

All referee comments are in bold and our answers in normal font. Changes in the manuscript text are kept in italics.

Referee #1

General comments:

This paper investigates the impacts of dust or iron additions on phytoplankton community with and without CO₂ enrichment in the northeast Pacific, which is known as a HNLC region. The experimental design is unique, and the incubation is conducted appropriately.

Reply: We thank the referee for this positive comment.

The authors present a novel dataset regarding the effects of dust addition and/or increased CO₂ on natural phytoplankton community in the HNLC area. However, the introduction and material and methods are insufficiently constructed to explain the importance of this work. For example, it is not clear from the manuscript why the authors use CJ-2 dust as Fe source. It would be better to cite previous works regarding Fe solubility and, if available, effects of dust addition on phytoplankton. The method of statistical analysis should also be described in sufficient detail with the threshold value (e.g., $p < 0.05$) to determine statistical significance. In addition, some of the discussions are lack of adequate referencing.

I recommend that the authors make a greater attempt to improve the manuscript. This paper can be accepted in the Biogeosciences after revision considering the comments below.

Specific comments:

Page 12284, lines 24-25. Is this statement correct? I believe that diatoms possess highly efficient CCMs than coccolithophores as described in the previous review by Reinfelder (2011, *Annu. Rev. Mar. Sci.*). Please revise this section.

Reply: The reviewer is right. The words were inverted.

The end of the paragraph, p 12284, starting line 24, has been changed.

New sentence: Coccolithophores and diatoms generally exhibit low- and high-efficiency CCMs, respectively (Reinfelder, 2011). Alternatively, fertilisation with FeSO₄ usually favors the growth of diatoms in HNLC waters (Boyd et al., 2007 and references therein). It is not known whether Fe-Dust deposition will favor diatoms or coccolithophores in the context of OA. OA could profoundly modify the structure and functioning of a phytoplankton community typically dominated by calcifying haptophytes.

Pages 12284-12285, Introduction. The topic of duct should be described and it should be separated from that of Fe, because it is unclear whether Asian duct might serve as a Fe source or not.

Reply: The following sentences have been added p 12284 starting line 9, to explain the importance of dust.

New sentence: *Sources of iron to the northeast subarctic Pacific include vertical mixing, eddies, tidal currents and convection (Cullen et al., 2009; Royer et al., 2010), volcanic ash (Mélánçon et al., 2014; Olgun et al., 2011), and desert dust (Boyd et al., 1998; Jickells et al., 2005). Dust, which is considered one of the most important sources, is deposited sporadically mostly in the spring during occasional dust storms originating from the deserts of northern Asia (Duce and Tindale, 1991). A natural strong dust deposition event has been shown to nearly double particulate organic carbon (POC) concentration in the northeast subarctic Pacific in 2001 (Bishop et al., 2002). The importance of eddies and vertical diffusion in the Gulf of Alaska was recently reviewed and found to be greater than previously thought (Crawford et al., 2007; Johnson et al., 2005; Lam and Bishop, 2008).*

Pages 12286-12287, Experimental setting and location. Please give the temperature and salinity in the sampling site.

Reply: The following sentence with the requested information was added p. 12286 starting line 23:

New sentence: *A CTD profile conducted at the same station 2h after all bags were filled showed a temperature of 13.5°C and a salinity of 32.6 at 10 m depth.*

Page 12288, lines 2-4. It is difficult to understand experimental conditions without the previous paper by Nishikawa et al. (2000) due to the lack of information about CJ-2 dust in the manuscript. Please describe the properties (e.g., size, chemical composition, and iron solubility) of CJ-2 dust in more detail in this section or discussion. Ooki et al. (2009, J. Geophys. Res. Atom.) might serve as a useful reference for the iron dissolution property.

Reply: The following sentence with additional information was added p. 12288 line 2:

New sentence: *Briefly, CJ-2 dust was collected from the Tengger desert surface soil, roughly sieved and blown through a wind tunnel designed to collect fine particles. Median diameter of CJ-2 dust is 24.1 µm. CJ-2 dust is characterized by a Fe content of 3.02 ± 0.12% and Fe solubility of 0.33% (Ooki et al., 2009).*

Page 12290, lines 19-21. In this sentence, the authors described that the 6 groups were quantified by CHEMTAX. However, table 3 shows 8 algal groups in the pigment ratio matrix for CHEMTAX analysis. Which is correct? In addition, the authors should provide the reference or method used to determine the initial pigment:chl a ratios in the table 3.

Reply: The matrix ratio used in the CHEMTAX analysis of the data is the same that is regularly used in northeast subarctic Pacific waters and was described in Royer et al. (2010). We agree that more information will be welcomed, especially considering the importance of speciation in the paper (see below). Regarding the question related to the algal groups, 8 were measured by the ratio matrix but 2 of them were below detection limit at P26, which resulted in the confusion between whether 6 or 8 groups have been identified. We are now clearly explaining

that 8 groups were measured but that prasinophytes and cryptophytes were below detection limit at our sampling station.

The end of the paragraph on page 12290 was changed (starting line 19).

New sentence:

The initial pigment ratio matrix loaded into the CHEMTAX program (Table 3) was obtained by averaging the minimum and maximum values of pigment : Chl a ratios given in Table 1 of Mackey et al. (1996) and is similar to that used by Suzuki et al. (2002) and Royer et al. (2010) for samples collected in the subarctic North Pacific. Eight algal groups were quantified using the chemotaxonomy program CHEMTAX (Mackey et al., 1996): cyanobacteria, pelagophytes, haptophytes (including coccolithophores), diatoms, dinoflagellates, prasinophytes, cryptophytes and chlorophytes. For a description of the pigment types, see Zapata et al. (2004).

And the following sentence was added as the second sentence of the Taxonomy section, p 12294:

New sentence: *Prasinophyte and cryptophyte biomarkers were not detectable during our experiments.*

Page 12292, Statistical analysis: The threshold value (e.g., $p < 0.05$) for determining statistical significance should be described in this section.

Reply: The following sentence was added to the section

New sentence: *The threshold value for determining statistical significance was $p < 0.05$.*

Page 12294, lines 14-16. “The highest POC concentration. . .” Is this statement indicates that the POC concentration in the Dust treatment was significantly higher than that of Dust+Acid treatment and the other treatments?

Reply: Yes. The point we want to make here is that POC concentrations were higher in the Dust treatment than in all other treatments, including the Dust+Acid treatment. The sentence was reworded to clarify our message

New sentence :

The highest POC concentration was measured in the Dust treatment ($287 \pm 30 \mu\text{g L}^{-1}$) and lowering the pH resulted in a 24% decrease in POC concentration at T4 (Dust+Acid treatment: $217 \pm 2 \mu\text{g L}^{-1}$) (Fig. 2c).

Page 12295, lines 20-21. “These results suggest a. . .” This statement would be described in the discussion.

Reply: It was deleted from the results sections as it is already referred to in the discussion p12300, lines 27-29.

Page 12295, lines 22-28. “Phytoplankton can acquire. . .is likely to be.” These statements seem better placed in the discussion.

Reply: As requested by referee #2, more details on what FeDFB uptake means in term of iron uptake was added to the method section 2.4.4. Consequently, the lines referred above were

removed from the manuscript as the information is given elsewhere in the new version of the manuscript.

Page 12297, line 25 (and elsewhere). It is better to use “ μatm ” instead of “ppm” for the unit used to express partial pressure.

Reply: Done

Page 12297, line 26. The period can be omitted.

Reply: Done.

Page 12297, lines 25-27. “The abrupt decrease. . .(Figs. 2a and 3c)” In this statement, why can the authors say that the community composition was unaffected by CO₂? Since the effects of acidification on the community composition were not described statistically in the results, it is hard to accept the statement. Rather, in the figure 3, chl a biomass of diatoms, haptophytes, and pelagophytes seems to have increased in the Control+Acid treatment relative to Control treatment.

Reply: These groups indeed look like they have a higher biomass, but due to a low number of replicates and poor statistical power, they were not significantly different at the threshold level of $p < 0.05$. We do agree that statistical results should be described in the results section, so we added the following sentence to the end of the taxonomy section in the results:

The only treatment to show a statistically significant difference with the Control is the Dust treatment, which had significantly higher concentrations of chl a attributable to diatoms and cyanobacteria than the Control at T4. No statistical difference between any treatment and its acidified counterpart could be detected.

Page 12298, line 11-14. In the discussion regarding the CO₂ effect on the HNLC water, it should be noted that the other experiment conducted using HNLC water demonstrated strong negative effects on diatoms at elevated CO₂ levels (Sugie et al. 2013, Biogeosciences; Endo et al. 2015, Biogeosciences).

Reply : The following sentence has been added to the paragraph :

Another experiment conducted with water from the diatom-dominated Fe-limited Bering Sea has shown a negative effect of elevated CO₂ on diatoms (Sugie et al., 2013, Endo et al., 2015), which was not observed in the Fe-enriched treatments.

Page 12299, lines 5-7. This statement requires supporting references regarding the response of DOC and/or TEP productions to ocean acidification. For example, Yoshimura et al. (2014, Deep Sea Res. I) reported minimal effects of increased CO₂ on DOC production in the Fe-limited phytoplankton communities.

Reply: This statement was supported by Riebesell et al., 2007 but we also added a reference to the paper of Engel et al. (2014) who also observed a stimulation of DOC production at elevated

CO₂. In addition, as suggested, we mention the paper by Yoshimura et al. (2014) in order to give a broader view of the types of responses observed so far.

New sentence: *Similar significant stimulation of DOC production at elevated CO₂ was reported by Engel et al. (2014) in coastal waters but only a weak and inconsistent CO₂ induced decrease in DOC production was observed by Yoshimura et al. (2014).*

Page 12300, lines 26-29. How did the authors make sure that phytoplankton growth was limited by Fe availability? I feel that the decrease in carbon fixation rate is insufficient to explain Fe limitation for phytoplankton without references. Given that Fe was released and re-adsorbed by the dust, DFe concentration might have remained constant by a balance between dissolution and re-adsorption. This paragraph needs to be reconsidered.

Reply: Decrease in the carbon fixation rate is only one of the supporting data for Fe-limitation. The high FeDFB uptake rates measured at that time represent a stronger indicator of Fe limitation. The paragraph has been changed to put more emphasis on the FeDFB uptake rate.

Modified paragraph:

Our results suggest that after a period of active growth, phytoplankton in the Dust treatment became Fe-limited 4 days into the experiment. This conclusion is supported by the very low absolute and chl a normalized C fixation rates in the Dust treatment as compared to the Control and FeSO₄ treatments at T4 (Fig. 4a), as well as by the FeDFB uptake rates normalized to C uptake rates which were higher in the Dust treatment than in the Control (Fig. 4b). Dust particles are known to efficiently adsorb Fe (Ye et al., 2011). The rapid return to Fe deficiency in the Dust treatment may thus result from a combination of increased Fe demand and re-adsorption of Fe onto dust particles. This explanation implies however that the re-adsorbed Fe becomes less prone to desorption, which needs to be demonstrated. These results suggest that the influence of the Fe released from dust lasted less than 4 days during our experiments.

Pages 12301-12302, “In order to further explore. . . was detected with the two factors”. These statements seem better placed in the material and methods or results.

Reply: This part of the text was moved to the results section and a new sentence was added to the paragraph from which the text was taken out to improve fluidity.

New sentence: *The two-factor ANOVA allowed us to increase the statistical power and detect a negative effect of acidification on chl a concentration after 4 days.*

Page 12302, lines 18-19. This statement is inconsistent with Fig. 3c, which indicates that chl a concentration of haptophytes increased in the Fe and Dust treatments relative to control treatment.

Reply: These increases are not statistically significant due to our small number of replicates. Therefore, we cannot state that there were more haptophytes in the Fe and Dust treatments.

The sentence has been reworded to:

This can be explained by the lack of significant difference in the abundance of DMSP-rich haptophytes in the Control, FeSO₄ and Dust treatments at T4.

Figures 1-5. Sample size should be described in the figure captions.

Reply: Done

References cited

Endo, H., Sugie, K., Yoshimura, T., and Suzuki, K: Effects of CO₂ and iron availability on *rbcl* gene expression in Bering Sea diatoms. *Biogeosciences*, 12, 2247-2259, 2015.

Ooki, A., Nishioka, J., Ono, T., and Noriki, S: Size dependence of iron solubility of Asian mineral dust particles. *J. Geophys. Res. Atmos.*, 114, D3202, 2009.

Reinfeldt, J. R.: Carbon concentrating mechanisms in eukaryotic marine phytoplankton, *Annu. Rev. Mar. Sci.*, 3, 291–315, 2011.

Sugie, K., Endo, H., Suzuki, K., Nishioka, J., Kiyosawa, H., and Yoshimura, T.: Synergistic effects of pCO₂ and iron availability on nutrient consumption ratio of the Bering Sea phytoplankton community, *Biogeosciences*, 10, 6309–6321, 2013.

Yoshimura, T., Sugie, K., Endo, H., Suzuki, K., Nishioka, J., and Ono, T.: Organic matter production response to CO₂ increase in open subarctic plankton communities: Comparison of six microcosm experiments under iron-limited and-enriched bloom conditions, *Deep-Sea Res. I*, 94, 1–14, 2014.

Added references

*Boyd, P. W., Jickells, T., Law, C. S., Blain, S., Boyle, E. A., Buesseler, K. O., Coale, K. H., Cullen, J. J., de Baar, H. J. W., Follows, M., Harvey, M., Lancelot, C., Lavoie, M., Owens, N. P. J., Pollard, R., Rivkin, R. B., Sarmiento, J., Schoemann, V., Smetacek, V., Takeda, S., Tsuda, A., Turner, S., and Watson, A. J.: Mesoscale iron enrichment experiments 1993-2005: Synthesis and future directions, *Science*, 315, 612-617, 2007.*

*Boyd, P. W., Wong, C. S., Merrill, J., Whitney, F., Snow, J., Harrison, P. J., and Gower, J.: Atmospheric iron supply and enhanced vertical carbon flux in the NE subarctic Pacific: Is there a connection?, *Global Biogeochemical Cycles*, 12, 429-441, 1998.*

*Crawford, W. R., Brickley, P. J., and Thomas, A. C.: Mesoscale eddies dominate surface phytoplankton in northern Gulf of Alaska, *Prog. Oceanogr.*, 75 (2), 287-303, 2007.*

*Cullen, J. T., Chong, M., and Ianson, D.: British Columbian continental shelf as a source of dissolved iron to the subarctic northeast Pacific Ocean, *Global Biogeochem. Cy.*, 23, 2009.*

Duce, R. A. and Tindale, N. W.: Atmospheric Transport of Iron and Its Deposition in the Ocean, *Limnol. Oceanogr.*, 36, 1715-1726, 1991.

Engel, A., Piontek, J., Grossart, H.-P., Riebesell, U., Schulz, K. G., and Sperling, M.: Impact of CO₂ enrichment on organic matter dynamics during nutrient induced coastal phytoplankton blooms, *J. Plankton Res.*, 36(3), 641-657, 2014.

Jickells, T. D., An, Z. S., Andersen, K. K., Baker, A. R., Bergametti, G., Brooks, N., Cao, J. J., Boyd, P. W., Duce, R. A., Hunter, K. A., Kawahata, H., Kubilay, N., laRoche, J., Liss, P. S., Mahowald, N., Prospero, J. M., Ridgwell, A. J., Tegen, I., and Torres, R.: Global iron connections between desert dust, ocean biogeochemistry, and climate, *Science*, 308, 67-71, 2005.

Lam, P. J. and Bishop, J. K. B.: The continental margin in a key source of iron to the HNLC North Pacific Ocean, *Geophys. Res. Lett.*, 35(2), 10.1029/2008GL033294, 2008.

Olgun, N., Duggen, S., Croot, P. L., Delmelle, P., Dietze, H., Schacht, U., Oskarsson, N., Siebe, C., Auer, A., and Garbe-Schoenberg, D.: Surface ocean iron fertilization: The role of airborne volcanic ash from subduction zone and hot spot volcanoes and related iron fluxes into the Pacific Ocean, *Global Biogeochem. Cy.*, 25, 2011.