

***Interactive comment on* “Factors limiting heterotrophic bacterial production in the southern Pacific Ocean” by F. Van Wambeke et al.**

Anonymous Referee #1

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General comments

This is an interesting manuscript reporting the roles of potential factors limiting bacterial growth along vertical and longitudinal gradients across the South Eastern Pacific Gyre. Overall, the manuscript is well written and the data are of high quality. The measurements appear to be of high quality and clearly presented. However, the statistical measurements are not clear enough. In several places the authors talk about one thing being higher (or less) than another, but no statistical test is given. In addition, some conclusions are not substantiated.

My suggestions for improvement of this work are shown below.

1. First, the manuscript lacks a description of the statistical methods used in their

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analyses. A detailed statistical analysis section needs to be included in the methods section. In addition, p values should be included in the results text wherever statistical analyses are mentioned (for example, page 3806, line 18). Also, the test that the authors use has a number of assumptions (normality in particular) that would make the test invalid if they were violated. The authors might want to comment on this as well and also assess the suitability of the test.

2. Secondly, for any statistical test involving rates (e.g. enrichment factor), the numbers should be arcsine transformed or logarithmic transformed. The authors do not say anything about this (e.g. Table 4; Fig. 4), so I assume they did not do the transformation.

3. I cannot understand how the authors can conclude that a labile organic carbon source could limit BP after relieving the N limitation in the GYR site from their TMC experiments (see below).

Specific comments

1) Page 3800, line 7: This sentence seems to be half-baked. “From the Marquesas plateau (8°W to approx 125°W), bacteria were not bottom-up controlled...”
8°W? Or 138°W?

2) Page 3802, line 1: “both autotrophic and photoautotrophic”? Do you mean “both heterotrophic and photoautotrophic”?

3) Page 3802, line 12-15: The authors should describe how to achieve the TMC condition. Or, relevant references should be given.

4) Page 3803, line 3-4: Since temperature can strongly influence heterotrophic bacterial growth, did you compare temperature in the on-deck incubator with in situ temperature when the bottles were incubated with “circulating surface seawater”?

5) Page 3804, line 1-3: The use of only leucine as an activity marker is possibly problematic. It is known from studies in several ecosystems that some bacteria are unable to take up leucine. A change in population could therefore be misinterpreted as a change in activity level. Have other substrates been tested?

6) Page 3805, line 17: “Fig. 2” should be inserted in the next sentence as a reference

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point.

7) Page 3805, line 21-22: The authors talk about one thing being less than another, but no statistical test is given. Also, this sentence is not in absolute agreement with Table 4. (e.g. chlorophyll increased similar to the primary production at the GYR and EGY sites after 24h.)

8) Page 3806, line 16: “There was no other additional effect after 24h”? It seems to should be “after 48h” according to the context?

9) Page 3808, line 18: The authors talk about one thing being higher than another, statistical test should be given (p-value).

10) Page 3809, line 10: Bacterial abundances appear to show a response only up to $\times 1.2$, not $\times 2$, in Table 4.

11) Page 3810, line 5: “(Table 4)”? It seems it should be “(Table 5)” according to the context?

12) Page 3811, line 16-17: I cannot understand how you can conclude that stimulation of bacterial production by nitrate + ammonium addition is direct. The factor of increase of leucine incorporation rate is not statistically significantly different from non-amended controls after the addition of N alone after 24h (in Table 4 and Fig. 2), although the magnitude of response of heterotrophic bacterial parameters seems to be higher than that of phytoplankton.

13) Page 3811, line 22-23: Also, it is difficult for me to understand how you can conclude that a labile organic carbon source could limit BP after relieving the N limitation from “N, FeN (Fe+N) and all (Fe+N+P) treatments stimulated exactly to the same degree phytoplankton properties, whereas the leucine incorporation rate was stimulated more progressively in these three treatments.” First of all, organic carbon (glucose) was not added in the above three treatments as a resource limiting factor. Secondly, there is no evidence indicating that progressively higher enrichment factors of leucine incorporation in these three treatments are due to progressively more labile organic carbon from the phytoplanktonic production, since phytoplanktonic parameters were

almost equal in the three treatments.

14) Page 3812, line 4: Is this sentence correct? “The rapidity of the **RESPONSE** of heterotrophic versus autotrophic **RESPONSE** after ...”?

15) Page 3812, line 10-11: “autotrophic process were also notably favoured in regard to respiration”? Do you mean “heterotrophic processes were also notably favoured in regard to respiration”?

16) Page 3813, line 9: Insert some appropriate references concerning the assumption of “20% efficiency”.

17) Page 3813, line 14: Insert some appropriate references regarding the UV radiation effects on transformation from labile DOC to refractory DOC.

18) Table 4: Were the ratios obtained using the MEAN values of triplicates? Please mention it in the legend.

19) Table 4, the legend: “Enrichment codes correspond to Table 2”?

20) Fig. 4: The authors should explain clearly how the graphs and the median and percentiles correspond, and what the error bars indicate (SD?).

Technical corrections

1) Page 3801, line 24: Use a “?” in “iii) direct versus cascade effects: are bacteria directly stimulated, or do they benefit from a surplus phytoplankton production also affected by the relieving of one key nutrient?”

2) Page 3804, line 19: ANOVA should be spelled out in full on first use.

3) Page 3804, line 26: Lack of a blank between “Chl *a*” and “varied”. Please use the same format for “Chl *a*” in the whole text.

4) Page 3805, line 3: “where”? Instead, “were”?

5) Page 3807, line 2: “ii)? Instead, “iii)?

6) Table 4, the legend: Double “Table 4”.

7) Fig. 2, the legend: “trace metal **CLEAN**”? “For GYR and **EGY**”?

8) Fig. 2: “HNL”?

9) Fig. 3: “T”? Or “C”?

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