Dear Emilio Marañón,

Please see our detailed responses to the reviewer comments, including detailed information on which parts of the manuscripts were changed, and how. Responses by us are in blue and cursive.

Reviewer 1

## General comment

The manuscript presents multi-factorial experiments to examine the potential impacts of ocean warming and Fe availability under future climate change scenarios in two different regions of the Southern Ocean. The manuscript demonstrates that warming does have an impact on phytoplankton growth and community structure, but that under many cases these impacts are less than the effect of Fe under current temperatures. The results raise discussion points for how additional factors, such as Manganese and light, should be studied in combination to provide a better understanding in future studies.

Overall the manuscript is very well put together, with my only main concern being that statistical tests between treatments focused primarily on bulk concentrations and not on the differences in rates (i.e. chlorophyll derived growth rates, nutrient drawdown rates etc).

*We looked at the net growth rates in combination to the bulk comparisons we made: i) for phytoplankton group specific net growth rates, please see our response to Reviewer's specific comment on this topic (comment on line 522), (ii) for the other variables (chlorophyll, dissolved and particulate metal concentrations) we had limited time points, preventing proper rate calculations, and (iii) statistical analysis based on nutrient drawdown rates rather than final concentrations gave the same results (with the exception that temperature was on the brink of being a significant factor affecting Si in bioassay A2, while it was a significant factor based on bulk concentrations, i.e., p= 0.08 for rates vs 0.02 for bulk concentrations). We thus did not add the nutrient drawdown rates to the manuscript, but will add total nutrient drawdowns to the Supplements.*

This study was also quite unique in that it reports not only changes in dissolved trace metal concentrations in the treatments but also changes in particulate trace metal concentrations. However, I could not find any reporting on contributions of lithogenic fractions from the initial starting water which may impact some of the particulate results.

*The initial lithogenic fraction of particulate trace metals was higher in Amundsen Sea compared to the Weddell Sea* (e.g., ca.60~~-80 – 90~~ % vs ca. 52 ~~30~~% for pFe). Please note that the lithogenic concentration did not change during the experiments, i.e. background concentration remains the same, and as such does not affect comparison between the treatments. Please also see our replies to later comment (on line 434) regarding this topic.*

**Please note the slight change in lithogenic percentage in response to Reviewer 1's previous comments, following our final analysis.*

There are some more general comments about areas that require some greater clarity or further expansion in the PDF.

Specific comments:

-line 47. I would raise the idea that you should be comparing statistically the growth rates between treatments, rather than just the bulk concentrations. I think this could definitely strengthen your conclusions and overcome any different initial starting points in bulk concentrations that may be impacting the results.

*See our first reply on the same topic.*

-line 70. Sallee et al. 2021 report trends of increasing stratification at depth. Would this not impact supply of Fe from below the Ferricline?

*We agree that an increasing stratification at depth could also impact the supply of Fe from below the Ferricline, adding uncertainty to predictions of future iron concentrations that are nevertheless expected to increase. We will change lines 65 onwards (changes from original manuscript are underlined for clarity): "Trace metal supply in the Southern Ocean follows a strong seasonal cycle where in winter Fe is replenished via deep water-mixing (Tagliabue et al., 2014) or sediment resuspension in coastal areas (Boyd et al., 2012), to be quickly depleted again by phytoplankton uptake in the next season. Predicted increases in stratification may weaken dFe supply to surface waters from below (Sallée et al. 2011), however, this is still uncertain as increased stratification might not have a strong effect or might even increase turbulent nutrient fluxes associated with breaking internal waves (van Haren et al., 2020). Additionally, increased stratification effects may be counteracted by a deepening of mixed layer depths (Sallée et al., 2021) and changes in gyre-scale circulations (Misumi et al., 2013). In general, Fe limitation for Antarctic phytoplankton is predicted to be at least partially relieved in the future (Bazzani et al., 2023) …"*

-line 73. There are also expected changes in atmospheric supply from areas such as Patagonian dust (Li et al., 2008; Portner et al., 2022).
Note that future climate conditions could decrease dissolution rates (Demasy et al., 2024, Frontiers in Marine Science).
*While the supply of iron from Patagonian dust to the Southern Ocean is predicted to increase, this may not necessarily enhance dFe concentrations in our study regions. This is because future climate conditions are expected to decrease the dissolution rates of particulate iron (pFe), potentially counteracting the increased dust input (Demasy et al., 2024). It is important to note that both the changes in dust supply and dissolution rates are uncertain, and the overall impact on dFe concentrations in the Southern Ocean remains unclear. Since the input of dFe from Patagonian dust is very uncertain, and dFe concentrations added via dust would be comparably small (Lancelot et al., 2009, Biogeosciences, Vol 6 (12)), we decided to not include this into the Introduction.*

It may also be worth mentioning what impact you expect temperature may have on bacteria, i.e., siderophore production?
*Temperature does not seem to have a direct effect on siderophore production in the Southern Ocean (Sinha et al., 2019). At the same time, ocean acidification may lead to pH levels where siderophore production is lowered, already at a pH of 7.5, siderophore production was lower compared to production levels at a pH of 8.5 (Sinha et al. 2019). This may be (partly) countered by enhanced siderophore concentrations due to increased growth rates of the siderophore-producing bacteria with warming (Sinha et al. 2019). We suggest adding the following to the Discussion (section 4.3): "Furthermore, the availability of dFe is likely changing due changes in sources (see introduction) but is also influenced by siderophore production (reviewed by Gledhill & Buck 2012, Frontiers in Microbiology, Vol 3) but warming of the Southern Ocean does not seem to have a direct effect (Sinha et al., 2019, Journal of Basic Microbiology, Vol 59 (4)). Warming likely increases the growth rates of siderophore producing bacteria (Sinha et. al. 2019), but this may be countered by reduced siderophore production due to ocean acidification (Sinha et al. 2019)."*

-line 16. Were large grazers removed before filling the containers? If not, what would you expect their impact to be on the results.
*Water was not filtered before filling the cubitainers (to avoid contamination risk), thus large grazers were not removed. We will add the following to the Discussion section 4.4: "Since the seawater was not filtered before distribution to the cubitainers to reduce contamination risk, there is a chance (although small, Voronina et al., 1994, Polar Biology, Vol 14) that large grazers were introduced to the incubations. We did not specifically sample for large grazers but did not notice any on the filters for Chl a and POC. Large grazers can be expected to graze on larger phytoplankton (Hansen et al. 1994, Limnology and oceanography, Vol 39 (2)), thereby reducing phytoplankton net growth. This would be most noticeable for the F and TF treatments, given the positive response of larger phytoplankton to Fe addition. Our results would then be underestimating the effect of Fe enrichment. Moreover, grazing*

*would likely enhance with temperature (e.g. Lewandowska and Sommer, 2010, Marine Ecology Progress Series, Vol 405; Karakuş et al., 2022, Journal of Geophysical Research: Biogeosciences, Vol 127 (10)), further reducing (and underestimating) net growth rates of larger phytoplankton specifically in the TF treatment. "*
*Moreover, if the (typically low abundance) large grazers were present it likely would have resulted in large variation between the replicates.*

-line 182. I think you should add a statement here that these were in custom built deck incubators as I didn't first understand this. Then the reader can go to the SI for all the extra details
*Thank you for pointing this out. We will add the following as a first sentence to section 2.2 Bioassay setup: "Incubations were performed in custom built deck incubators (see Supplement Bioassay Setup for more information)."*

-line 298. Is there any way to ascertain sigmaPSII from this method? As this information could help to provide more insight into the interacting effects of Fe and light from your experiments. If it is not a standard output then do not worry.
*Unfortunately, it is not possible to ascertain sigmaPSII from the instrument we used here.*

-line 301. Schuback et al. 2021 recommend low light adaptation to remove any impacts of quenching. Did you subsample for photopshysiology at night or during the day?
If you subsampled during the day, what impact may you think this dark adaptation would have on the final Fv/Fm values you recorded?
*Thank you for the reference. The incubations were usually sampled during the day, but we also had light during the night, which could potentially have led to slight underestimation of Fv/Fm, more so for the Weddel Sea as light intensities for Amundsen Sea Bioassays were already low. However, earlier tests with different phytoplankton including polar phytoplankton did not show an effect of different dark-adaptation times (mentioned in original manuscript, line 303).*

-line 303. What impact do you think this UP water could have on the cells? Are there any risks of cell rupture due to contact with low saline UP water? Would rinsing the cuvette with the actual incubation water have not reduced this risk?
*UP water was removed from the cuvette by shaking and placing the cuvette upside down on lint-free paper towels to remove last droplets. Technical replicates were measured for each sample, and the cuvette was not rinsed between technical replicates. We tested in response to your comment for potential effects of remaining UP water droplets and found no significant difference in $F_v/F_m$ values between technical replicates (non-parametric Kruskal-Wallis ANOVA, p = 0.95). We will add this to the Material and Methods section: "The cuvette was rinsed with ultra-pure (UP) water between samples, which was removed by shaking the cuvette and placing it upside down on lint-free paper towels to remove any remaining droplets (testing technical replicates did not show a significant effect of UP rinsing, non-parametric Kruskal-Wallis ANOVA, p = 0.95)."*

-line 304. What was the % range of your blanks relative to Fm?
*We used 0.2 µm filtered seawater sample for blanking (Cullen and Davis 2003, L&O Bulletin, Vol 12 (2)). The % range for the Weddel Sea bioassays was on average 13%, while for the Amundsen Sea bioassays it 46%. The highest % were measured in bioassay A2 where chlorophyll concentrations were lowest, resulting in having to adjust the photo-multiplier to higher settings with higher blanks. Overall, the Fv/Fm values did not differ majorly when blanks were higher compared to lower.*

-line 410. Did W1 have a larger initial diatom abundance compared to the other experiments?
*Yes, W1 did indeed have a larger initial diatom abundance compared to the other Bioassays, see line 497 in the original manuscript.*

-line 434. This is thus assuming that all of the particulates is biogenic in nature. Did you calculate the contribution of lithogenic particles in your incubation water?
It is also important to consider how much of this is also authigenic as well.
*We will add the following text to the Material and Methods section 2.5, following line 262: "The lithogenic fraction and concentration of pFe and other particulate metals discussed was determined by assessing the ratio between the particulate metal of interest and particulate aluminium (pAl), assuming all pAl originates from crustal material using the approach described in more detail in van Manen et al (2022). For example, we are using the observed pFe/pAl ratio in the samples and the known crustal ratio of 0.21 mol mol$^{-1}$ (Taylor and McLennan, 1985, The continental crust) to calculate the lithogenic pFe fraction and concentration~~, see supplemental data (Table S10) for more details~~."*
*And: "The EDTA oxalic acid wash used on particulate samples prior to filtration should effectively remove surface-bound metals, also minimizing the authigenic Fe fraction" following line 246.*

*In the Results (following line 435), we will add that lithogenic particles provided a consistent background that did not affect observed changes between the treatments, and we will add the respective data to the Supplements.*

-line 476. Did you attempt to calculate any Chl-a or POC derived growth rates from the experiments? Significant differences between treatments may become clearer, especially when you log transform the rates. Just a suggestion to help tease apart whether some of the small not significant changes you see here may actually be significant between treatments.
*Because we only have sampled at the start and the end of the bioassays for both Chl a and POC, calculating derived growth rates is not appropriate and would not help teasing those effects apart.*

-line 522. I would also add the same suggestion here to calculate the growth rates of abundances for your different groups and log transform for statistical comparison.
*The net growth rates for the different phytoplankton populations showed largely comparable results (based on statistics) as the comparisons based on abundances on the last day of the incubations. Still, we agree that addition of these net growth rates is helpful, and we will add them to the Results section (changes from original manuscript are underlined for clarity):*

*Line 526 - 552 of the original manuscript will be changed to: "Phyto 19 increased in abundance and share (Fig. 6a) <u>specifically</u> in the temperature treatments, <u>with net growth rates of 0.40 ± 0.08 d$^{-1}$ and 0.52 ± 0.005 d$^{-1}$ for the T and TF treatments (compared to 0.35 ± 0.11 d-1 and 0.30 ± 0.09 d-1 for C and F treatments, p < 0.04) and final abundances</u> of 2,800 and 3.500 cells mL$^{-1}$ for T and TF (compared to 1,700 and 1.300 mL$^{-1}$ for C and F, p < 0.01). <u>Phyto 3 also showed higher abundance-derived net growth rates with warming (0.33 ± 0.13 and 0.32 ± 0.002 vs 0.26 ± 0.06 d$^{-1}$ for the T, TF and C treatment, respectively), but with abundances being only significantly higher for the TF treatment (776 ± 37 vs 542 ± 107 cells mL$^{-1}$ for TF and C treatments).</u> Phyto 24 was positively impacted by <u>Fe addition, particularly</u> the TF treatment <u>resulted in higher net growth rates and final abundances (i.e., 0.32 ± 0.09 vs 0.15 ± 0.06 d$^{-1}$, and</u> 595 ± 62 vs 361 ± 9 cells mL$^{-1}$ for TF compared to the C treatment; p < 0.05). When converted to cellular carbon based on cell volume using 237 and 196.5 fg C µm$^{-3}$ as conversion factors for Pico- and Nanophytoplankton, respectively (Fig. 6e), the strong positive response of the phytoplankton to the TF treatment was mostly due to this larger-sized Phyto 24 (average diameter of 19 µm, p = 0.01, stat: 0.92) and to smaller extent Phyto 19 (p < 0.01).
Bioassay A2 presented the highest share of picoeukaryotes, especially Phyto 3 (59 % compared to max. 18 % in the other bioassays, Fig. 6b). Only few treatment-specific responses were recorded. Phyto 19 increased somewhat with warming (p = 0.04), <u>and Phyto groups 16 and 17 showed increased net growth rates with dFe addition (0.31 ± 0.22 and 0.23 ± 0.06 vs 0.09 ± 0.16 and 0.31 ± 0.06, 0.30 ± 0.06 and 0.23 ± 0.06 for the F, TF and C treatments of Phyto 16 and 17, respectively, p < 0.02 for both).</u> The phytoplankton populations in W1 were distributed more equally (Fig. 6c), with higher abundances of especially Phyto 16 and 17 for the Fe addition treatments (p < 0.05, most pronounced for TF with average abundances of 3,103 ± 1,290 vs 948 ± 218 cells mL$^{-1}$ and 2,041 ±572*

*vs 1,158 ± 216 cells mL$^{-1}$ for Phyto 16 and 17 in the TF vs C treatments, respectively). Their specific net growth rates were up to 2.2-fold higher for the Fe addition treatments than the control (0.29 ± 0.02, 0.38 ± 0.10 and 0.20 ± 0.02, and 0.16 ± 0.02, 0.21 ± 0.06 and 0.09 ± 0.02 d$^{-1}$ for the F, TF and C treatment of Phyto 16 and 17). When expressed in carbon, Phyto 16 was still a recognisable indicator species (p = 0.03) but at the same time the larger-sized Phyto 21 (average cell diameter of 10 um) and diatoms Phyto 22-24 (13-19 µm) showed clear positive responses to Fe addition (Fig. 6g, p < 0.05 for all). Net growth rates were largely comparable for these phytoplankton groups: 0.23 ± 0.02, 0.19 ± 0.01, 0.17 ± 0.04, 0.20 ± 0.05d$^{-1}$ for Phyto 21-24 in the F treatment (and similar net growth rates in the TF treatment) compared to 0.09 ± 0.07, 0.14 ± 0.03, 0.04 ± 0.04, 0.12 ± 0.02 in the C treatment, respectively (p < 0.03). Bioassay W2 also showed a distinct shift in favour of Phyto 16 and Phyto 17 (away from Phyto 13) with Fe addition, already early in time (Table S8), both for abundances and cellular carbon (Fig. 16d, h, p < 0.01 for all). The F treatment net growth rates of Phyto 16, 17 and Phyto 13 were 0.42 ± 0.02, 0.34 ± 0.03 and 0.21 ± 0.09 d$^{-1}$ (again with similar growth in the TF treatments) compared to 0.20 ± 0.03, 0.17 ± 0.04 and 0.37 ± 0.02 d$^{-1}$ in the C treatment (p < 0.03). Diatoms 23 and 24 also responded positively to Fe addition with ~2-fold higher net growth rates than the control (Fig. 16h, p < 0.01). Phyto 23 net growth rates were 0.37 ± 0.06 and 0.39 ± 0.04 d$^{-1}$ for F and TF compared to 0.19 ± 0.06 d$^{-1}$ for the C treatment (p = 0.004), and Phyto 24 net growth rates were 0.38 ± 0.08 and 0.32 ± 0.05 for F and TF treatments vs 0.22 ± 0.09 for the C treatment."*

*Line 694 to:*
*"GLM analysis revealed that temperature alone was a significant factor for total phytoplankton abundances, however more specifically, only Phyto 3, Phyto 19 and Phyto 22 abundances displayed significant positive responses to temperature alone (T treatment of Amundsen Sea Bioassays).*

*And line 714 to:*
*"Phaeocystis antarctica showed higher net growth rates for Fe-addition treatments in both bioassay W1 and W2, however, the effect was not very apparent and overall, P. antarctica seemed to handle the other treatments consistently well."*

*Moreover, we will add how the phytoplankton abundance-derived net growth rates were calculated in the Material and Methods section: "Phytoplankton net growth rates were calculated using exponential trendlines. For total abundances, the full incubation period was taken into account (i.e., day 1 - 6 for Amundsen Sea and day 2 - 8 for Weddell Sea bioassays). Starting abundances were taken prior to filling of the cubitainers and hence not taken into account. For the phytoplankton group specific rates only those time points (>3 but most often 4-5 time points) with a consecutive increase in abundances were selected."*

-line 623. There may be higher Mn requirements for reactive oxygen species, but there is also a Mn requirement for photosynthesis. So if you provide more Fe, and phytoplankton can build more reaction centers then their Mn requirement will also increase. See Raven 1990.
*Please see response to the next comment.*

-line 628. The work of Hawco expands on this idea, that if you just relieve Fe limitation then you likely force phytoplankton into Fe limitation due to the requirements of both TMs in photosynthesis.
So indeed your higher dMn at A1 meant that the phytoplankton community here was not forced into Mn limitation.
I think you need to be more explicit here about the Fe and Mn requirements of photosynthesis and how your initial Mn concentrations impacted your results.
*We thank the Reviewer for pointing out that the Fe and Mn requirements of photosynthesis is not explicit yet. We will add to section 4.2 lines 628 onwards: "This duality in the pMn:POP ratios is not surprising as Mn demand may not only increase under Fe stress, but it should also increase with Fe addition, as both Mn and Fe are required for photosynthesis (Raven 1990, New Phytologist, Vol 116, McCain 2021, Hawco et al. 2022). Hence, in an environment with low Mn concentrations, Fe addition*

*can consequently lead to Mn limitation (e.g., Hawco et al., 2022). Dissolved Mn concentrations at the start of bioassay A1 were relatively high, and indeed pMn:POP ratios increased with Fe addition, while concentrations of dMn decreased during the experiment. However, the low (potentially phytoplankton growth limiting) dMn concentrations in Weddell Sea bioassays from the start might have prevented a noticeable positive effect of Fe addition on dMn uptake. The higher biomass and cell abundance after Fe addition in these experiments implies the community had to make due with less Mn per cell than in the treatments without Fe addition (likely resulting in relatively low Mn quota despite elevated demand), potentially explaining why there was an increase in the pMn:POP ratios in the C and T treatments of W2, whereas this was not observed in W1 with even lower dMn starting concentrations. Such variation in apparent Mn demand and quotas likely reflects adaptive changes in nutrient uptake and storage mechanisms under nutrient stress but could also be due to different phytoplankton community compositions and/or environmental conditions."*

-line 648. I would be hesitant to call them non-essential metals. Zinc plays an important role in both intracellular CO2 transport and phosphate cycling, where copper and cadmium can be substituted in its place occasionally. Our growing understanding of these metals in other metabolic processes, outside of photosynthesis, means that whilst they may not be at limiting concentrations in the Southern Ocean, they still have a strong role to play in phytoplankton growth.
*The Reviewer is correct, we only meant Cd is non-essential, Zn and Cu definitely are. We added now that by essential metals we refer to manganese, zinc and copper, and by non-essential to cadmium.*

-line 661. Do you think warming and/or Fe would have impacted the bacterial community which may be contributing to the reported POC concentrations?
*We checked bacterial abundances (based on flow cytometry enumeration) and whilst temperature did not affect bacterial abundances significantly, iron led to higher abundances in both Weddell Sea bioassays (4.6 and 5.0 x $10^5$ cells $mL^{-1}$ for Fe addition treatments in W1 and W2 compared to 3.3 and 4.1 x $10^5$ cells $mL^{-1}$ for non-Fe treatments). However, bacterial carbon made up less than 3% of the reported POC concentrations. We will add the following to the results section, when talking about POC (line 482 onwards): "Only bioassays W1 and W2 showed a significant increase in bacterial abundances with Fe addition (final abundance 4.7 ± 0.9, 4.5 ± 0.5 vs 3.1 ± 1.0 and 4.7 ± 0.6, 5.4 ± 0.2 vs 4.4 ± 0.1 for F, TF vs C treatments in W1 and W2, respectively). However, bacteria did not have a major effect (less than 3%) on total POC concentrations." Furthermore, we will also add to the Materials and Methods: "Samples for bacterial abundances were fixed with EM-grade glutaraldehyde (0.5% final concentration; Sigma- Aldrich, Zwijndrecht, The Netherlands), flash-frozen in liquid nitrogen and stored at -80°C until analysis using flow cytometry (Marie et al.; 1999). Bacterial carbon concentrations were calculated assuming 12.4 fg C $cell^{-1}$ (Fukuda et al.; 1998, Applied and environmental microbiology, Vol 64 (9))."*

-line 669. I wonder whether you could use the different light conditions to make any inferences about how climate change is expected to alter light availability. Whilst I know it was not one of your specific treatments, the different conditions between your study areas may provide some insight.
For instance, there is conflicting evidence of both shallower and deeper mixed layers which could alter light availability in the future. Coupled with the idea of the Southern Ocean being more cloudy.
*We will add the following to the discussion, following line 674:*
*"Future light conditions in the Southern Ocean will vary for the different regions, e.g. lower sea ice coverage may enhance light availability (Leung et al., 2015, Biogeosciences, Vol 12 (19); Petrou et al., 2016, Journal of Plant Physiology, Vol 203; Krumhardt et al., 2022), whereas increased cloud coverage in the Antarctic Circumpolar Current region would reduce it (Grise et al., 2013 Geophysical Research Letters, Vol 40; Kelleher and Grise, 2021, Atmospheric Science Letters, Vol 23 (1); Krumhardt et al., 2022). Moreover, there are conflicting reports about whether mixed layer depths will increase (Leung et al., 2015) or decrease (Krumhardt et al., 2022), which directly impacts light conditions for the phytoplankton. Our results from the low light bioassay A2, showing only a small effect of Fe on phytoplankton, suggest that in regions or periods with low light, Fe increase will not drastically*

*stimulate phytoplankton growth. This highlights the importance of including light availability in Southern Ocean ecosystem (modelling) predictions."*

-line 754. Where your two study sites in similar bloom phases to make them comparable? If not, what impact do you think it has on the results?
*The bioassays in the Amundsen Sea were initiated in late February, which is towards the end of the reported bloom period (Arrigo et al., 2012, Deep Sea Research Part II: Topical Studies in Oceanography, Vol 71). The Weddell Sea bioassays were initiated in late December/early January, which is during the start of the bloom (von Berg et al., 2020, Geophysical Research Letters, Vol 47 (11)). The bloom phases were thus not comparable. We will add this information to the Material and Methods section. The differences we found between the two regions seem, however, more driven by differences in light availability, trace metal co-limitation and starting phytoplankton community. We will address this point briefly in the Discussion (line 658 onwards): "The Weddell Sea bioassays exhibited stronger Chl a accumulation, a stronger increase in $F_v/F_m$ and increased phytoplankton abundances in response to Fe addition than the Amundsen Sea bioassays, which is likely due to the lower dFe concentrations (and hence higher degree of Fe limitation for the phytoplankton typical for the Weddell Sea) at the start of the incubations. Indeed, given that the Weddell Sea bioassays were performed early in the productive season, these results imply more severe Fe limitation in the Weddell Sea whereas any Fe limitation in the Amundsen Sea likely only develops later in the season."*

-line 760. I think this may warrant some further expansion as to where and when Fe input may increase. Maybe you also need to discuss here what would be the case if Fe inputs do not increase under future climate change scenarios. Would rising temperatures alone lead to significant ecological shifts?
*Regarding differences in dFe input: In the Amundsen Sea, increased Fe input is likely to occur due to enhanced glacial melt and runoff, particularly during the summer months when melting is most pronounced (Van Manen et al., 2022). Increases in seawater temperature may affect the availability of dFe for phytoplankton, since temperature affects the oxidation of the bioavailable Fe(II) to Fe(III) (e.g. Millero et al., 1987, Geochimica et Cosmochimica Acta, Vol 51 (4)), however, Aflenzer et al. (2023) did not observe a lower bioavailability of added dFe with increased temperatures. In the Weddell Sea, Fe input may increase through upwelling of Fe-rich deep waters and meltwater from ice shelves, but this is less certain (Klunder et al., 2011, Deep Sea Research Part II: Topical Studies in Oceanography, 58 (25-26)). Seasonal variations in sea ice cover and glacial melt will play a significant role in determining the timing and magnitude of Fe input in these regions.*

*We will add this to the Introduction (line 73 original manuscript)*

*Regarding the effect of only temperature: Temperature alone showed a limited effect on phytoplankton, with only 3 phytoplankton groups (Phyto 3, Phyto 19 and diatom Phyto 22) increasing in abundances, and only Phyto 19 showing a consistent effect. Still, these groups represent pico-sized as well as larger phytoplankton (2, 8.1 and 13.3 µm diameter). Earlier studies also showed temperature to have only a limited effect on (natural) phytoplankton communities (Rose et al., 2009). Indirect effects of warming (e.g. locally high ice-melt induced freshening, dFe increase) will likely have larger impact on phytoplankton community compositions. Ice-melt induced freshening already led to a shift from diatom to cryptophyte and flagellate dominated communities in the Western Antarctic Peninsula region (reviewed by Deppeler and Davidson 2017), and increased dFe concentrations will affect phytoplankton community composition even more so when combined with temperature increases (this study; Rose et al. 2009). Furthermore, the availability of dFe is likely changing due changes in sources (see introduction) but is also influenced by siderophore production (reviewed by Gledhill et al., 2012) but warming of the Southern Ocean does not seem to have a direct effect (Sinha et al., 2019). Warming likely increases the growth rates of siderophore producing bacteria (Sinha et. al. 2019), but this may be countered by reduced siderophore production due to ocean acidification*

*(Sinha et al. 2019). Overall, predictions about future conditions and their consequences are complex and have large uncertainty, but it seems likely conditions will be temporally and spatially heterogenous with varying changes in temperature and availability of Fe (and light). For example, while the warming of surface water in the Amundsen Sea has already been observed, Weddell Sea surface temperatures for the deep basin seem relatively stable at the moment with significant warming only below 700 m (Strass et al., 2020 Journal of Climate, Vol 33(22)). However, upwelling of this warm water leads to local temperature increases in notably coastal regions (Darelius et al., 2023), potentially increasing future temperatures by over 2 °C warmer in troughs that connect the open ocean to ice shelfs (Teske et al., 2024), increasing not only temperatures but likely also glacial melt derived Fe supply. This makes it prudent to assess not only individual, but also combined effects of increasing Fe and temperature as discussed in the next section.*

*We will add these arguments to the Discussion (section 4.3).*

Reviewer 2
General comment:
This study uses bioassay experiments in the Weddell Sea and in the Amundsen Sea Polynya to study the effects of increasing Fe and temperature conditions on natural phytoplankton communities. Given the current predictions about upcoming changes in the Southern Ocean, it is critical to understand what effects these changes may have on natural communities. The manuscript is very well written, and this study is greatly strengthened by the extent of trace metal results reported (including both dissolved and particulate data).
However, several points could be improved. First, the differences in initial conditions could be discussed in more details (coastal vs offshore, difference in macronutrients, etc.).
*We will include a comparison of the initial conditions for the different bioassays to the Results (including information from Material and Methods and Table 1), as well as include depth profiles for the stations in the Supplements to provide a general overview of the initial differences between bioassays. Lines 460 – 461 will be moved to the regional differences section in the Results.*

*"3.1 Sample site characteristics*
*The in-situ temperature was below zero for all bioassays, with lowest values for Amundsen Sea bioassay A2 and Weddell Sea bioassay W2 (-1.6 °C and -1.4 °C, respectively, compared to -0.6 °C and -0.3 °C for A1 and W1). The daily average irradiance at sampling depth on day of sampling was lowest for A1 and A2, i.e., < 6 µmol quanta $m^{-2}$ $s^{-1}$, compared to 18 and 98 µmol quanta $m^{-2}$ $s^{-1}$ for W1 and W2. Dissolved inorganic macronutrient concentrations were relatively comparable between bioassays, except the silicate concentration in W1 being ~20 µM lower than for the other bioassays (but still far from limiting). Initial dFe concentrations in the Weddell Sea were lower compared to the Amundsen Sea (Table 1), as were dMn concentrations (Fig. 2). Bioassay A1 had the highest Chl a concentrations (sampled within the ASP), followed by W1. Both bioassays also had the highest share of >20 µm Chl a. The Chl a concentration of A2 was almost exclusively made up of < 20 µm sized phytoplankton (98% of total Chl a, Table 1). Flow cytometry derived phytoplankton abundances were highest for the Amundsen bioassays. The photosynthetic efficiency Fv/Fm at the start of the incubations was 2-fold lower for the Weddell Sea bioassays compared to the Amundsen Sea bioassays (i.e., 0.3 vs 0.6 r.u., respectively). The station for bioassay W2 was closest to the coast, followed by A1, A2 and W1, however distance to land did not seem to have a major impact on either phytoplankton community composition, or nutrient concentrations."*

Further, the photophysiological results (Fv/Fm and more?) should be referred to more often to strengthen some statements.
*We will refer to the $F_v/F_m$ results more consistently, e.g. in lines 594 and 658 of the original manuscript. See also our reply to the comment on the topic by Reviewer 1.*

You could also add more information on the biological results that were briefly mentioned (e.g., Chl:C ratios) to confirm your hypothesis.
*We thank the Reviewer for their suggestion. We will add the following at 666 of the original manuscript: "The relatively low Chl:POC ratios in the Weddell Sea bioassays (average over all treatments 0.003 ±0.003 vs 0.008 ±0.002 for the Amundsen Sea bioassays) may indicate stronger Fe limitation, since Fe limited cells are known to have a lower Chl:POC ratio compared to non-limited cells (Moore et al. 2007, Deep-Sea Research Part II: Topical Studies in Oceanography, Vol 54 (5-7))."*

The figures could also be improved with bigger panels and the statistically significant differences displayed to help result visualisation and interpretation.
*We will adapt the figures as suggested (e.g. increase panel size and change the colour for the TF treatment). Adding significance indicators to the figures will in our opinion give the impression that we used pairwise comparisons. However, the test used is not based on pairwise comparisons, hence we cannot add meaningful indicators for statistical significance to the figures. We will add a table to the supplements providing the statistical results for the test used.*

Finally, some typos need fixing throughout the manuscript (use of abbreviations and then not, some references to fix, some commas near period).
*We carefully checked the manuscript and tried to make sure the revised manuscript is thoroughly corrected.*

Below, I have provided line comments which I hope will help improve the manuscript.
Line comments
-L68 This first long intro paragraph could be split for easier reading.
*Thank you for bringing this to our attention. We will add paragraph breaks in line 65 and line 78 to facilitate reading.*

-L71 fix ref
*We will double check all references and fix them, where necessary.*

-L105-107 This info could be removed from the introduction as you describe it well in your method.
*We will shorten this sentence in the introduction to: "Our bioassay treatments comprised a full factorial combination of Fe and temperature increases."*

-L55-120 Considering how many times you speak of Mn in your results and discussion, I wonder if you should talk about it in your introduction and describe its essential roles in phytoplankton.
*We will add to the introduction line 77 onwards: "*
*Besides Fe and temperature, there are also other factors, e.g., other bio-essential metals (Mn, Co, Ni, Cu and Zn), where notably Mn has been shown to be (co-)limiting in the Southern Ocean (Wu et al; 2019, Browning et al., 2021, Balaguer et al.; 2022, Hawco et al.; 2022). Mn is essential for phytoplankton photosystems (Raven et al., 1990) and a co-factor for enzymes dealing with oxidative stress (Wolfe-Simon et al., 2005, Journal of Phycology, Vol 41 (3)). Moreover, light is another major limiting factor for phytoplankton growth in Southern Ocean (e.g. van Oijen et al.; 2004, Journal of Plankton Research, Vol 26 (8), Strzepek et al.; 2019, Proceedings of the National Academy of Sciences, Vol 116 (10), Vives et al.; 2022, Journal of Marine Systems, Vol 234, Latour et al.; 2023, Limnology and Oceanography Letters, Vol 9 (2))."*

-Figure 1: I think you are missing in your result and discussion a general description of your different sites. Both regions are separated but within them you present results from coastal and (almost?) offshore data? This may greatly impact your results too.
*We are hesitant to put too much emphasis on regional differences, given that we only performed two experiments in each region, and given that light conditions differed. We will incorporate a section on*

*initial characteristics of the locations as a first section in the Results, please see our response to your first general comment.*

-L137 what is 'PVDF'?
*PVDF is a type of plastic (Polyvinylidene fluoride). We will spell it the first time we use it.*

-L143 ASP previously defined
*Thank you for noting. We will change accordingly.*

L146 Why this difference in duration?
*The difference in duration between the Amundsen Sea and Weddell Sea bioassays was due to logistical constraints (we will add this to the Materials and Methods section). Amundsen Sea bioassays were conducted before Weddell Sea bioassays and unfortunately, time was very limited. Given the small effects observed in the Amundsen Sea bioassays, we decided to adapt light conditions and extend the duration of the Weddell Sea bioassays to allow for a more comprehensive assessment of the treatment effects.*

-Table 1: could the depth profiles of each station be presented in SI so we can better visualize the different initial conditions?
*We will add depth profiles for each station to the Supplements.*

-L183 typo
*Thank you for pointing this out, we will change the text accordingly.*

-L191 How were these light levels achieved in your experiment?
*Light levels were adapted using neutral density screens, we will clarify this in the Material and Methods following line 193.*

-L245 Can you clarify if you expect your pTM measurements to include lithogenic material as well?
*Yes, our pTM measurements include lithogenic material. We will add the following text to the Material and Methods section 2.5, following line 262: "The lithogenic fraction and concentration of pFe and other particulate metals discussed was determined by assessing the ratio between the particulate metal of interest and particulate aluminium (pAl), assuming all pAl originates from crustal material using the approach described in more detail in van Manen et al (2022). For example, we are using the observed pFe/pAl ratio in the samples and the known crustal ratio of 0.21 mol mol$^{-1}$ (Taylor and McLennan, 1985) to calculate the lithogenic pFe fraction and concentration~~, see supplement xx for more details.~~"*
*And: "The EDTA oxalic acid wash used on particulate samples prior to filtration should effectively remove surface-bound metals, also minimizing the authigenic Fe fraction" following line 246.*

*In the Results (following line 435), we will add that lithogenic particles provided a consistent background that did not affect observed changes between the treatments, and we will add the respective data to the Supplements.*

-L248 at the end of the experiment?
*Yes, all filters collected (stored at -20°C) were taken back to the NIOZ at the end of the cruise to be processed and analysed.*

-L298 could you add info on the type of measurements (flash sequence etc…)
*The measuring light frequency used was set to level 5 (25 Hz) with an intensity of 8, the SAT-pulse width was set to 0.8 seconds and the far-red pulse width was set to 10 seconds, with intensities of 10 and 6, each. We will add this information to the Material and Methods, section 2.8.*

-L303 Did you also rinse the cuvette with the (filtered) sample itself?
*No, the cuvette was not rinsed with the sample before measuring, but UP water was removed from the cuvette by shaking and placing the cuvette upside down on lint-free paper towels to remove last droplets. We also measured technical replicates per biological replicate, between which the cuvette was not cleaned with UP water. There was no significant difference in Fv/Fm between the different replicates (non-parametric Kruskal-Wallis ANOVA, p = 0.95), indicating that rinsing with UP water did not impact reported values. We will add this to the Materials and Methods section.*

-L307 Can measurements of the functional absorption cross section of PSII be derived from this instrument too?
*Unfortunately, the functional absorption cross section of PSII cannot be determined with the instrument used.*

-Figure 3: it would be helpful to have the significant differences displayed on the figure.
*Adding significance indicators to the figures will in our opinion give the impression that we used pairwise comparisons. However, the test used is not based on pairwise comparisons, hence we cannot add meaningful indicators for statistical significance to the figures. We will add a table to the Supplements providing the statistical results for the test used.*

-Figure 4: please fix the x-axis label.
*We will change the x-axis label accordingly.*

-L594-596 You could also refer to Fv/Fm to support this statement.
*The reviewer is correct, we will add Fv/Fm in this sentence (here underlined for clarity): "However, since phytoplankton abundances, $F_v/F_m$ and Chl a concentrations were not higher in T treatments compared to the control, and since phytoplankton requires less Fe at higher temperatures (Jabre & Bertrand, 2020), this is less likely."*

-L596 It is hard to visualize this trend because of the different scales in Figure S2. It would be good to refer to the lower Si initial conditions at W1 compared to A1 and A2. Also, the t0 of Figure S2 does not match your value in Table 1 for A1 and W2? Same for NOx of A1.
*W1 showing the strongest decline in Si concentrations should still be visible in Fig. S2, given that the tick marks have the same distance despite the different scales. We will add to the caption that tick marks represent the same concentration intervals aiding comparison.*
*We are sorry for the typos in Table 1. The correct values are the ones displayed in figure S2, we corrected the Table.*

| Bioassay | Station | Lat. (°S) | Long. (°W) | Temp. (°C) | Salinity (psu) | Irradiance (µmol quanta $m^2$ $sec^{-1}$) | Si (µM) |
|---|---|---|---|---|---|---|---|
| A1 | 31 | 73.50 | 116.50 | -0.6 | 33.99 | 5.0 | 77.9 |
| A2 | 52 | 72.00 | 118.42 | -1.6 | 33.89 | 3.1 | 77.5 |
| W1 | 17 | 65.00 | 000.00 | -0.3 | 33.90 | 17.7 | 58.3 |
| W2 | 36 | 70.08 | 011.08 | -1.4 | 33.82 | 97.6 | 71.8 |

| Bioassay | $PO_4$ (µM) | NOx (µM) | Fe (nM) | total Chl *a* (µg $L^{-1}$) | < 20 µm Chl *a* (%) | Phyto ($x10^3$ $mL^{-1}$) | $F_v/F_m$ r.u. |
|---|---|---|---|---|---|---|---|
| A1 | 1.8 | 24.3 | 0.28 | 3.0 | 42 | 8.4 | 0.6 |
| A2 | 2.0 | 28.2 | 0.10 | 0.4 | 98 | 7.1 | 0.6 |
| W1 | 1.6 | 24.0 | 0.05 | 1.5 | 24 | 5.6 | 0.3 |
| W2 | 1.9 | 27.9 | 0.03 | 0.6 | 65 | 4.4 | 0.3 |

-L610 How did you calculate the growth rates?

*Phytoplankton net growth rates are based on an exponential trend line per replicate for each treatment per bioassay and were originally only determined for total phytoplankton abundances. We have now included phytoplankton group specific growth rates. We will include how phytoplankton net growth rates were calculated in the Material and Methods: "Phytoplankton net growth rates were calculated using exponential trendlines. For total abundances, the full incubation period was taken into account (i.e., day 1 - 6 for Amundsen Sea and day 2 - 8 for Weddell Sea bioassays). Starting abundances were taken prior to filling of the cubitainers and hence not taken into account. For the phytoplankton group specific rates only those time points (>3 but most often 4-5 time points) with a consecutive increase in abundances were selected."*

-L642 are you referring to the 3 experiments? In your results I think you mentioned differences to the C and not the T.

*We calculated the difference in pCu:POP ratios for the Fe treatments compared to the control to avoid too many comparisons. However, statistical results indicated that Fe addition treatments showed differences in the pCu:POP ratios compared to both treatments without Fe addition (i.e. control and T treatment), and both the control and T treatment showed higher pCu:POP ratios. We are referring to all 3 experiments, however, the effect was most visible for bioassay W2 and will change the text to make this clearer (changes underlined for clarity): "For example, cellular Cu requirements increase under Fe limitation (Schoffman et al., 2016), which could explain the higher pCu:POP ratios in the C and T treatments compared to the Fe addition treatments <u>in all bioassays analysed</u> (Fig. 3)."*

-L659 maybe I missed it, but you should also discuss that some bioassays have an increase in Chl in the control compared to t0 (A1, A2, W2) while W1 did not and why is that.

*We will add discussion on the topic when we relate the results of W1 with the bioassay S54-65 from the Viljoen et al. (2018) study: "The location the seawater for bioassay W1 was taken has similar coordinates as bioassay S54-65 in a study by Viljoen et al. (2018). These authors sampled 3 weeks later (different year) and at a comparable depth (30 m vs 20 m in our study) and found largely similar responses by the phytoplankton to dFe addition, i.e., total Chl a increased by ~2 µg Chl a $L^{-1}$ and diatoms dominated the phytoplankton community. In contrast to W1 but comparable to our other bioassays, total Chl a concentration in bioassay S54-65 (Viljoen et al. 2018) increased in the control over the duration of the bioassay. The lack of increase in Chl a in the control (and T) treatment of W1 might be explained by a lower in-situ dFe for W1, indicating a stronger limitation of dFe. At the same time, POC (and < 20 µm Chl a) concentrations did show an increase over time in the control (and T)*

*treatment of bioassay W1. Moreover, bioassay W2, with even lover starting concentrations of dFe, showed an increase in Chl a over time for the control. Given the lowest dMn concentrations in W1, it might be that dMn and not (only) dFe was limiting the production of reaction centres (Raven et al.; 1990), resulting in Chl a concentrations to not increase. Given the increased requirement for Mn under low Fe (Peers & Price; 2004), Fe addition may have relieved Mn limitation in the Fe addition treatments slightly, resulting in the observed increase of Chl a in those treatments."*

-L661 you could also refer to your Fv/Fm results here.
*The Reviewer is correct, we will add Fv/Fm to this part of the Discussion:*
*"The Weddell Sea bioassays exhibited stronger Chl a accumulation, a stronger increase in $F_v/F_m$ and increased phytoplankton abundances in response to Fe addition than the Amundsen Sea bioassays, which is likely due to the lower dFe concentrations (and hence higher degree of Fe limitation for the phytoplankton typical for the Weddell Sea) at the start of the incubations."*

-L664 Could these results (Chl:POC) be presented in SI? So we can see the difference between treatments. Results of fluorescence per Chl may also be interesting in this context?
*We will add the Chl:POC ratios as a Supplement, as well as F0 values from our $F_v/F_m$ measurements.*

-L666-669 Do you mean that in-situ phytoplankton were light-limited? It is expected which such low levels.
*Indeed, we expect in-situ phytoplankton to be light-limited, as we indicated in line 670.*

-L671 Figure 2 still shows a great depletion of the added Fe by day 3. Do you think it is due to phytoplankton uptake or could be sorption? POC does not show any change but Chl does although with n=1. Do you think another parameter could have become limiting after day 3?
*The observed depletion of added Fe by day 3 could be attributed to several factors, but the most likely is a combination of precipitation and wall adsorption, and perhaps luxury uptake as explained in section 2.3. Even in UP water, the Fe concentrations decreased albeit less rapidly than in natural seawater (with more (particle) surfaces available), where the difference between Fe amended and control treatments often became undisguisable based on concentrations alone (see section 3.1). This implies sorption of Fe due to its high affinity for surfaces plays a role, where we envision an equilibrium concentration (balance between sorption and dissolution) develops after Fe addition that is driven by the type and amount of (particle) surface area available. If there is also significant biological uptake, the equilibrium will re-establish, leading to desorption maintaining the values near equilibrium despite uptake (till all available Fe is used). As such, not much can be concluded from the dissolved concentration alone and hence, we switched to the practice of using $d^{57}Fe$ for the Weddell Sea experiments to better track the Fe addition as described in this manuscript.*
*The decline in dFe is what we would expect, so we should be careful not to overinterpret this feature. Regarding whether another parameter became limiting in A2 after day 3: It is possible that another parameter became limiting, but given that we measured all macro- and micronutrients, and none was drawn down to limiting concentrations, it is unlikely it was nutrient limitation. Light conditions were likely limiting, potentially causing POC levels not to increase (please also see our response to the next comment).*

-L674 these low light levels are comparable to this study: https://doi.org/10.1002/lol2.10366.
*Thank you for the reference. We will change lines 671 and following: "Although earlier studies reported positive responses of phytoplankton to Fe addition also under low light conditions (Viljoen et al., 2018; Alderkamp et al., 2019), the light intensities used for the low light treatment in those studies were still relatively high (i.e., 15 and 30 µmol quanta $m^{-2}$ $s^{-1}$) and well above those in A1 and A2 (average of 3.4 and 1.5 µmol quanta $m^{-2}$ $s^{-1}$). In addition to higher light levels, the lower initial dFe concentrations in the Ross Sea study (Alderkamp et al. 2019) compared to our study indicate a stronger Fe limitation and subsequently a stronger response to dFe addition. A recent study on Southern Ocean deep chlorophyll maximum phytoplankton responses to Fe addition (Latour et al.,*

*2023) reported a Chl a increase at low light intensities (similar to our Amundsen Sea bioassays) and no change of POC (similar to bioassay A2) until light levels increased. This supports our suggestion that the low light condition in A2 was a determining factor for the lack of a response to dFe addition."*

-L676 Do you think you could have seen a response if the experiment lasted longer? With the initial combination of very low temperature, light and biomass at that station.
*Given that the light levels stayed low, we do not think extending the incubation would have made much diference. Both $F_v/F_m$ and total phytoplankton abundances over time already showed levelling off at the day the experiment was ended. Only in case weather conditions had improved, leading to higher light levels, may we have seen a response.*

-L677 Or maybe phytoplankton were just not Fe-limited at the A2 station, as supported by the high Fv/Fm. It is likely the cells were content with the slow growth rates and would only need more Fe if light levels were increased (refer to the paper mentioned in one of the previous comments + https://doi.org/10.1073/pnas.1810886116)
*We agree that maybe phytoplankton in bioassay A2 were not limited by dFe concentrations as indicated in line 670 – 671 of our original manuscript. We will clarify better and add the following: "The high initial $F_v/F_m$ value suggests that the phytoplankton may not have been limited by dFe (under these low light conditions) and would only require more dFe once light intensities increased again (Strzepek et al.; 2019, Vives et al.; 2022, Latour et al.; 2023). The small increase in $F_v/F_m$ in the Fe addition treatments may suggest growth became dFe limited during the incubation (Fe-addition did show a significant effect on $F_v/F_m$ at the last day of the incubations), despite the light conditions remaining low."*

-L686 Do you mean for A1 only? It is hard to visualize this treatment effect in Figure 4.
*The effect was stronger in bioassay A1, however also bioassay A2 showed a significant positive effect of Fe addition on Fv/Fm values. We will adapt the symbol-sizes in the figure to make differences easier to see.*

-L696 Which figure are you referring to? I cannot see this in Figure 6.
*We agree that for some phytoplankton groups, differences are hard to see in this figure and will increase the panel sizes of the figures to make it easier to see such differences. The effect of temperature on phytoplankton groups 19 and 22 can also be seen in the Supplemental data where all phytoplankton abundances for each day of the bioassays are given. Phyto 19 showed higher abundances in the T and TF treatments in bioassays A1 and A2 (not present in Weddell Sea bioassays) and Phyt 22 showed an increased abundance in the T and TF treatments for both A2 and W2. We will also include Phyto 3 here, since it showed increased net growth with temperature for bioassay A1.*

-L700 change in what?
*L700 in the original manuscript refers to changes in phytoplankton abundances. We will clarify this.*

-L764 but POC results were not extensively described in this study.
*L764 refers to how a change in phytoplankton community composition and size structure may affect organic matter fluxes in the Southern Ocean. This part of the Conclusions is now moved to the Discussion based on a suggestion by Reviewer 3.*

Reviewer 3
General comments:
A comprehensive examination of iron and temperature modulation of natural phytoplankton communities was conducted through two sets of bioassay experiments in two different regions of the Southern Ocean, namely the Amundsen Sea and the Weddell Sea, over the summer period. The experimental design was well-planned and executed with consideration of future climate change predictions. The resulting data and supporting information are structured and comprehensively

presented in the manuscript. The future recommendations further enhance the relevance of this research and the propensity for continued investigations into related fields by highlighting several knowledge gaps.

Detailed and technical comments:
I have outlined some suggested changes to consider for each section, as well as some minor technical corrections. My main suggestion would be that some statements in the discussion could benefit from including more recent publications where relevant to confirm or explain the results. Additionally, the conclusion could further be finessed to enhance clarity and impact.
*We will include the suggested references to the Discussion and will change the Conclusion to be more concise (see our response to your Conclusion-specific comment later on).*

Introduction
The introduction adequately highlights the knowledge gaps and thus provides the rationale for the study on temperature-enhanced effects of iron on the natural phytoplankton community in the different Southern Ocean regions. However, the results and discussion additionally introduce other parameters which could have been briefly included as part of the literature in the introduction.
*We will add to the Introduction line 77 onwards: "Besides Fe and temperature, there are also other factors, e.g., other bio-essential metals (Mn, Co, Ni, Cu and Zn) where notably, Mn has been shown to be (co-)limiting in the Southern Ocean (Wu et al; 2019, Browning et al., 2021, Balaguer et al.; 2022, Hawco et al.; 2022). Mn is essential for phytoplankton photosystems (Raven et al., 1990) and a co-factor for enzymes dealing with oxidative stress (Wolfe-Simon et al., 2005). Moreover, light is another major limiting factor for phytoplankton growth in Southern Ocean (e.g. van Oijen et al.; 2004, Strzepek et al.; 2019, Vives et al.; 2022, Latour et al.; 2023)."*

Line 60: The references listed for 'phytoplankton growth often becomes limited by low iron (Fe) availability' are some examples of the many existing publications. Rather indicate that these are some references ('e.g.') to the magnitude of studies that have indeed established that phytoplankton growth becomes limited by low Fe in different ocean basins.
*The Reviewer is correct and we will add an "e.g.," to these references.*

-Line 62: Consider adding 'Milligan and Harrison, 2000' for the 'nitrate assimilation' reference (https://onlinelibrary.wiley.com/doi/full/10.1046/j.1529-8817.2000.99013.x).
*We thank the Reviewer for the reference and will add it accordingly.*

-Line 70: Another reference to consider for 'Fe supply by increased wind-driven mixing' is 'Moreau et al. 2023' (https://www.nature.com/articles/s41467-023-36992-1).
*We thank the Reviewer for the reference and will add it.*

-Line 71: Typo 'L.' in the citation: 'L. Seyitmuhammedov et al., 2022'.
*We fixed the typo.*

-Line 92: Could you expand a little on the 'Fe from a variety of sources' apart from the seafloor?
*We will add the underlined part to the sentence in line 92: "The subpolar cyclonic Weddell Gyre circulating in the Weddell Sea basin isolates the centre of the Weddell Sea from marginal Fe sources such as melt or sediments, whilst the currents on the edges of the gyre have the potential to pick up Fe from a variety of sources, such as the seafloor, bathymetry driven mixing with deeper water masses, and sources associated with ice melt (Raiswell et al., 2008, Geochemical transactions, Vol 9; Shaw et al., 2011, Deep Sea Research Part II: Topical Studies in Oceanography, Vol 58 (11-12); Klunder et al., 2014, Biogeosciences, Vol 11 (3); Annett et al.; 2015; Sherrell et al., 2015; Lannuzel et al., 2016; Raiswell et al., 2016, Biogeosciences, Vol 13 (13); Hopwood et al., 2019; Van der Merwe et al., 2019; Gerringa et al., 2020; Sieber et al., 2021, Earth and Planetary Science Letters, Vol 567; Seyitmuhammedov et al., 2022; Tian et al., in prep.)."*

Materials and Methods
The materials and methods section was succinct. The section on the setup verification is much appreciated to remove any doubt of contamination issues, particularly for incubations performed while out at sea.
Figure 1: I would have appreciated seeing some information on the hydrography of the sampling sites, or even the Chla distribution in the map (separately). However, I understand that it is not so trivial, given the different sampling timelines.
*We will add a depth profile for each station to the Supplements, as well as Chl a data based on NASA worldview at the time of sampling.*

-Line 141: What was the average PAR under the 'dimmed light conditions?
*We did not measure the PAR in the clean container, however, the samplers are light proof and cubitainers were covered with black opaque bags to avoid light stress when transported on deck. We will add to line 193 in the Material and Methods section 2.2: "During transport on deck, cubitainers were covered with black light-proof bags to avoid light stress."*

-Line 147: '28 December 2018 to 5 January 2019'
*We will change the text accordingly.*

-Line 153: Table 1:

- In the methods you refer to 'silicic acid'. Ensure it is clear that silicic acid is indeed the reported 'silicate'. If not, make this clear.
- It should be clearer if 'Fe' refers to dissolved Fe (dFe) only.
- Chl *a* (italicize a)

*Silicic acid is indeed the reported silicate, we will change the text accordingly and be consistent with the names. We use dFe when referring dissolved iron only, and Fe when talking about iron in general, and will check that we stay consistent in this.*

-Line 204: Could you please clarify what threshold/range is 'consistently low'?
*The ranges measured are given in lines 201 – 203 of the original manuscript. Concentrations within one standard deviation of the mean starting values (0.12 nM) were considered consistently low.*

-Line 298: Section 2.8: Limited information on the instrument operation for the photophysiological data acquisition is given. Was any form of post-processing conducted on the raw Fv/Fm data? Or was this not necessary based on the data acquisition from the PAM?
*No post-processing was necessary on the raw $F_v/F_m$ data, and we will add more information on the measurement specifics.*

-Have you considered investigating the effective absorption cross-section (σPSII) from the photophysiological results? Could it further support the outcomes and contextualize the results in terms of stress on the photosystem or help estimate the primary production rates?
*Unfortunately, the instrument used does not permit for us to calculate the effective absorption cross section.*

-Line 305: Perhaps you could reference 'Cullen and Davis, 2003' for the choice of 0.2 µm filtered blank corrections (Cullen JJ, Davis RF (2003) The blank can make a big difference in oceanographic measurements. Limnol Oceanogr Bull 12:29–35)?
*We thank the reviewer for the reference and will add it.*

Results:
In the introduction, you highlighted that 'Generally, the Weddell Sea has a relatively low primary productivity', while 'the west Amundsen Sea and specifically the Amundsen Sea Polynya (ASP) is

known as one of the most productive regions in the Southern Ocean in terms of net primary production per net area'.
Based on these statements, it would be nice to see a brief contrast of the initial conditions, as well as the results obtained in these two areas. This may also be a precursor to a concluding statement about the differences in the temperature-enhanced effects of the phytoplankton from these two regions.
In the abstract, I missed specific outcomes that are expected to be different for these two regions
*We are hesitant to put too much emphasis on regional differences, given that we only performed two experiments in each region, and given that light conditions differed. We will, however, add a section on initial differences between the bioassays in the Results.*
***"3.1 Sample site characteristics***
*The in-situ temperature was below zero for all bioassays, with lowest values for Amundsen Sea bioassay A2 and Weddell Sea bioassay W2 (-1.6 °C and -1.4 °C, respectively, compared to -0.6 °C and -0.3 °C for A1 and W1). The daily average irradiance at sampling depth on day of sampling was lowest for A1 and A2, i.e., < 6 µmol quanta $m^{-2}$ $s^{-1}$, compared to 18 and 98 µmol quanta $m^{-2}$ $s^{-1}$ for W1 and W2. Dissolved inorganic macronutrient concentrations were relatively comparable between bioassays, except the silicate concentration in W1 being ~20 µM lower than for the other bioassays (but still far from limiting). Initial dFe concentrations in the Weddell Sea were lower compared to the Amundsen Sea (Table 1), as were dMn concentrations (Fig. 2). Bioassay A1 had the highest Chl a concentrations (sampled within the ASP), followed by W1. Both bioassays also had the highest share of >20 µm Chl a. The Chl a concentration of A2 was almost exclusively made up of < 20 µm sized phytoplankton (98% of total Chl a, Table 1). Flow cytometry derived phytoplankton abundances were highest for the Amundsen bioassays. The photosynthetic efficiency Fv/Fm at the start of the incubations was 2-fold lower for the Weddell Sea bioassays compared to the Amundsen Sea bioassays (i.e., 0.3 vs 0.6 r.u., respectively). The station for bioassay W2 was closest to the coast, followed by A1, A2 and W1, however distance to land did not seem to have a major impact on either phytoplankton community composition, or nutrient concentrations."*

-Line 483 and elsewhere: When referring to significant differences in Chla concentrations from bioassays, the Chla concentrations are a useful baseline. However, Chla growth rates are nuanced for assessing significant differences in phytoplankton responses from the bioassay experiments. Thus, it might be useful to reassess the significant differences in Chla by evaluating their growth rates instead. You already mentioned elsewhere Chla growth rates, but I did not easily find how this was calculated.
*The net growth rates mentioned in the manuscript are based on total phytoplankton abundances, not on Chl a. Phytoplankton growth rates are based on an exponential trend line per replicate for each treatment and each bioassay, which we will add to the Material and Methods section. For Chl a, we unfortunately had limited time points, preventing proper rate calculations.*

-Line 420: Figure 2: Typo: 'Weddell Sea (W1: c, e, i; W2: d, f, j)'?
*Thank you for noticing the typo, we will change the figure legend.*

-The red and purple lines blend too well with each other. Consider a darker shade of purple or another colour. Apply comments to other similar figures.
*We will adapt the figures accordingly.*

-Line 423: missing ')' after 'day 3'?
*We will add the missing bracket.*

-Line 451: Figure 3: Typo: 'Amundsen Sea A1 (a, f, i, l, o)?
*Thank you for noticing the typo, we will change the figure legend.*

-Line 470: Figure 4: '(a), (b), (c) and (d)' – change to lowercase to be consistent.

We will change this.

-Line 109-110: Since short-term local temperature increases can be expected in the Weddell Sea, can you comment on the short-term temperature increase effects from your bioassays in W1 and W2? Would using lower incubation temperatures for this region be justifiable instead?
*Temperature alone did not have a major effect on phytoplankton in the Weddell Sea, thus the short-term small temperature increases (Darelius et al., 2023) as such are unlikely to have a large, direct impact. However, if such temperature increase occurs in a period of time when dFe concentrations are high(er), we could expect some restricted local and shorter-term) phytoplankton responses (growth, increase in Chl a concentrations and POC). When these short-term increases occur on top of general warming, it may cause a (small) response by the phytoplankton. Still, we expect mostly an indirect effect from associated freshening (Darelius et al., 2023).*

-Line 399: I missed how the nutrient drawdown was calculated. It would be nice to see a summary table or figure for the nutrient drawdown and the Chla drawdown, respectively. It is confusing to follow the results otherwise.
*We did not calculate nutrient drawdown rates but compared nutrient concentrations at the start and end of the incubation period as nutrient drawdown, and additionally compared end concentrations between treatment to determine stronger and/or weaker nutrient drawdowns. We will add total drawdowns to the Supplements.*

-Line 406: 'Silicate acid' or just 'silicate'? Check consistency in the use of terms throughout the manuscript.
*It should indeed be silicate. We will change this throughout the manuscript to be consistent.*

-Line 551: potential typo: '(Fig. 6d,h, $p<0.01$ for all).' And (F and TF, Fig. 6h, $p < 0.01$)?
*Thank you for noticing the typo, we should indeed be referring to Figure 6, not 16.*

Discussion:
Line 608: 'Dissolved Mn is known to (co-)limit Southern Ocean phytoplankton growth and community composition (Balaguer et al., 2022).'
Mn is known to (co-)limit together with? Are these co-limitations necessarily seasonal?
Consider: Pausch, et al. 2019.
(https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0221959)
Latour, et al. 2023. (https://online.ucpress.edu/elementa/article/11/1/00022/197210/Seasonality-of-phytoplankton-growth-limitation-by)
*We thank the Reviewer for the references. We will edit the following section (following line 607; changed sections are underlined for easier following): "Dissolved Mn is known to (co-)limit Southern Ocean phytoplankton growth and community composition together with Fe (Browning et al. 2021, Balaguer et al. 2022). Under such conditions, dFe addition alone positively impacts Chl a concentrations, phytoplankton abundances and POC concentrations, but a combination of dFe and dMn addition can lead to higher increases in these variables (Pausch et al. 2019, Browning et al., 2021). Nevertheless, dMn addition effects can often be masked by the effects of dFe addition (Latour et al. 2023), and dFe addition alone can already lead to increases in Chl a even in primarily Mn-limited areas (Browning et al., 2021). This fits our results showing increases in in Chl a concentrations with dFe addition. Also net growth rates based on total phytoplankton abundances showed increases (i.e. 1. 5 (0.20 ± 0.05 vs 0.12 ± 0.02 d-1) and 1.4-fold (0.24 ± 0.01 vs 0.18 ± 0.01 d-1) higher for Fe-addition treatments (F and TF) compared to the control for bioassays W1 and W2. The lower starting concentrations of dMn in W1 compared to W2 may have contributed to the 2-fold lower phytoplankton net growth rates in W1 compared to W2, independent of the treatment. Our data indicate potential dMn/dFe colimitation in the Weddell Sea already in early summer. Since the requirements for dMn and dFe differ between different phytoplankton groups (Arrigo, 2005; Twining & Baines, 2013; Balaguer et al., 2023), we suggest that the (co-)limitation of dMn and dFe may be*

*affected by phytoplankton community composition. Considering that Mn limitation can be seasonal (Latour et al., 2023), we also urge to study different stages of the phytoplankton bloom period."*

-Line 635: Typo: '.,'
*Thank you for noticing, we will change the text accordingly.*

-Line 637: Is this increased Mn uptake consistent with the needs of the phytoplankton community found at A1?
*Given that diatoms are highly abundant in the Fe addition treatments (both F and TF), and it is known for diatoms that their dMn demand increases with dFe addition (McCain 2021 Raven 1990, Hawco et al. 2022), we do think that the increase in Mn uptake is consistent with the needs of the phytoplankton community for those treatments in bioassay A1. However, diatoms are also known to have an increased Mn demand under Fe limitation (Peers & Price 2004). There is evidence (Hawco et al. 2022) for an increased Mn demand with increasing dFe concentrations for both a diatom- and a nanophytoplankton based model, suggesting that the increased Mn uptake observed for bioassay A1 is indeed consistent with the needs of the phytoplankton community. We will add the following to the Discussion on pMn:POP: "This duality in the pMn:POP ratios is not surprising as Mn demand may not only increase under Fe stress, but it should also increase with Fe addition, as both Mn and Fe are required for photosynthesis (Raven 1990, McCain 2021, Hawco et al. 2022). Hence, in an environment with low Mn concentrations, Fe addition can consequently lead to Mn limitation (e.g., Hawco et al., 2022). Mn concentrations at the start of bioassay A1 were relatively high, and indeed pMn:POP ratios increased with Fe addition, while concentrations of Mn decreased during the experiment. However, the low (potentially phytoplankton growth limiting) Mn concentrations in Weddell Sea bioassays from the start might have prevented a noticeable positive effect of Fe addition on Mn uptake. The higher biomass and cell abundance after Fe addition in these experiments implies the community had to make due with less Mn per cell than in the treatments without Fe addition (likely resulting in relatively low Mn quota despite elevated demand), potentially explaining why there was an increase in the pMn:POP ratios in the C and T treatments of W2, whereas this was not observed in W1 with even lower Mn starting concentrations."*

-Line 664: The average Chl a:POC ratio over all treatments for the Weddell Sea bioassays were 0.003 ± 0.003. Could you comment as to why the Chl a content is very low relative to the POC? Is this because of a significant variability in this ratio across different treatments?
*The significant difference between treatments is only partly explaining the low Chl:POC ratios for the Weddell Sea. Fe addition did have a significant (positive) impact on Chl:POC ratios for Weddell Sea bioassays, however these ratios are still lower compared to the Amundsen Sea bioassays. Ratios were 0.003 and 0.004 for the F and TF treatment in both W1 and W2, with ratios for the C treatment being 0.002 and 0.003 for W1 and W2, respectively (no difference between C and T treatments). In contrast, Chl:POC ratios for Bioassays A1 were 0.006, 0.008, 0.009 and 0.011, and for Bioassay A2 0.005, 0.006, 0.007 and 0.009, for the C, T, F and TF treatments, respectively. As mentioned in line 665 in the original manuscript, we assume that Chl:POC values were higher in the Amundsen Sea as an adaption to low light. Moreover, the higher Chl:POC ratios in the Amundsen Sea (and consequently the lower ratios in the Weddell Sea) might also be a sign for the difference in Fe limitation (and possibly Mn limitation) between the Weddell Sea and Amundsen Sea bioassays, since Fe-limited cells are known to have a lower Chl:POC ratio compared to non-limited and/or replete cells (Moore et al. 2007).*
*We will add to line 666 in the original manuscript: "The relatively low Chl:POC ratios in the Weddell Sea bioassays (average over all treatments 0.003 ±0.003 vs 0.008 ±0.002 for the Amundsen Sea bioassays) may indicate stronger Fe limitation, since Fe limited cells are known to have a lower Chl:POC ratio compared to non-limited cells (Moore et al. 2007)."*

-Line 673: The bioassays conducted by Viljoen et al. (2018) were in the Weddell Sea, while Alderkamp et al. (2019) conducted their bioassays in the Ross Sea. The bioassays presented in this manuscript were conducted in both the Amundsen Sea and Weddell Sea. The sampling season and location of

W1 coincided with bioassay 'S54–65' by Viljoen et al. (2018). However, no comparisons seem to have been made or conclusions drawn regarding the outcomes based on similar and variable initial conditions to the overall outcomes from the bioassays.
Instead, only the 'low light conditions' where the light intensities differed due to Sea regions between this manuscript and Viljoen et al. (2018) as well as in Alderkamp et al. (2019) were highlighted.

*We thank the Reviewer for pointing out that bioassay S54-65 by Viljoen et al 2018 was performed at a similar/closeby location as bioassay W1 and will add a comparison of these specific bioassays to our discussion: "The location the seawater for bioassay W1 was taken has similar coordinates as bioassay S54-65 in a study by Viljoen et al. (2018). These authors sampled 3 weeks later (different year) and at a comparable depth (30 m vs 20 m in our study) and found largely similar responses by the phytoplankton to dFe addition, i.e., total Chl a increased by ~2 µg Chl a $L^{-1}$ and diatoms dominated the phytoplankton community. In contrast to W1 but comparable to our other bioassays, total Chl a concentration in bioassay S54-65 (Viljoen et al. 2018) increased in the control over the duration of the bioassay. The lack of increase in Chl a in the control (and T) treatment of W1 might be explained by a lower in-situ dFe for W1, indicating a stronger limitation of dFe. At the same time, POC (and < 20 µm Chl a) concentrations did show an increase over time in the control (and T) treatment of bioassay W1. Moreover, bioassay W2, with even lover starting concentrations of dFe, showed an increase in Chl a over time for the control. Given the lowest dMn concentrations in W1, it might be that dMn and not (only) dFe was limiting the production of reaction centres (Raven et al.; 19990), resulting in Chl a concentrations to not increase. Given the increased requirement for Mn under low Fe (Peers & Price; 2004), Fe addition may have relieved Mn limitation in the Fe addition treatments slightly, resulting in the observed increase of Chl a in those (F and TF) treatments."*
*Moreover, we will add the following on the Alderkamp et al. 2019 paper to line 675:*
*"In addition to higher light levels, the lower initial dFe concentrations in the Ross Sea study (Alderkamp et al. 2019) compared to our study indicate a stronger Fe limitation and subsequently a stronger response to dFe addition."*

-Line 680: The Fv/Fm results are minimally discussed, and do not provide much insight into the changes in the phytoplankton health together with both the temperature and iron changes and the confounding influence on the changing communities.

*As suggested by Reviewer 2, we will refer to the $F_v/F_m$ results more often throughout the Discussion, where they strengthen our discussion points (e.g. in lines 594 – 596 and line 658 of the original manuscript), rather than leaving them out. We will link Fv/Fm with sampling time in the productive season following line 661 (original manuscript). Moreover, we will refer to $F_v/F_m$ values to show that bioassay A2 may not have been limited by dFe concentrations (based on a comment by Reviewer 2 on line 677). We will also change the order of the Discussion on $F_v/F_m$ values to avoid switching between bioassays: "Indeed, given that the Weddell Sea bioassays were performed early in the productive season, these results imply more severe Fe limitation in the Weddell Sea whereas any Fe limitation in the Amundsen Sea likely only develops later in the season. Consistent with the lower dFe concentrations was the reduced in-situ $F_v/F_m$ of the phytoplankton in W1 and W2, which stayed low for non-Fe addition treatments throughout the experiments, as it is a common indicator of Fe stress in the Southern Ocean (Greene et al., 1992; Mills et al., 2012; Olson et al., 2000; Jabre and Bertrand, 2020). In addition, the low dMn concentration may have contributed to the low $F_v/F_m$ (Wu et al., 2019). The decrease in $F_v/F_m$ in the F and TF treatments towards the end of the Weddell Sea bioassays seem to indicate that the added Fe had depleted again to limiting conditions or that Mn became (co-)limiting." Following line 671, we will add (about bioassay A2): "The high initial $F_v/F_m$ values suggest that the phytoplankton may not have been limited by dFe (under these low light conditions) and would only require more dFe once light intensities increased again (Strzepek et al.; 2019, Vives et al.; 2022, Latour et al.; 2023). The small increase in $F_v/F_m$ in the Fe addition treatments may suggest growth became dFe limited during the incubation (Fe-addition did show a significant effect on $F_v/F_m$ at the last day of the incubations), despite the light conditions remaining low."*

Conclusion:
I struggle to clearly see all the concluding points made from this study, particularly in the latter half of the conclusion. The conclusion partly reads like a literature review and does not sufficiently highlight some of the main conclusions. This undermines the value based on the breadth of the experiments and outcomes of this study. I would suggest focusing on synthesizing key findings and clearly articulating the study's contributions and implications in the conclusion.
*We will move respective parts where fitting (Introduction and Discussion) or leave out. We will change the Conclusion section focusing only on our immediate results and on an outlook for future experiments. We will delete lines 733 – 735 and 766 – 773. Lines 754 – 759 will be moved to the Introduction. Lines 762 – 766 will be moved to the Discussion. The Conclusions now read (changed and/or added text is underlined): "Our study stands out in that it combined trace metal chemistry and biology, Chl a, and population abundance to examine co-effects using natural Antarctic phytoplankton communities at environmentally realistic Fe concentrations (+ 2 nM) and a predicted (2 °C) temperature increase (Boyd et al., 2015; Jabre et al., 2021; Andrew et al., 2022). Bioassay incubations were performed under trace metal clean conditions (for the entire duration) and with temperature remaining stable over the course of incubations (maximum fluctuation of temperature ± 0.3 °C). We stress the importance of trace metal clean working conditions to avoid inadvertently assigning Fe addition effects on phytoplankton to temperature when working in low Fe regions (i.e. Southern Ocean, but also open oceans in general). The differences we found between the F and TF treatment may have been assigned to temperature alone under non-trace metal clean working conditions (as Fe would inadvertently have been introduced), whilst our results show that temperature alone did not have a (major) effect. Our data also shows the importance of considering other regional and/or seasonal factors potentially limiting phytoplankton growth, such as e.g. light availability (limiting light conditions in bioassay A2) and dMn availability (potentially limiting in W1), when studying the effect of future climate on Southern Ocean phytoplankton. Additionally, our data indicates a trend of increased uptake of trace metals under dFe limitation, suggesting there are many adaptive strategies employed by phytoplankton in navigating nutrient scarcities under varying environmental conditions, with potential impact on the stoichiometry of global (micro-) nutrient distributions due to the central role of the Southern Ocean.*
*In general, the addition of dFe was the primary factor for observed stimulatory effects, with temperature enhancing the effect of dFe. Especially large diatoms benefitted from Fe addition, although several smaller-sized phytoplankton populations showed enhanced abundances upon Fe addition. Climate change is predicted to lead to a shift towards smaller phytoplankton (Deppeler & Davidson, 2017; Krumhardt et al., 2022). Our study shows, however, that enhanced Fe input counteracts this warming-induced shift, assuming macronutrients will not become limiting. Given that the intensity of the observed effects varied between the experiments with distinctly different phytoplankton communities, this study emphasizes the need for studying diverse regions of the Southern Ocean and performing multiple bioassays over the productive season to better understand and predict potential future changes, especially as future changes in Fe availability are region-specific (Tagliabue et al., 2016; Van Manen et al., 2022).*
*The Southern Ocean biogeochemical cycling and ecosystems dynamics are complex and need to be better studied in field and modelling studies. The current study underlines the need for assessing consequences of near future temperature changes at environmentally relevant dFe concentrations."*

-Line 753: The reference to Brookes and Crowe (2019) appears in the conclusion with the statement that dual treatments may affect the responses. However, this referenced statement does not seem to appear among the discussion points or even as an inference.
*We decided to take this part out of the manuscript, since it did not fit well after all.*

-Lines 754-759: These lines read as introduction sentences, rather than providing a strong context for the conclusion from the study: i.e. enhanced Fe input in such regions may *partly* overturn the warming-induced shift, given that macronutrients will not become limited.
Consider revising as this is not a compelling conclusion of the study.

*We will change the Conclusions to be more concise (please see our response to your previous comment on this).*

-Line 764: 'only will the flow of organic carbon through the food web be affected,'
*We will change the text accordingly.*

-Lines 767-773: Again, I feel that these literature points can be better contextualized to your actual results.
*We agree and changed the Conclusions (see reply to above comment).*

Data availability: It seems that one needs to have an account with the NIOZ dataverse to access the data presented in the manuscript. Will this be publicly accessible later on?
*Data should already be available using the password and username provided by the editors. However, data will be publicly accessible once the manuscript is accepted.*

Supplementary Information:
Generally, the panel sizes of all figures in the manuscript and supplementary could benefit from being slightly larger, so as to better see trends and the differences between the treatments or days.
*We will adapt the figures accordingly.*

Supplement Figure S1: This gives a nice overview of the physical setup. However, I struggled to fully comprehend and follow the details presented in the 'Bioassay set-up' in the supplementary text.
*We will go through the text and adapt where necessary.*

Supplement Figure S2: 3 Typos: '...the Amundsen Sea A1 (a, e, i), A2 (b, f, j) and the Weddell Sea…'
'The black dotted line represents the control (C) treatment, the red solid line the temperature (T) treatment, the blue solid line the iron (F) treatment, and the purple solid line the combined temperature and iron (TF) treatment.'
It is not very clear to see the 'black dotted line' from the panels without having to zoom in significantly. Are these supposed to be 'black solid line'? Perhaps you could change the scale/size of the y-axis to facilitate larger panels.
The purple line seems to blend too well with the red line. Consider using another contrasting colour like green or darker purple?
'Averages of triplicates with error bars represent the standard deviation'.
*We will change the figures accordingly.*

Supplement Figure S3: Again here, is the 'black dotted line' maybe meant to be 'black solid line'? Same comment regarding the purple and red solid lines blending.
*We will change the figures accordingly.*

Supplement Figure S4: Panel 'd' is missing brackets '(d)'.
*Thank you for pointing this out, will change the figures accordingly.*

'Average biovolume was calculated using total phytoplankton volume assuming spherical cells and dividing by total phytoplankton abundances.'
*Thank you for noticing the typos (missing brackets etc.). We will change the text and legend where necessary and will also increase figure size and change the colour of the TF treatment (currently purple) to either a different colour or a different shade of purple.*