**Interactive comment on “Heterotrophic prokaryote distribution along a 2300 km transect in the North Pacific subtropical gyre during strong La Niña conditions: relationship between distribution and hydrological conditions” by M. Girault et al.**

Anonymous Referee #2

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Review of manuscript “Heterotrophic prokaryote distribution along a 2300km transect in the North Pacific subtropical gyre during strong La Niña conditions: relationship between distribution and hydrological conditions” by M. Girault et al.

The authors explored the spatial distribution of heterotrophic prokaryotes along a north-south latitudinal transect (33°N - 12°N) crossing three different hydrographic areas (Kuroshio region, Subtropical gyre and transition zone). The biotic and abiotic parameters collected were used to investigate the relationships between the environmental parameters and the three prokaryotic populations (VHNA, HNA and LNA) distinguished by flow cytometry and nucleic acid staining according to their green fluorescence versus side scatter signature. Furthermore, the authors analyzed the results obtained using principal component (PCA) and redundancy analysis (RDA) in order to statistically identify the main parameters controlling the prokaryotic distribution. Finally, the authors showed a significant correlation between the hydrographic conditions and the prokaryotic communities distinguished by flow cytometry.

**Major Comments**

The manuscript presents a very interesting dataset in a poorly study area, however the data analysis needs to be substantially improved before publication. The statistical analyses presented do not allow to answer the main scientific question of the manuscript, i.e. “Which are the main controlling factors for the three prokaryotic populations along a north-south latitudinal transect characterized by different hydrographic conditions?” Furthermore, the discussion is often very descriptive and speculative, hence I strongly suggest the authors to refocus the manuscript pointing out the main findings according to the new results obtained. Finally, I find La Niña section not relevant for the manuscript, as there is no data available to prove any effect of La Niña on the distribution of the prokaryotic community.

The authors statistically analyzed the “phytoplankton-related variables (Chla and silicic acid)”, however; they never included the pico-phytoplankton (Prochlorococcus, Synechococcus and pico-eukaryotes) counts obtained by flow cytometry in the analyses. Thus, they did not use this data in the manuscript, although they mention to have it. I suggest the author to include this data in the next manuscript version.

In the manuscript the authors discussed the role of nutrients in the distribution of HNA and LNA populations. What about the VHNA population? Please include the VHNA population in the discussion. Instead of using the HNA/LNA ratio in your analyses you could use the relative contribution of the three prokaryotic populations to the bulk prokaryotic community.
Minor Comments

Page 15801, line 16-23. This sentence can be moved to the methods section.

Pages 15801-15802. Please add the standard deviation to the average concentration of LNA, HNA, VHNA populations.

Page 15802. Please consider using in this section the relative contribution of the three prokaryotic populations instead of the HNA/LNA ratio (figure 5).

Page 15805-15806. The paragraph has to be revised in a more concise way, the discussion on the role of silicic acid is too long and speculative.

Page 15806. Here for the first time the authors discussed about Synechococcus abundance in the Subtropical Gyre and in the Kuroshio regions, however this data is not presented at all in the results section. Please add more information about the pico-phytoplankton counts along the transect.

Pages 15807-15808. As I mentioned before I find La Niña section not relevant for the manuscript.

Pages 15809. So far it is not really clear what is the ecological role of the prokaryotic populations distinguished with the flow cytometer (HNA versus LNA) (Bouvier et al 2007 EM). Please comment on that in the manuscript.

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