

## ***Interactive comment on “Distribution of ultraphytoplankton in the western part of the North Pacific subtropical gyre during a strong La Niña condition: relations with the hydrological conditions” by M. Girault et al.***

**Anonymous Referee #1**

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The authors presented, for the first time, the distribution patterns of ultraphytoplankton and heterotrophic prokaryotes in the subtropical western North Pacific during a La Niña condition. Although flow cytometry (FCM) has become a conventional analytical technique for determining the abundance of ultraplankton in seawater in the field of Oceanography since 1990s, the combination of their FCM and macronutrient data at nanomolar levels enabled them to examine the relationships between these parameters. Overall, this paper seems to be rather descriptive, but it could be publishable after revision - I have a few major concerns about their analytical procedures used in this

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study.

Major comments:

In terms of FCM, how many replicate samples did you analyze? Also, according to Campbell (2001), thousands of events for each population should be acquired for accuracy. In this study, the authors found that concentrations of nanocyanobacteria were very low ( $< 600$  cells  $\text{cm}^{-3}$ ) and the detection limit was 5 cells  $\text{cm}^{-3}$  (P5712, L22-28). If the authors consider that these data are statistically reliable, please indicate both the runtime of your FCM with its approximate flow rate and the method for determining the detection limit in the text. For nanocyanobacteria, how were you able to separate nanocyanobacteria from cryptophytes which could possess phycoerythrin? Why did you select the conversion factors of Tuit et al. (2004), Karayanni et al. (2005) and Caron et al. (1995) out of the literature values published previously? In P5707 and L24, SYBR Green II should be replaced by SYBR Green I, because the former is a stain for single stranded DNA and RNA and Marie et al. (1999) did not use SYBR Green II.

Campbell, L. (2001) Flow cytometric analysis of autotrophic picoplankton. In: Marine Microbiology (Ed., J. H. Paul), Methods in Microbiology, 30, 317-343, Academic Press.

The authors used principal component analysis (PCA), a simple, unconstrained ordination technique with one data matrix. PCA is a good statistical method to grasp the major structures of the whole data with a reduced set of orthogonal axes. However, PCA can hardly explain relationships between the parameters used (i.e., plankton and environmental variables in this case) - the authors tried to examine these, but the relationships obtained were qualitative. If the authors would like to compare their plankton data with the environmental variables, I believe they should use canonical ordination methods such as redundancy analysis (RDA), a popular statistical technique combining regression and PCA with multiple data matrices, and discuss the outputs. For example, Fehling et al. (2012) and Peng et al. (2012) succeeded in determining the

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environmental factors controlling the distribution patterns of each phytoplankton group using RDA.

Fehling, J. et al. (2012) The relationship between phytoplankton distribution and water column characteristics in North West European Shelf Sea Waters. PLoS ONE, 7, e34098, doi: 10.1371/journal.pone.0034098.

Peng, S. et al. (2012) Distribution and controlling factors of phytoplankton assemblages in a semi-enclosed bay during spring and summer. Marine Pollution Bulletin, 64, 941-908, doi: 10.1016/j.marpolbul.2012.03.004.

Minor comments:

P5703, L20-24: I do not agree on your statement that HPLC pigment analysis is impractical for estimating phytoplankton community structure in oligotrophic waters. It is true that cellular pigment content becomes low in such waters with high irradiance. However, this weakness can be retrievable by increasing the filtration volume of seawater. Please amend or delete the sentence.

P5703, L27 and P5724, L19: "a" after chlorophyll should be italicized.

P5704, L25 and thereafter: Remove the space between number and % (i.e., 1%).

P 5706, L14: because

P 5706, L15: was referred to

P 5706, L 16: were not shown

P5708. L5-19: What statistical software did you use?

P5709, L14: Please cite a more appropriate literature instead of the paleoceanographic paper of Oba and Murayama (2004), because they did not indicate the hydrographic characteristics of Kuroshio waters.

P5709, L14-15 and P5718, L3: the second group Subtropical gyre (stations 5-8)

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P5709, L18-19: the last group Transition zone

P5710, L18: nitrogen availability in diatoms (Fig. 7).

P5711, L13: Use station 8, not 22.83°N.

P5712, L24: Nanocyanobacteria were mainly

P5717, L10: modify salinity, especially

P5719, L16: Figs. 13 and 14

P5720, L2: zone was mostly

P5723, L16: diatoms

P5726, L12: be mainly controlled

Figs. 12-14: "Prochlorococcus" and "Synechococcus" should be italic.

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Interactive comment on Biogeosciences Discuss., 10, 5701, 2013.

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