Biogeosciences Discuss.,
https://doi.org/10.5194/bg-2017-567-AC1, 2018

Biogeosciences Discussions

Interactive comment on "In depth characterization of diazotroph activity across the Western Tropical South Pacific hot spot of N_2 fixation" *by* Sophie Bonnet et al.

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Received and published: 2 May 2018

Response to Referee #1

We thank Reviewer #1 for his constructive comments. Below are copied the comments in regular font with our point by point responses below. Changes in the manuscript appear in 'track change' mode.

Bonnet and coauthors measured nitrogen fixation rates and diazotroph abundance along a west-east transect in the western tropical South Pacific Ocean. They report some astonishingly high rates along this transect and offer explanations for the driving

factors. This is a solid piece of work with some important and interesting findings. Most of my comments below are minor although there are a couple of major typographical errors that need to be fixed. But this manuscript can be improved to make it something more than a data report by providing a good oceanographic context to the observations reported. It seems very likely that the researchers encountered different water masses along the transect that account for some of the variances in nitrogen fixation rates reported and providing that context would be useful. I suggest not using acronyms when not necessary – the difference between GY and gyre is two letters and it just makes it easier to read. I also suggest some minor modifications to the figures to make them more useful.

I do have a couple of pet peeves to express and hope the authors will pay attention to at least the second and change the manuscript accordingly. 1) While I realize that this was a major oceanographic expedition with many groups, all working at different pace and thus necessarily, some results are available earlier and already published while others more recently processed, it is still frustrating to read a manuscript where critical bits of information are presented elsewhere, either already published, in review or in preparation. It is unfortunate that success in the modern scientific enterprise is measured by numbers of papers and careers of especially young scientists are determined by first authorships, resulting in piece meal papers. I don't expect the authors can do much about this but do wish to raise this issue because it is especially important for major interdisciplinary field expeditions such as OUTPACE.

I totally understand this comment and share this view in some ways. The OUTPACE special issue is divided in 27 papers, each dealing with a specific part of the 'story'. Since the submission of this manuscript, several of the cited papers are now available online on the special issue webpage https://www.biogeosciences.net/special_issue894.html, and some are accepted, which should improve accessibility. I acknowledge that a synthesis paper would be necessary to give a broad and multidisciplinary view of the ecosystem functioning in this region.

2) The word “hotspot” is starting to get overused. It would seem that each investigator’s favorite geography is a “hotspot” and I am having a difficult time with the concept of claiming a quarter or even one eighth of the largest ocean (western tropical South Pacific) as a hotspot. As the authors themselves say, “WTSP is a vast oceanic region” (page 2, line 21). The data to support the idea that the entire WTSP is a hotspot is still sparse and much too variable - 631286 in Melanesian archipelago waters - is a range of almost 45% in this cruise alone. The findings in this manuscript are significant even without that claim. In addition, there is one, potentially two real hotspots within this transect that are important in my view and that get lost when the claim is made to the whole area – I am both supportive and excited by the idea that there is a “sweet spot” (to use a different term) for diazotrophy at the interface where there is a supply of iron and phosphorus (around station LDB). This zonal gradient is similar to the meridional gradient in Fe and P that Moore et al characterized in the Equatorial Atlantic. But the South Pacific is more complex and thus interesting in that there is clearly some sort of island effect with higher rates closer to the islands as well as Fe supply from the seafloor.

We agree with this comment and made a more reasonable use of the term hot spot throughout the new version of the manuscript. In addition, we now discuss the possible origin of the high N2 fixation rates at LDB, in relation with physical parameters and nutrient inputs page 15 line 10-28.

Specific comments and suggestions: Page 2, line 5 – is it ammonia or amino acids?

It is first transformed into ammonia (see reaction below) then in amino acids N2 + 8 H+ + 8 e–+16ATP $\rightarrow$ 2 NH3 + H2 + 16 ADP + 16 Pi

Page 7, line 4 – should be per cell, not par cell

Yes, sorry, this has been fixed

Page 7, line 18 (and elsewhere) – it would be good to discuss what is special about

station LDB. This station is clearly a hotspot. Why? Was there any eddy activity here? Why is the water warmer here? Why is the chl higher all along the water column? Are we at the edge of water masses? Actually for that matter, what is going on at LDA where warmer waters are mixed down to almost 150 m.

We added a section regarding the bloom at LDB page 15, line 23-28 and also refer to a video showing the evolution of the origin of the bloom: ‘However, the huge surface bloom observed at LDB (Figure 1) and extensively studied by (de Verneil et al., 2017) was mainly sustained by N2 fixation (secondary fueling picoplankton and diatoms) (Caffin et al., in review, 2018)), rather than deep nutrient inputs (de Verneil et al., 2017). This bloom had been drifting eastwards for several months and initially originate from Fiji and Tonga archipelagoes (https://outpace.mio.univ-amu.fr/spip.php?article160), which may have provided sufficient Fe to alleviate limitation and triggered this exceptional diazotroph bloom as previously proposed by Shiozaki et al. (2014)’.

Page 8, line 2 – what is DL? If it is detection limit, what is it? Why is the range 0-4 in line 4 – i.e. why is this not from detection limit?

We acknowledge that DL was not defined in the submitted version. We have added in the Methods section the following sentence: ‘The minimum quantifiable rate (quantification limit, QL) calculated using standard propagation of errors via the observed variability between replicate samples measured according to Gradoville et al. (2017) was 0.035 nmol N L-1 d-1.’ We now refer to QL instead of DL in the Results section. Line 4 has been fixed as well.

Page 9, lines 34-38. This is a very important and interesting finding. While pressure could be one reason, clearly temperature would also play a role (although that would be in the opposite direction?) – i.e. there is a temperature gradient of 6-8 C between surface and 150m.

We acknowledge that there is temperature gradient with depth, but if temperature would

have played a role on dissolution, we would have had higher dissolution at depth (colder temperatures) compared to the surface, which was not observed. However, we thought that it was important to be mentioned and we have added a sentence as follows in the revised version: 'Despite the AN2 value was different according to the incubation mode, it did not change with the depth of incubation on the mooring line, indicating that a slightly higher pressure than atmospheric pressure (1.5 bar at 5 m depth) is enough to promote the 15N2 dissolution. It also indicates that the slightly lower seawater temperature (22-24°C) recorded at ∼100-180 m where the deepest samples were incubated likely did not affect the solubilization of the 15N2 gas'.

We partly agree with this comment. For samples collected above 50 m, the seawater temperature in the deck incubators and in situ was identical, so it seems that only pressure played a role in the higher dissolution. However, we acknowledge that seawater temperature is lower below 50 m and this may have enhanced the gas dissolution. Therefore, we have modified the text as follows: 'The seawater temperature checked regularly in the on-deck incubators was equivalent to ambient SST and likely did not explain the differences observed for samples collected above 50 m. However, we cannot exclude that the colder temperatures measured below 50 m-depth (∼23-26°C instead of 29-30°C in surface) may have, in addition to pressure, slightly enhanced the 15N2 gas dissolution, despite the AN2 value did not change with depth'.

Page 10, line 11 – suggest using "differing" or some other word such as changing rather than differed?

We changed by 'Despite the AN2 value was different according to the incubation mode'

Page 10, line 15 – what does under in-situ-simulated mean? Why not just on-deck incubations that simulated appropriate light levels?

We agree and changed the text accordingly

Page 10, line 21 – it would appear that there is quite a bit of variability even in the

archipelago waters. I am concerned that the contouring for figure 2 makes it appear as though it is a lot more uniform than it really is. While I understand the attraction of presenting the data along a linear transect this way, I do worry that real numbers are getting lost in this presentation and that the rates are actually a whole bunch more variable.

We now present all vertical profiles as supplementary information, clearly showing that rates are higher at some stations, which is also discussed in the new version of the manuscript.

Page 11, lines 3-22 – why only discussion of DIP – what about DOP? Trichodesmium can use DOP and it would have been interesting to see what was going on with that.

We have added a section regarding DOP in section 4.2: 'During the OUTPACE cruise, TDIPs were variable but were close or below two days in MA waters (Moutin et al., 2017), indicating a potential limitation by DIP at some stations. Trichodesmium, one of the major contributor to N2 fixation during the cruise, is known to synthesize hydrolytic enzymes in order to access the dissolved organic phosphorus pool (DOP) (Sohm and Capone, 2006). It is thus likely that DOP played a role in maintaining high Trichodesmium biomass in MA waters when TDIP locally dropped below two days as it was the case at station LD B for example (Moutin et al., 2017)'.

Page 11, lines 26-27 – what is the range for the DFe concentrations?

The range of DFe for MA waters was 0.2-66.2 nM and 0.2-0.6nM for GY waters. This has been added in the revised version page 14 lines 11-12.

Page 12, lines 15-16 – Is it not the other way around – PAR explains depth?

We changed the text as follows 'They were also negatively correlated with depth and logically positively correlated with PAR and seawater temperature, two parameters which are depth dependent'.

Page 12, line 35 – dominated not dominating

This has been fixed

Page 13, line 28 – suggest saying “more than” rather than above

I do not understand this comment

Page 14, line 18-19 – the sentence construction suggests that rates have gone up rather than our understand of rates have changed

Yes, we totally agree and have changed the sentence as follows: ‘The number of N2 fixation estimates have increased dramatically at the global scale over the past three decades (Luo et al., 2012)’.

Page 24, Table 3. It would seem that the table header for the second row is wrong. Spent a lot of time trying to figure out why the numbers were different till I figured out that it is actually for UCYN B rather than Trichodesmium

Thanks a lot, it was a mistake, it is fixed in the new version.

Page 25, Table 4. Why are the numbers for cDNA gene copies different from that reported in the text?

I am not sure to get this comment. The cDNA numbers are not reported in the text. However, the nifH copies per liter from the qPCR assay are given in Table 3 and in the text to give the context of the nanoSIMS studies. Those numbers are different of course but it is well specified in the text page 10 lines 30-31 that we are not talking about cDNA in this section but about quantification of nifH.

Page 27 Figure 1: Suggest improving 1a and show the ocean currents better – the superimposition of a big arrow does not do much. I am not clear how 1b was done – am just surprised that there are no clouds in the image. This is not critical expect to understand if the high chl patches seen are temporally relevant.

The currents have been redrawn on Figure 1a. Figure 1b is actually a quasi-Lagrangian weighted mean Chl map in which the satellite data are weighted in time by each pixel’s

distance from the ship’s average daily position for the entire cruise. As there were a large number of images due to the 45 days duration of the cruise, the resolution is nice despite there were clouds.

Page 28, Figure 2: Suggest adding the parameters to the various subfigures.

The parameters have been added

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