

## ***Interactive comment on “Dynamics of phytoplankton community structure in the South China Sea in response to the East Asian aerosol input” by C. Guo et al.***

**Anonymous Referee #1**

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This manuscript by C. Guo et al. presents an interesting data-set from onboard grow-out incubation experiments conducted in the South China Sea to determine the phytoplankton growth responses to the additions of East Asian aerosol leachate. However, it is difficult to investigate the appropriateness of their attempt to mimic atmospheric aerosol deposition in the microcosm experiments for several reasons.

- The leaching procedure is a key to design this kind of experiments. No information was given by the authors regarding source of the seawater used for the leaching, time-length of the leaching, pH of the leachate, method for storage of the leachate, and background nutrients and trace-metals concentrations in the original seawater. If the

C2983

authors used natural seawater, concentration and characteristic of metal-binding organic ligands may affect biological availability of trace-metals in the leachate. Freezing of the aerosols before leaching may change the solubility of elements in the particles. Based on the data shown in Fig. 2, trace metals and nutrients concentrations in the leachate (4000 ml/5 ml = 800 times of the concentration shown in Fig. 2) could be micro-molar to sub-milli-molar levels. Such high concentrations in seawater could result in formation of metal (iron, etc.) colloidal particles and thus biological availability may change through adsorption and precipitation processes in the leachate. So application of the observed phytoplankton responses in the microcosm to natural atmospheric deposition event could be problematic.

- Trace-metal clean techniques should be applied to test the effect of trace-metals in the leachate. The authors used acid (10% HCl)-cleaned bottles for their incubation experiments, but it is not clear that they used trace-metal clean techniques through the experiments; from the collection of surface seawater and sample treatments, to the sub-sampling and parameter measurement. The authors should present initial and final concentrations of trace-metals in the control bottles to confirm that there was no serious contamination throughout the experiments, because discussions on iron-limitation are included in this paper. Stock solutions of inorganic nutrients used for the experiment 3 should be purified to remove trace-metal impurities.

- The corresponding amount of aerosols added in the experiments was 0.02 and 0.2  $\mu\text{g/L}$  ((70 mg/450 ml) $\times$ (0.5 or 5 ml/4000 ml)) for the low- and high-treatments. The authors should explain validity of these additions by comparing these values with the observed aerosol deposition fluxes in the South China Sea. Total amounts of added leachate were different between Exp. 1 (0.5 or 5 ml) and Exp. 2 (0.2 or 2 ml  $\times$ 4 = 0.8 or 8 ml), and it is difficult to compare the observed results directly.

- Although the authors measured picoplankton abundance and Fv/Fm every 24 hours, only the Fv/Fm values for PM7 and S412 experiments were shown. Without knowing daily changes in phytoplankton abundance and nutrient concentrations during the

C2984

experiments, evaluation based only on the results obtained at the end of the experiments (Day 3 or 4) could be misleading. It is possible that the phytoplankton in the low-treatments responded within 24 hours and then became nutrient-limitation again after Day 2.

#### Specific comments

P.6641; L.10 The "high-aerosol, low Chl" condition may suggest that the atmospheric nutrient supply is not enough for phytoplankton to grow or the hydrographic conditions such as light-limitation by deep mixing during winter playing an important role on the control of phytoplankton production.

P.6642; L.12 It is not clear why the authors used aerosol particles only smaller than 2.5  $\mu\text{m}$ . Most of the dust particles may be in the fraction larger than 2.5  $\mu\text{m}$ , and most of the aerosol samples would be anthropogenic origins.

P.6643; L.3 Material and cleaning procedure of the screen should be described.

P.6643; L.6-7 Please show the temperature range during the incubations and reasons for selecting 40% light intensity for the incubation.

P.6643; L.19-21 The concentrations of added inorganic nutrients were different from those supplied from the addition of the aerosol leachate, so that direct comparison of the yield of phytoplankton biomass is inappropriate.

P.6646; L.9 Soaking in DMF at  $-20^{\circ}\text{C}$  for 2 hour might not be enough to extract plant pigments effectively.

P.6647; L.17 Based on the data in Fig. 2, DIN concentration is about 2.5  $\mu\text{M}$  and phosphate is about 0.01  $\mu\text{M}$ , so that N/P ratio should be  $>200$ .

P. 6647; L.19- In the section 3.2, please explain possible reasons for the observed decrease in Chl concentration in controls bottles of the experiments S412 and A1.

P.6649; L.10-11 An increase in cell abundance in the low-treatments can only be seen

C2985

in *Synechococcus* at station PM7 and S412.

P.6649; L.17-19 An increase in abundance of picoeukaryotes is significant only in SEATs and S412 experiments.

P.6650; L.2-4 Abundance increased only in the high-treatments.

P.6650; L.9-11 It is not clear whether the authors counted dinoflagellates cell number only for autotrophic species or both autotrophic and heterotrophic species. Explanation for inconsistency between the decreasing trend in the dinoflagellates cell number and the increasing trend in the peridinin pigment concentration is needed.

P.6650; L.12 Initial abundance of the protist grazers are needed to confirm the increasing trend.

P.6650; L.23-25 The magnitudes of variability in Fv/Fm driven by changes in phytoplankton community structure often exceed that induced by nutrient limitation (Suggett et al. 2009, Mar Ecol Prog Ser, 376:1-19), and so the observed shift in dominant phytoplankton from pico-cyanobacteria to diatoms should be considered before interpretations of physiological status.

P.6651; L.2 Cellular Chl a and carbon contents are lower than what? Side scatter signal (Fig. 8-k, m and n) and red fluorescence (Fig. 9-k and n) showing significant increases in some experiments.

P.6652; L.5-6 In the experiment PM7, the observed increase in chl-a was 0.35  $\mu\text{g}$  for the addition of 0.2  $\mu\text{g/L}$  aerosol, and so the Chl/aerosol ratio should be much higher.

P.6652; L.13-15 Total amount of added DIN is different between the high-treatments and N+P/N+Si, and thus direct comparison of the final biomass is wrong. Discussion on iron-limitation does not have profound meaning without data on iron addition treatments and in situ dissolved iron concentrations. The reported dissolved iron concentration (0.2-0.3 nM) seems to be enough for oceanic phytoplankton species to grow with atmospheric N supplies.

C2986

P.6652-3; section 4.2 No statistically significant decrease in chl-a relative to the controls was observed in this study, instead significant increase in abundance of picophytoplankton was observed in some of the low treatments by FCM measurements. FCM data on cellular red fluorescence suggesting that decrease in cellular chl-a content of pico-eukaryotes was one of the reason for a slight decrease in total chl-a concentration.

P.6654; L.5-6 Reference is needed for this suggestion.

P.6654; L.9 It seems to be useful to calculate growth rates of phytoplankton species based on the microscopic enumeration data to examine the degree of growth enhancement by the aerosol leachate.

P.6654; L.11-24 Again, discussion on iron-limitation does not have profound meaning without data on iron addition treatments and in situ dissolved iron concentrations. The FCM data on cellular red fluorescence and side scatter signal shows that both *Prochlorococcus* and *Synechococcus* enhanced physiologically in the high-treatments.

P.6655; L.2 Trace-metal toxicity depend on their chemical speciation and the information about organic metal-binding ligands are key to evaluate the metal toxicity in the SCS.

P.6655; L.3 It is not clear what kind of the observed negative effect the authors are talking here.

P.6655; L.4-7 Is the difference in abundance between the high-aerosol and the N+P treatments is statistically significant? From Fig. 2, DIN concentration in the high-aerosol treatment (about 2.5  $\mu\text{M}$ ) seems to be higher than that of the N+P treatment (1  $\mu\text{M}$ ). FCM data on cellular red fluorescence showing physiological status in the high-treatment was better than the controls in the experiment 3. How about the effect of grazing on the *Prochlorococcus* abundance.

P.6655; L.23-27 The selective grazing should be happen in the control bottles, but there was no significant change in the cellular red fluorescence and side scatter signal in the

C2987

control.

P.6662; L.14 The manuscript in preparation should be deleted from the references.

Figs. 4-9 Initial values should be included in the figures.

Fig. 4 Please show the date of the sampling for each measurement. Data for PM7 is missing.

Figs. 5, 7, 9 and 10 It is not clear which panel showing the data obtained at 48 hour. Why the authors showing the data only for 48 and 96 hours?

Fig. 6 Is it true that all the data obtained at 96 hours? Data for A1 is missing.

Fig. 7 Why ciliate data for PM7 and A1 is missing? No measurements or abundance was low?

Fig. 10 Please show the data for C3a, SEATs and A1.

According to the text, Figs 8 and 9 should be Figs 9 and 10, and Fig. 10 should be Fig. 8.

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C2988