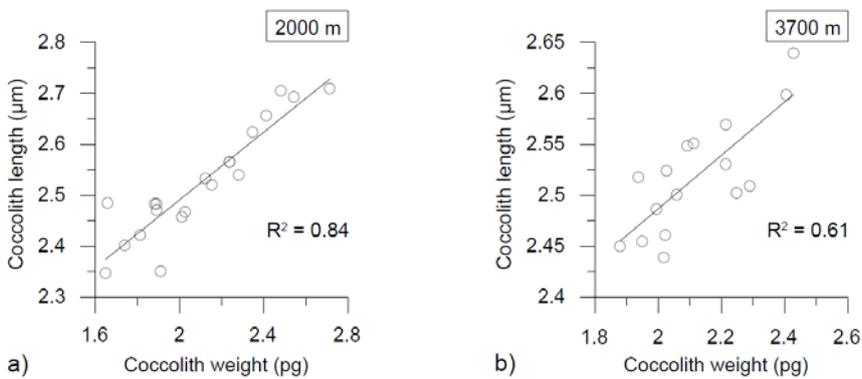
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731 **Figure 6:** Mean and standard deviation of coccolith weight and length over the sediment
 732 trap deployment period at 2000 m and 3700 m at the 61°S site. The red solid line
 733 represents a 3-point running average. Grey strips represent summer.



734

735 **Figure 7:** Regression plots between *E. huxleyi* coccolith mass (pg) and length (µm) at the
 736 2000 m (a) and 3700 m (b) sediment traps.

737 **4. Discussion**

738 **4.1 Origin, magnitude and composition of the coccolithophores**

739 Since there is a current debate about the potential expansion of *E. huxleyi*
 740 populations south of the PF, during recent decades (Cubillos et al., 2007; Saavedra-

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744 Pellitero et al., 2014; Winter et al., 2014; Malinverno et al., 2015; Patil et al., 2017), it is
745 important to evaluate the likely origins of the sinking coccolith assemblages collected at
746 station 61°S. This assessment is particularly needed in the case of deep-moored,
747 sediment-trap experiments because the source area of the particles collected by the traps
748 can be as wide as hundreds of square kilometres (Buesseler et al., 2007).

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749 Several lines of evidence strongly suggest that the coccolithophore fluxes
750 registered by the traps were produced in waters of the Antarctic Zone. Firstly, the mooring
751 was deployed in a quiescent area of the AZ-S (Trull et al., 2001b), between the stronger
752 flows associated with the southern branch of the PF and the SACCF (Fig. 1). The
753 relatively weak currents around the sediment trap location greatly reduce the area of likely
754 origins of the particles intercepted by the traps, i.e. the statistical funnel (Siegel and
755 Deuser, 1997; Siegel et al., 2008). Moreover, the large magnitude of the coccolith export
756 fluxes at both depths, plus the long duration of the period of enhanced coccolith flux
757 (about 3 months), rule out the likelihood of a transient lateral transport event (e.g.,
758 transport by mesoscale eddies) of a coccolithophore bloom produced in more northerly
759 latitudes. Lastly, the composition of the biogeochemical fluxes and diatom assemblages
760 collected by the traps are characteristic of AZ waters (Rigual-Hernández et al., 2015a),
761 further supporting the idea that the coccolithophores captured by the traps were produced
762 close to the site. All this clearly indicates that in 2001 *E. huxleyi* was an established
763 member of the phytoplankton communities of the Antarctic Zone to the south of
764 Australia.

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765 The annual coccolith export to the deep ocean at the 61°S site (1.03×10^{11}
766 coccoliths $m^{-2} yr^{-1}$) is one sixth that registered by Wilks et al. (2017) (6.5×10^{11} coccolith
767 $m^{-2} yr^{-1}$) in the SAZ waters (station 47°S; Fig. 1) north of the study site. The lower
768 abundance of coccolithophores at the sampling site is most likely due to the negative
769 effects of low temperature and low light levels on coccolithophore growth (Paasche,
770 2002; Boyd et al., 2010), but important also is the competitive advantage of diatoms over
771 coccolithophores in the silicate-rich waters of the AZ-S. The lower coccolithophore
772 production in the AZ-S is also reflected in the lower carbonate export at this site, i.e. 6 g
773 $m^{-2} yr^{-1}$ versus 10-13 g $m^{-2} yr^{-1}$ at the 47°S site (Rigual-Hernández et al., 2015b; Wilks et
774 al., 2017). The non-proportional latitudinal change in coccolith and carbonate fluxes (i.e.
775 sixfold versus twofold changes, respectively) is most likely due to variations in the
776 contribution of heterotrophic calcifiers (i.e. foraminifers and pteropods) to total carbonate
777 export. There are also differences in the carbonate content per coccolith of the

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784 coccolithophore species and the morphotypes of *E. huxleyi* dwelling in each zonal system.
785 Indeed, mean coccolith weight can vary up to two orders of magnitude between small
786 species such as *E. huxleyi* (2-3.5 pg) and large and heavily calcified taxa such as
787 *Coccolithus pelagicus* (~150 pg) (Giraudeau and Beaufort, 2007). Intraspecific size
788 variability is also common in most coccolithophore species, mainly due to growth
789 variations driven by different environmental factors and by genotypic variability (e.g.
790 Knappertsbusch et al., 1997; Poulton et al., 2011).

791 Based on the significant genetic variability found between Southern Ocean
792 populations of morphotypes A and B/C, Cook et al. (2011) classified these morphotypes
793 as *E. huxleyi* var. *huxleyi* and *E. huxleyi* var. *aurorae*, respectively. Since only
794 morphotype B/C had been reported at and south of the Antarctic Polar Front, Cook et al.
795 (2013) concluded that the rapid drop in water temperature occurring at the Antarctic Polar
796 Front may act as an open-ocean barrier to gene flow between these the two Southern
797 Ocean *E. huxleyi* morphotypes/varieties. The nearly monospecific coccolith assemblages
798 of *E. huxleyi* morphotype B/C collected by the 61°S site traps (Plate 1) are consistent with
799 those studies and supports the idea that the physiological differences in light-harvesting
800 pigments of morphotype B/C compared to other *E. huxleyi* varieties (Cook et al., 2011)
801 may represent a critical ecological advantage in the cold and low-light waters of the AZ_
802 S south of Australia.

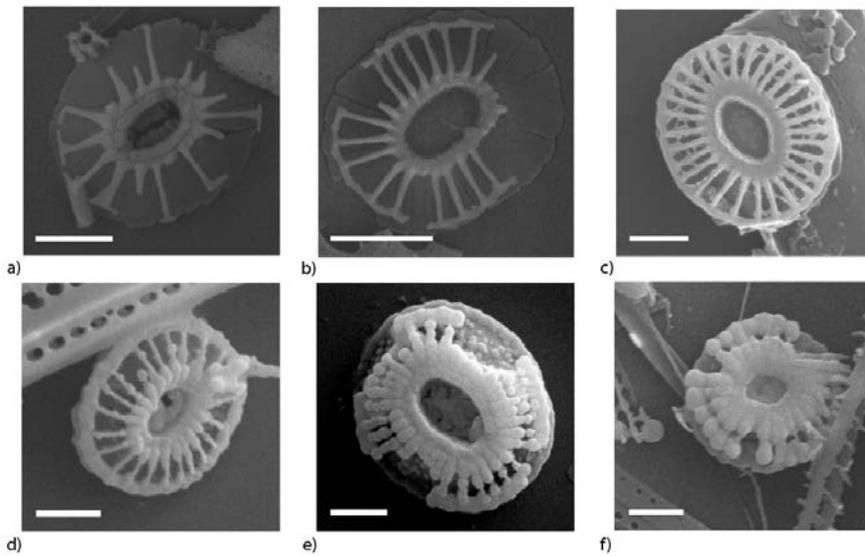
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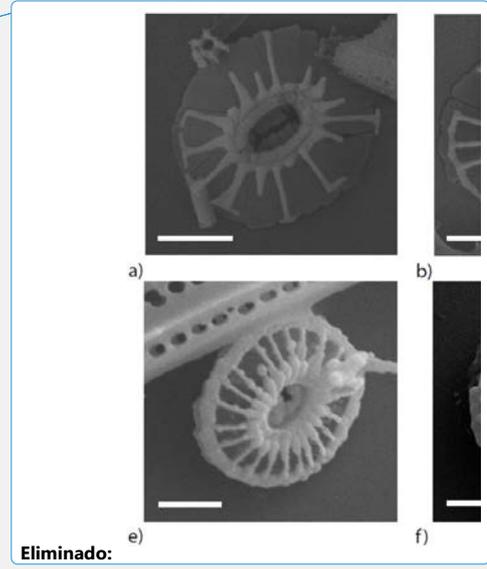
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809 **Plate 1:** SEM photos showcasing the different morphologies of *Emiliana huxleyi*
 810 morphotype B/C coccoliths found in the sediment traps of the 61°S site. Scale bars =1
 811 μm.

812

813 **4.2 Seasonal dynamics of the calcareous and siliceous phytoplankton fluxes**

814 The eight-day sampling resolution during spring and summer enabled us to
 815 monitor the detailed temporal dynamics of phytoplankton fluxes at the 61°S site.
 816 Comparison of satellite-derived PIC and [chlorophyll-a](#) concentrations for the study region
 817 with coccolith fluxes registered by the sediment trap suggests a time lag of about two
 818 months between their surface maxima and peak coccolith fluxes registered by the
 819 shallower trap (Fig. 4). Therefore, the growth phase of the *E. huxleyi* bloom probably
 820 took place between October and December 2001, a period characterised by very low SSTs
 821 (0.1-0.9 °C). It was before development of any significant stratification in the upper water
 822 column (Fig. 2b and 4a). These observations indicate that the very cold temperatures (near
 823 0°C) and strong mixing of the water column in the Antarctic waters during spring are not
 824 an impediment for the development of an *E. huxleyi* bloom. [The very low *C. leptoporus*](#)
 825 [and *Gephyrocapsa* spp. fluxes throughout the annual cycle suggest that the environmental](#)
 826 [conditions of the AZ-S must represent an ecological limit of these species. Peak fluxes of](#)



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830 *C. leptoporus* and *Gephyrocapsa* spp at both sediment traps coincide with those of *E.*
831 *huxleyi* indicating that the summer solar irradiance increase is the main factor stimulating
832 coccolithophore growth irrespectively of the species. ✓

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833 The onset of seasonal increase in coccolithophore arrivals in the traps occurred at
834 the same time as that of diatoms, suggesting a rapid response of both phytoplankton
835 groups to enhanced light levels. Although both coccolith and diatom fluxes exhibited a
836 pronounced and nearly parallel increase throughout December (Fig. 5), coccolith fluxes
837 peaked one week later than those of diatoms. A similar succession was observed in late
838 summer, when coccoliths displayed a secondary flux maximum, one sampling interval
839 later (8 days) than that of diatoms (Fig. 5). These observations agree with the bloom-
840 dynamics scheme proposed by Barber and Hiscock (2006) (the so-called coexistence
841 theory), in that neither phytoplankton group seems to outcompete the other during the
842 development of the bloom. Interestingly, diatoms seem to decline earlier than
843 coccolithophores, a feature often (but not always) observed in other parts of the world
844 ocean (e.g. Margalef, 1978; Holligan et al., 1983; Lochte et al., 1993; Sieracki et al., 1993;
845 Thunell et al., 1996; Balch, 2004). Indeed, a recent study of the phenological
846 characteristics of coccolithophore blooms by Hopkins et al. (2015) concluded that they
847 often follow those of diatoms in many regions, the sequencing driven by increasing
848 stabilization and/or nutrient depletion (mainly silicate and/or iron, and possibly also
849 favoured by associated increase of carbonate saturation; Merico et al, 2004) of the surface
850 layer. The slightly different seasonal pattern observed at both sampling depths (Fig. 5) is
851 mainly attributed to the fact that the area of the ocean from which the particles have been
852 produced increases with depth (Siegel and Deuser, 1997).

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853 Lack of nutrient and mixed-layer-depth measurements during the sediment trap
854 deployment precludes us from establishing robust links between changes in physical and
855 chemical parameters in the upper water column and the observed phytoplankton
856 succession. Nonetheless, some shipboard observations of mixed-layer properties from
857 years previous to the sediment trap deployment (Fig. 2; Trull et al., 2001b) can provide
858 some insight about the mechanisms driving the phytoplankton succession. Macronutrient
859 measurements indicate that, although considerable nutrient draw-down often occurs by
860 mid-summer, the AZ-S waters never reach potentially limiting concentrations (i.e. below
861 10 μM) of silicate, nitrate or phosphate (Fig. 2a; Trull et al., 2001b). Thus, macronutrient
862 limitation was not a likely driver of the observed phytoplankton succession at the 61°S
863 site traps. Iron levels in the AZ-S, on the other hand, are low year-round (0.1-0.2 nM;

867 Boyd et al., 2000b; Sohrin et al., 2000) and exhibit clear seasonality in the AZ (Tagliabue
868 et al., 2014). So, iron availability does represent a potential driver for the observed
869 phytoplankton succession. Indeed, laboratory experiments have shown that *E. huxleyi* has
870 lower minimum iron requirements for growth than oceanic diatoms (Brand et al., 1983;
871 Muggli and Harrison, 1997). This physiology likely provides an ecological advantage
872 over diatoms in the later stages of the spring-summer bloom, when most iron has been
873 stripped from the mixed layer.

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874 In regard to the mechanism underlying the second diatom-coccolith succession
875 observed at both depths in February (Fig. 5), it is possible that a vertical mixing event –
876 as frequently reported in the AZ (e.g. Brzezinski et al., 2001) – supplied waters rich in
877 iron and macronutrients to the euphotic zone, resetting the phytoplankton succession.
878 Alternatively, the part of the *E. huxleyi* populations accumulated at or just above the
879 nutricline may have increased using the iron moved by diapycnal diffusion through the
880 pycnocline (Tagliabue et al., 2014). Their deposition in February could have been
881 triggered by a drop of the light levels (Fig. 4). This second hypothesis is also consistent
882 with the following observations: (1) the presence of a sub-surface chlorophyll-*a*
883 maximum in the study region during spring and summer (Parslow et al., 2001; Trull et
884 al., 2001b); (2) reports of high *E. huxleyi* cell accumulations associated with the nutricline
885 in other settings of the world ocean (Beaufort et al., 2008; Henderiks et al., 2012) and (3)
886 peak annual sedimentation in late February of the diatom *Thalassiothrix antarctica*
887 (Rigual-Hernández et al., 2015a), a typical component of the “shade flora” (Kemp et al.,
888 2000; Quéguiner, 2013). Further sampling and taxonomic analysis of the vertical
889 distributions of phytoplankton in the AZ-S south of Australia are required to assess these
890 hypotheses.

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891 892 **4.3 Seasonal variability in coccolith calcification**

893 Two main factors have been proposed as driving seasonal changes in coccolith
894 weights of *E. huxleyi*: a seasonal shift in the dominant morphotypes/ecotypes — each
895 with a different degree of calcification (Poulton et al., 2011) — and the physiological
896 response of a given morphotype to the seasonal variation of environmental parameters
897 (e.g. Smith et al., 2012; Meier et al., 2014). SEM analysis of the sediment trap samples
898 revealed that only morphotype B/C, *sensu* Young et al. (2003), thrives in the AZ-S waters
899 south of Tasmania. That is consistent with a report by Cubillos et al. (2007) of dominance
900 of B/C south of 50°S. Therefore, a seasonal shift in the dominant morphotype can be ruled

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905 out in respect to changing coccolith weight. The observed decrease in coccolith weight
906 could have been caused by a change in coccolith calcification or reduction in coccolith
907 dimensions. Young and Ziveri (2000) showed that coccolith weight is approximately
908 linearly correlated to the cube of coccolith length. Applying that, the decrease in length
909 by 7.5% (a reduction to 92.5%) observed from the pre-bloom to the summer bloom in the
910 2000 m traps (i.e. difference in minimum coccolith lengths in cups 5 and 8) corresponds
911 to a coccolith weight loss of 21% ($0.925^3 \approx 0.79$). That is similar to the observed weight
912 reduction in the 2000 m trap between the pre-bloom and summer bloom coccolith
913 assemblages (16.2 - 27.6%, respectively Fig. 6). When the linear correlation between
914 coccolith length and weight proposed by Young and Ziveri (2000) is also applied to the
915 3700 m trap coccoliths, the predicted reduction of coccolith weight between the pre-
916 bloom and bloom assemblages is 12%. That is again very similar to the reduction in
917 coccolith weight observed in the *E. huxleyi* coccoliths intercepted by the 3700 trap (10%).
918 It is strongly suggested that the seasonal changes in coccolith weight at the 61°S site were
919 mainly driven by changes in coccolith length and were not due to significant changes in
920 their degrees of calcification.

921 Laboratory, mesocosm and field studies have shown that multiple environmental
922 factors including irradiance, temperature, [salinity](#), macronutrient concentrations and iron
923 availability affect coccolith formation by *E. huxleyi* cells (e.g. Paasche, 2002; Zondervan,
924 2007; Langer and Benner, 2009; Feng et al., 2017). Since calcification in *E. huxleyi* is a
925 light-dependent process (Paasche, 1999, 2002), the observed decrease in coccolith weight
926 during summer in both traps was somewhat unexpected. Some field experiments have
927 shown that calcification in coccolithophores can occur at low light levels, or even in the
928 absence of light (e.g. van der Wal et al., 1994). However, it is often reduced compared to
929 that at higher irradiance levels (Zondervan, 2007).

930 [In terms of temperature effects, Saruwatari et al. \(2016\) described a decrease in](#)
931 [coccolith size with increasing temperature cultivating *E. huxleyi* strains \(morphotype B/C,](#)
932 [strains MR57N and MR70N\) from the Bering and Chukchi Seas. However, comparison](#)
933 [of our results with those of Saruwatari et al. \(2016\) should be done with great caution due](#)
934 [to two reasons. Firstly, the *E. huxleyi* coccolithophores living in the Arctic seas most](#)
935 [likely correspond to a different ecotype than those dwelling in the AZ waters, and](#)
936 [therefore they may potentially exhibit different physiological responses to water](#)
937 [temperature changes. Secondly, the SST range in our study site was remarkably lower \(0](#)
938 [- 3°C\) than that used by Saruwatari et al. \(2016\) in their cultures \(5 - 20°C\). These](#)

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940 limitations make drawing inferences from Saruwatari et al. (2016) difficult. Feng et al.
 941 (2017), on the other hand, showed that optimal temperature for calcification of *E. huxleyi*
 942 cells retrieved in the Southern Ocean (morphotype A, strain NIWA1108) was ~20°C,
 943 while temperatures below 10°C resulted in a dramatic reduction of calcification rates and
 944 severe malformations of coccoliths, such as incomplete distal shield elements. Although
 945 *E. huxleyi* morphotype B/C found at the 61°S site likely represents an ecotype more
 946 tolerant to low temperatures than morphotype A (Cubillos et al., 2007; Cook et al., 2013),
 947 the frequent variations in the structure of the coccoliths (e.g. incomplete distal shield
 948 elements; Plate 1) captured by the traps suggest some degree of low-temperature stress.
 949 Despite the important role of temperature in coccolithophore growth (Paasche, 2002),
 950 enhanced summer SSTs may lead to an increase in coccolith weight, a response opposite
 951 to that observed at both traps. Therefore, it is unlikely that seasonal SST variations at the
 952 61°S are behind the observed variability in coccolithophore weight.

953 Bollmann and Herrle (2007) identified a close relationship between changes in
 954 SSS (gradient from 33 to 38) and the length of *E. huxleyi* coccoliths using a global
 955 compilation of core top and plankton samples. However, based on the almost negligible
 956 annual variability in SSS (values ranging between 33.7 to 33.9 psu) in the study region,
 957 salinity most likely did not play a significant role on the observed seasonal variability in
 958 coccolith morphology observed in our traps.

959 In regard to the possible impact of macronutrient concentrations on coccolith
 960 weight, both nitrate and phosphate are known to have a pronounced effect on coccolith
 961 calcite content and morphology (Zondervan, 2007). However, as mentioned previously,
 962 none of these macronutrients reach limiting concentrations throughout the annual cycle
 963 in the AZ-S (Fig. 2; Trull et al., 2001b). and, therefore, their influence in the calcification
 964 of coccolithophores is likely to be low or negligible.

965 On the other hand, low iron levels have been reported to have a pronounced
 966 negative effect on CaCO₃ production by *E. huxleyi* cells (Schulz et al., 2004), so it
 967 represents a candidate driver of seasonal changes in coccolith weight. During winter, deep
 968 water mixing re-stocks the mixed layer with iron (Tagliabue et al., 2014). As soon as light
 969 levels become sufficient for photosynthesis in early spring, phytoplankton rapidly
 970 develops under non-limiting concentrations of macro- and micronutrients. These
 971 favourable conditions for coccolithophore growth could explain the heavier and larger
 972 coccoliths registered in early December (Fig. 6). As the phytoplankton bloom develops,

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980 the dissolved iron stock is rapidly depleted in the photic zone possibly resulting in a size
981 and weight reduction of coccoliths of the already substantial *E. huxleyi* populations. From
982 late summer throughout autumn, some recycling of iron in the upper water column by
983 increasing summer populations of zooplankton feeding on the bloom (Tagliabue et al.,
984 2014), coupled with increasing light levels and the continued shallowing of the mixed
985 layer, would allow coccolithophores to produce again longer and heavier coccoliths (Fig.
986 6).

987 Iron-limitation, therefore, represents the most likely environmental driving factor
988 for the seasonal variability in coccolith weight and length of *E. huxleyi* assemblages at
989 the 61°S site. However, we note again that the absence of *in situ* measurements of
990 chemical and physical parameters of the water column, means that control of coccolith
991 weight by varying iron availability in the AZ-S remains an hypothesis needing validation
992 by future studies.

993

994 4.4 Effects of calcite dissolution on the sinking coccolith assemblages

995 The similar average annual coccolith weight registered at both traps indicates that
996 negligible coccolith dissolution occurs at meso- and bathypelagic depths in the AZ-S
997 south of Australia. That is despite the fact that coccolith sinking assemblages captured by
998 the deeper trap were exposed to potentially intense dissolution after crossing the CSH
999 (located at 3000 m in the study region; Fig. 2). The similar coccolith values observed at
1000 both depths can be attributed to the formation of algal and faecal aggregates in the mixed
1001 layer that include fine mineral particles (Passow and De La Rocha, 2006) and provide
1002 protection against dissolution. They also facilitate rapid transport of the coccoliths down
1003 through the water column. The aggregate-formation hypothesis is supported by the
1004 findings of Closset et al. (2015) who estimated that sinking rates at the 61°S site were, at
1005 least 213 m d⁻¹ during the productive period, a value consistent with the sinking rates of
1006 algal and/or faecal aggregates (Turner, 2002, 2015).

1007 Despite not finding increased dissolution with water depth between 2000 and 3700
1008 m, it is possible that coccoliths experienced some carbonate dissolution before reaching
1009 the traps. Milliman et al. (1999) suggested that the same biological processes that
1010 facilitate aggregate formation and flocculation, such as ingestion, digestion and egestion
1011 by grazers, may be responsible for significant carbonate dissolution at epipelagic depths
1012 (i.e. depths shallower than 800-1000 m. Indeed, the negligible amounts of coccospheres
1013 found in both traps, together with the high sinking velocities, suggest that grazing could

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1017 have been an important influence on export. That is supported by findings of Ebersbach
1018 et al. (2011) in the PFZ north of our study location. They documented that an important
1019 fraction of the particles sinks from the mixed layer as faecal aggregates. On the other
1020 hand, the small spherules often observed on the coccoliths captured by the traps suggest
1021 some degree of coccolith dissolution followed by remineralisation. We speculate that
1022 some of the coccoliths captured by the traps could have experience partial dissolution in
1023 the upper water column leading to the exposure of their organic coccolith scaffold (Gal
1024 et al., 2016; Lee et al., 2016), to the environment. It is possible that salts dissolved in the
1025 water column subsequently precipitated over this scaffold structures resulting in the
1026 formation of the recrystallised structures observed in some coccoliths (Plate I, e-g).
1027 However, the available data are insufficient to evaluate the impact of carbonate
1028 dissolution in the upper water column and processes leading to secondary recrystallisation
1029 in the coccoliths.

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1031 4.5 Calcium carbonate content of *Emiliana huxleyi* coccoliths

1032 A broad range of calcite contents for *E. huxleyi* coccoliths (1.4 - 7.0 pg) has been
1033 proposed in the literature (e.g. Young and Ziveri, 2000; Beaufort, 2005; Holligan et al.,
1034 2010; Poulton et al., 2011). The differences in these estimates are most likely due to
1035 variability in the amount of coccolith calcite between morphotypes and to the varied
1036 methodological biases associated with the three main approaches for estimating coccolith
1037 mass: morphometrics, regression and birefringence. Since *E. huxleyi* morphotype B/C is
1038 considered to be geographically restricted to the Southern Ocean (Cubillos et al., 2007;
1039 Cook et al., 2013) we limit the comparison of our results to studies conducted only in the
1040 Southern Ocean reporting this morphotype.

Eliminado: is more weakly calcified than other morphotypes (Young and Ziveri, 2000) and

1041 Average annual coccolith quotas at both trap depths at the 61°S site (2.11 ± 0.96
1042 and 2.13 ± 0.91 pg per coccolith at 2000 m and 3700 m, respectively) are almost identical
1043 to that estimated by Holligan et al. (2010) (2.20 ± 0.60 pg ; morphotype B/C) in the Scotia
1044 Sea using a regression line between the number of coccoliths against PIC. Moreover, our
1045 estimates are slightly higher, but with a considerable overlap in the ranges of coccolith
1046 weight, than those estimated by Poulton et al (2011) for the *E. huxleyi* morphotype B/C
1047 populations found in Patagonian shelf waters (1.40 ± 0.6 pg). The greater standard
1048 deviation of our data is most likely due to the time periods compared. While the average
1049 coccolith weight estimated for our traps reflects an integration of the annual variability in
1050 coccolith weight, the shipboard observations by Poulton et al. (2011) provide a snapshot

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1055 of the summer coccolithophore populations, that likely exhibit lower coccolith size and,
1056 thus, variability.

1057 Because our coccolith weight estimates are similar to those of Poulton et al. (2011)
1058 and Holligan et al. (2010), we can estimate the fractional contribution of coccolithophores
1059 to total carbonate production in the AZ-S south of Australia. Coccolithophores account
1060 for approximately 2-5% of the annual deep-ocean CaCO₃ fluxes at mesopelagic depths at
1061 the 61°S site. The contribution of coccolithophores to the annual CaCO₃ budget in the
1062 AZ-S south of Australia is similar to the estimate by Salter et al. (2014) for the
1063 macronutrient-rich, but iron deficient M6 site in the Indian sector of the AZ (12%) and
1064 remarkably lower than an estimate for the iron-fertilised station A3 over the central
1065 Kerguelen Plateau (85%; Rembauville et al., 2016). Due to the different methodologies
1066 for estimating coccolithophore contributions to carbonate production, comparison of our
1067 results with these other studies should be treated with caution. While only whole
1068 coccoliths were counted for our calculation, therefore providing a conservative estimate,
1069 Salter et al. (2014) and Rembauville et al. (2016) estimated the weight of the < 20 µm
1070 fraction using inductively coupled plasma-atomic emission spectrometry. That approach
1071 often results in overestimates of the coccolith contribution to bulk carbonate content.
1072 There can be non-negligible contributions of non-coccolith fragments to the fine fraction
1073 (Giraudeau and Beaufort, 2007). Despite the biases associated with both methodologies,
1074 the general trend appears clear: the fractional contributions of coccolithophores to bulk
1075 carbonate export are lower in the iron-limited waters of the AZ compared to those in
1076 naturally iron-fertilised settings of the Southern Ocean. These findings underscore the
1077 secondary role of this phytoplankton group in the biological carbon pumps (both the in
1078 organic carbon and carbonate counter pumps) south of the PF where non-calcifying
1079 phytoplankton - mainly diatoms and *Phaeocystis* - largely control the biologically-
1080 mediated CO₂ exchange between the ocean and the atmosphere.

1081

1082 Conclusions

1083 Analysis of the materials captured by two sediment traps deployed at the 61°S site
1084 allowed for the characterization and quantification of coccolith assemblages in Australian
1085 sector of the Antarctic Zone. The data presented here provide a baseline of the state of
1086 coccolithophore populations in this region against which future changes can be assessed.
1087 More specifically, our study has shown the following:

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- 1097 • Coccolithophores were a consistent member of the phytoplankton communities of the
1098 Antarctic Zone south of Australia in year 2001. Coccolithophore assemblages in this
1099 region are monospecific being composed almost entirely of *Emiliana huxleyi*
1100 morphotype B/C. This observation supports the hypothesis that the physiological
1101 differences in light-harvesting pigments of morphotype B/C (or *E. huxleyi* var.
1102 *aurorae*), compared to other Southern Ocean *E. huxleyi* varieties (Cook et al., 2011),
1103 may represent an ecological advantage in the cold, low-light and iron-limited
1104 environment of the Antarctic Zone.
- 1105 • The onset of the coccolithophore productive period took place at the same time as that
1106 of diatoms, indicating that neither phytoplankton group outcompetes the other during
1107 the development of the bloom. We speculate that the diatom-coccolithophore
1108 succession observed during the peak phase of the productive period could result from
1109 the lower minimum iron requirements for growth of *E. huxleyi*, a feature that may
1110 confer a competitive advantage over diatoms.
- 1111 • A decrease in coccolith weight and size during the summer months was observed at
1112 both sediment trap depths. After assessing the potential influence of several
1113 environmental parameters, increasing iron limitation seems to be the most likely
1114 candidate to drive this change. This hypothesis, however, will need to be validated in
1115 future field and laboratory culture experiments with morphotype B/C.
- 1116 • The similar weight of *E. huxleyi* coccolith assemblages captured by the 2000 and 3700
1117 m sediment traps indicates that negligible coccolith dissolution occurs during transit
1118 through meso- and bathypelagic depths in the study region. This is most likely due to
1119 a rapid transport of the coccoliths in algal and/or faecal aggregates.
- 1120 • Coccolith weight values calculated for both sediment trap records using a
1121 birefringence-based approach were similar to previous estimates of *E. huxleyi*
1122 morphotype B/C in other Southern Ocean settings using regression and morphometric
1123 methods (Holligan et al., 2010; Poulton et al., 2011, respectively).
- 1124 • Coccolithophore fluxes at the 61°S site account for only 2-5% of the annual deep-
1125 ocean CaCO₃ fluxes, suggesting that heterotrophic calcifiers must represent the main
1126 biogenic carbonate producer in the AZ-S south of Australia.

1127 **Acknowledgments**

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