

Review of revised manuscript by K. Leblanc et al., “Phytoplankton community structure in the VAHINE MESOCOSM experiment”

Leblanc and colleagues present a revised version of their manuscript on floristic results from a LNLC mesocosm experiment in Noumea designed to stimulate diazotrophy and follow the transfer of newly fixed N through the ecosystem. The original manuscript, though well-written and interesting, was missing key datasets and methods, which made it impossible to consider as a standalone paper. The authors have addressed those deficiencies adequately and I now recommend publication after minor revisions. I do not think another round of review is necessary; however, I ask the Editor to ensure that these remaining minor points are adequately considered by the authors:

Title: Shouldn't “MESOCOSM” be all lower case? It doesn't appear to be an acronym.
OK

Line 20: “phosphate” should be “phosphorus”. OK

Line 21: “Initially, the diazotrophic community was...”. OK

Line 23: Add comma before “which”. OK

Line 24: Delete comma before “that”. OK

Line 39: N₂ is “a” major source, not “the” major source of new N to the ocean. OK

Line 49: “diazotrophs” (plural). OK

Line 123: The glycerol uncoupling method for PE is an *in vivo* method, not an extractive method.

In answer to this comment + the one on line 232-238, the text in the method section was modified as follows :

“Water samples (4.5 L) were filtered onto 0.4 µm Nucleopore polycarbonate membrane filters (47 mm diameter) and immediately frozen in liquid nitrogen until analysis. In the laboratory, particles retained on the filter were resuspended in a 4 mL glycerol-phosphate mixture (50/50) after vigorous shaking, according to the *in vivo* method (Wyman, 1992). The PE fluorescence excitation spectra were recorded between 450 and 580 nm (emission fixed at 605 nm), using a Perkin Elmer LS55 spectrofluorometer and emission and excitation slit widths adjusted to 5 and 10 nm, respectively (Neveux et al. , 2009). As this method was developed for small *Synechococcus* cells, potential packaging effect could occur when measuring PE in larger cells such as *Trichodesmium*, but this remains to be documented. Estimates of phycoerythrin were obtained from the area below the fluorescence excitation curve, after filter blank subtraction. PE analyses were made only at 6 m-depth in the three mesocosms and in lagoon waters.

Line 182: What is “gentle” peristaltic pumping? Come on now...

OK “gentle” was removed

Line 208: Was the enrichment of the ¹⁵N stock assessed by MIMS? This point needs to be made clear. In practice, the measured enrichment is typically lower than that expected from calculations, due to air N₂ contamination during preparation.

Yes it was. Following text was added :

“The ¹⁵N enrichment of the N₂ pool was measured by Membrane Inlet Mass Spectrometer according to Kana et al. (1994) and was found to be 2.4 ± 0.2 atom%.”

Kana, T. M., Darkangelo, C., Hunt, M. D., Oldham, J. B., Bennett, G. E., and Cornwell, J. C.: Membrane Inlet Mass Spectrometer for Rapid High-Precision Determination of N₂, O₂, and Ar in Environmental Water Samples, *Analytical Chemistry*, 66 (23), 4166-4170, doi: 10.1021/ac00095a009, 1994.

Line 212: Superscript “15”. OK

Line 216: Need to state the chemical form of the added ¹⁴C. OK

Lines 232-238: The glycerol uncoupling method was developed for *Synechococcus* (very small cells); because it is not extractive, it is possible that there are packaging effects associated with larger PE containing organisms such as *Trichodesmium* and DDAs. To my knowledge, these potential artifacts have never been characterized. That is, the method is likely only semi-quantitative for the larger organisms. This point should be made here in the context of PE changes over time.

Line 242: “whose” not “which”. OK

Line 250: “In contrast to...” not “Contrary to...”. OK

Line 266: “nano-phytoeukaryote” (singular). OK

Line 266: Delete “comprised”. OK

Line 275: “whose” not “which”. OK

Line 349: “diazotroph” (singular). OK

Line 352: I don’t think it is correct to say that these rates were “among the highest ever reported”. Certainly many terrestrial and freshwater systems exhibit higher N₂ fixation rates, and as far as marine systems go I suspect in the Baltic, for example, rates can be extremely high. Perhaps these rates are among the highest ever reported from this lagoon. Be specific.

Following text was added “ever reported for oceanic systems”

Line 429: Add “of” before “DDN”. OK

Lines 449-453: Could the increase in *C. closterium* be due to a mesocosm wall effect? You do mention that they can be common in benthic environments.

Cylindrotheca closterium could in effect grow on cell wall, but similarly to all other pennates. Yet it is more likely that they benefited from the locally modified conditions and synchronous increase with UCYN-C. Cylindrotheca closterium is a most opportunistic diatom and often blooms in mesocosms / microcosms experiments. Even if it grew on cell walls (which was not verified under this experiment) it would still grow there because of beneficial bottom-up conditions in the water tanks, so I am not sure adding a comment without any proof either way would make this point any clearer.

Line 492: Change “clearly” to “likely”. OK

Lines 497-500: If you are going to tell me that “Clear differences” between the mesocosms and the

lagoon exist, you'll need to state the statistical test and P-value. Otherwise, use less strong wording.
OK

Figures 3, 12, 13 and 14: I'm not a fan of the funny European habit of using commas in place of decimal points; either way, you need to be consistent throughout. OK decimal points were corrected in the Figure file.