

Interactive comment on “Seasonal development of iron limitation in the sub-Antarctic zone” by Thomas J. Ryan-Keogh et al.

Anonymous Referee #1

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This manuscript presents an attempt to evaluate the seasonal cycle of iron stress in the sub-Antarctic zone using three bottle-scale iron enrichment experiments conducted in December–February. The novel aspect of the study is the reoccupation of the experimental site over 2 months. Whilst these observations cannot, by some margin, be used to confidently state overarching changes in SAZ iron stress in these months, they are still valuable to the scientific community and worthy of publication in Biogeosciences. I however have a number of comments that should be addressed prior to publication. In particular I think the authors should more carefully/critically evaluate how far their experiments can actually be used to evaluate the seasonal development of Fe limitation in the sub-Antarctic zone without an analysis of supporting depth-resolved Fe, mixed layer depths, and PAR data. Upon reflection of the former, some rephras-

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ing of the manuscript is required. Some additional important method details are also lacking.

The paper is in general well written and referenced and the figures and tables are clear and complete. My comments below are listed in order through the manuscript, not by importance.

Specific comments:

Line 18: Variability in iron supply also includes dust (not mentioned)

Line 21–22: ‘incubation experiments were used to determine the importance of (iron) supply mechanisms’. Can they really be used for this? All they actually indicate is the biological response to incubation, not sources? A thorough analysis of Fe supply and demand, in conjunction with the bioassays, would be needed to do this (only 3 Fe values are reported).

Line 26–27: The results presented do not support the claim of progressive Fe depletion—phytoplankton appear to respond more to the Fe later in summer, but the in-situ Fe concentrations stay the same.

Line 72–73: Whilst excess nitrate+nitrite excretion under Fe stress may play a role, I think most would argue that high rates of resupply (relative to inefficient biological removal) overwhelmingly control the elevated residual nitrate in the Southern Ocean.

Line 97: ‘were’ change to ‘that were’?

Line 99: Do the results actually indicate a change the photosynthetic efficiency? As the authors say themselves, most of the Fv/Fm change could well be due to pigment changes that have little to do with PSII efficiency.

Line 99: Change ‘biomass’ to ‘chlorophyll-a biomass’?

Line 113: Table ‘X’?

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Line 115: CTD abbreviation defined?

Line 122: Why was water 'allowed to settle' in Go-Flo samplers? To increase the overall phytoplankton concentrations in the incubation bottles?

Section 2.2: - More details on the incubation experiment setup needed: What was the actual incubator? A culture cabinet? Something custom built? Please give details.
- How does the PAR values supplied in the incubator differ from in-situ values? For this the authors will need to calculate an average ML PAR using their observations of ML, CTD PAR and ship-instrument PAR. Although this might be a pain, it might really help to pick apart the difference in growth environments experienced by the community just prior to incubation, and thereby help to interpret their response to the altered conditions.

Lines 130-131: Which experiments were in duplicates and which in triplicates? Figure 2 states $n=3$ or $n=5$, so I do not understand this. Please clearly indicate number of biological replicates (number of bottles with the same treatment) and technical replicates (i.e. FRR/chl measurements made from the same bottle).

Line 133-134: Can this approximate time of sub-sampling be included in Table 1? This might help to interpret the results (for instance, if daytime, the Fv/Fm increase in Exp 1 due to differences in PSII damage/down regulation between days).

Line 144: Exactly how were the dFe samples filtered (method, on-ship/land)?

Line 154: Was the instrument a FastTrackall?

Line 168: Was the FastPro software used or was custom code used? If the latter please give details.

Section 2.8: What was the test for means comparison between treatments? T-test?

Lines 303-304: 'The rapid increase in Fv/Fm in both treatments at 24 h is likely due to bottle effects i.e. a change in light environment.' - This experiment needs discussing

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in more detail as the community response is clearly compatible with relief of resource limitation of larger cells. How did the light change between in-situ conditions and that in the incubator? What was the integrated PAR over the previous hours prior to the Fv/Fm measurement being made (if not at night time)? Perhaps different levels of PSII damage/down-regulation could be an explanation. Could the observed community shift not contribute to the Fv/Fm increases, i.e. do larger cells not typically have higher Fv/Fm (Sugget et al. 2009)? Should any chance of Fe contamination in the control bottles (e.g. during the 50L carboy) be acknowledged, as this would also be consistent with the observed responses?

References to seasonal Fe supply (e.g. lines 315-318, 341 onwards): Reduced Fe concentrations through the growing season are not actually observed. Furthermore, Fv/Fm values are low at both the beginning and end of the growing season. The only data the authors have to go on is the more pronounced phytoplankton responses to Fe relative to controls later in the growing season. Please rephrase these sections to more clearly indicate specifically what your bioassay results can actually say about seasonal changes in Fe supply to mixed layer waters.

Lines 311-312: 'large diatoms would require an increased silicate concentration, which is a limiting macronutrient in this region'. Is silicate a limiting nutrient at your site? Concentrations over 1 μ M were measured and chlorophyll-a and diatoms were enhanced in all +Fe treatments without added silicate.

Line 369-373: The authors state mean chlorophyll over the euphotic zone was higher than that over that of the mixed layer and then interpret this as a result of insufficient iron within the mixed layer. Whilst this is a possibility, could accumulation of lower light acclimated (higher chlorophyll/cell) phytoplankton below the mixed layer not equally play a role?

Lines 374-378: The mechanism the authors describe to explain the lack of Fe stress despite low Fe concentrations is not clear. The authors state that the 'limiting' nutrient

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'would be expected to be severely depleted through biological uptake regardless of resupply'. But the authors state that they observe no iron limitation early in the season, which is at odds with this explanation. If Fe was not limiting it should either accumulate in the dissolved phase, be scavenged, or taken up by the phytoplankton and stored even though it is not limiting. A re-phrase here might be necessary.

Lines 383-384: 'The short transient periods of increased wind stress thus appear to provide temporal relief from Fe stress'. Where is the data (wind, mixed layer depths, dFe concentrations, Fe stress status) to support this?

Lines 389: Increased nitrate concentrations throughout the growing season: do the authors need to invoke iron limitation as reducing the availability of photosynthetic reductant for nitrate reduction? The increase in nitrate concentration is large $\sim 8\mu\text{M}$: could physical process, such as a greater contribution of more recently upwelled water, be used to explain this? Or is the temperature-derived ML not capturing enhanced surface stratification later in the season that restricts downward mixing of more nitrate-depleted surface-most waters down to the incubation water collection depth? To test this the authors could calculate the buoyancy frequency in addition to the mixed layer depth.

Line 406-407: In the high latitude of North Atlantic and potentially North Pacific the cryosphere is important to seasonal dynamics? (ice/ground melting leading to enhanced stratification etc.)

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