Review of Caffin *et al.*

General comments:
In their study, Caffin *et al*. use a Lagrangian approach to determine N sources (N_2 fixation rates, NO_3^- supply from vertical diffusion and N from atmospheric depositions) and sinks (particulate N export) at 3 stations in the Western South Tropical Ocean. They also measured primary production using ^{14}C tracer incubations. Their main findings are that 1) N_2 fixation is the major source of new N (>90%) at all stations, regardless of whether the diazotroph community was dominated by *Trichodesmium* or unicellular cyanobacteria, 2) carbon export relative to primary production is high in this region, and 3) the sum of N input in the photic zone exceeds particulate N export. Overall, this study is interesting and timely as this region has recently been identified as a N_2 fixation hotspot. However, I have some concerns. First, they report a low input from atmospheric depositions but did not consider atmospheric sources other than NO_3^- and NO_2^- (e.g., NH_4^+ DON, PON). They also measured relatively high N_2 fixation rates and I am wondering if they considered the possibility of a contamination of their ^{15}N-N_2 stock with ^{15}N-NO_3^- and NH_4^+, which would artificially increase their N_2 fixation rates, as recently reported by Dabundo *et al*. (2014). Some of the references cited (this issue) were unavailable on the *Biogeosciences Discussions* online forum at the time of this review, making it impossible to evaluate these parts of the manuscript. I was also a bit confused regarding the novelty of their dataset: were the <u>same</u> N_2 fixation rates, qPCR or any other data collected at the same stations during the OUTPACE cruise already published in previous studies? The authors should make a clear distinction of the new data contributed by their study versus the data already published elsewhere in other manuscripts in the special issue.

Specific comments:

Title
The title is a bit long and not focused on the main point of the study. I suggest changing for: "N_2 fixation as the dominant new N source in the Western Tropical South Pacific Ocean (OUTPACE cruise)"

Introduction
Page 2, line 15: Knapp *et al.* (2008) and Bourbonnais *et al.* (2009) also observed a low $\delta^{15}N$ of NO_3^- (relative to $\delta^{18}O$-NO_3^-) in surface waters in the western and eastern subtropical Atlantic Ocean, supporting the role of N_2-fixers in these regions.

Page 4, lines 6: What factors influence the distribution of *Trichodesmium* or UCYN? I believe temperature is an important factor (see Moisander *et al*., 2010). This point should be discussed a bit more.

Experimental procedures
Page 5, line 19: They only considered NO_3^- and NO_2^- when quantifying N atmospheric depositions. They should also consider NH_4^+ or organic nitrogen (particulate or dissolved). For instance, Cornell *et al*. (1995) estimated that organic nitrogen was a significant component of atmospheric N depositions even in remove marine regions.

Page 5, line 26: Did they check their commercial ^{15}N Eurisotop gas for possible contamination with ^{15}N-labeled dissolved inorganic nitrogen (NO_3^-, NO_2^- and NH_4^+)? Microbial assimilation of contaminant ^{15}N labeled dissolved inorganic nitrogen would artificially increase N_2 fixation rates. Dabundo *et al*. (2014) recently reported significant concentrations of ^{15}N contaminants in ^{15}N-labelled N_2 gas supplied by Sigma-Aldrich and Campro Scientific.

Page 6, lines 5-7: A better way to assess whether equilibration was complete would be to try different treatments in triplicate, i.e., shake the bottles for different times and intensity before the *in-situ* incubations.

Page 6, line 6: Add "incomplete" before equilibration.

Page 6, lines 7-10: I assume the 12 mL subsample was collected without contact with the atmosphere?

Page 6, lines 16-24: What is the detection limit for their N_2 fixation rates?

Page 7, line 21: Define UCYN-B, UCYN-A1, het-1 and het-2. Which bacteria are represented by these different groups?

Page 8, line 1: Were samples with qPCR reaction efficiency below 95% reported? Why not repeat analysis for these samples?

Results

Page 9, lines 24 to 27: How are these rates different from the one measured in Bonnet *et al*., 2017 (This issue). Are they the same rates as reported in Bonnet *et al*. (2017)? The Bonnet *et al.* paper was not yet available at the time of this review, making it impossible to effectively evaluate this part of the manuscript.

Page 11, line 11: Were *Trichodesmium* data for LD A (150 m) not available or below detection limit (as stated on page 10, line 32)?

Discussion

Page 11, lines 27-29: Perhaps atmospheric deposition measured during OUTPACE are low because they neglected contributions from organic nitrogen and NH_4^+. This possibility should be discussed (see my previous comment, page 5, line 19).

Page 12, lines 24-27: Again, it would be relevant to check for possible contamination of their $^{15}N_2$ Eurisotop stock by ^{15}N-labelled dissolved inorganic nitrogen (see Dabundo *et al*., 2014).

Page 14, line 22-23: The Berman-Frank paper was not submitted at the time of this review. Also, define PCD.

Page 16, lines 1-6: This paragraph is not clear. Do they mean the dead and live "swimmers" zooplankton were not distinguishable? Rewrite accordingly.

References

The following cited references were not accessible on the *Biogeosciences Discussion* online forum at the time of this review:
Berman-Frank *et al.* (This issue)
Bonnet *et al.* (This issue)
Bouruet-Aubertot *et al.* (This issue)
Caffin *et al.* (This issue)
Moutin *et al.* (This issue) – there is a Moutin *et al.* submitted but with a different title
Van Wambeke *et al.* (This issue)

Tables

Table 5: Include contributions from atmospheric depositions in this table.

Figures

Figure 3: Why PAR and DCM are decoupled at station LD B?

Technical considerations:

Review the manuscript for grammatical errors and typos. Here are a few examples:

Page 1, line 21: replace "Thanks to a Lagrangian…" for "Using a Lagrangian…".

Page 1, line 34: replace "while there contribution…" for "while their contribution…"

Page 9, line 13: replace for "… in-situ incubation method ranged from 0.0 – 19.3 nmol N L^{-1} d^{-1} …"

Page 10, line 11: replace by "was strongly influenced by the vertical diffusion coefficient".

Page 11, line11: replace "LDA" by "LD A".

Page 11, line 21: replace "whatever" for "regardless of", i.e., "… N_2 fixation was the major external source of N to the WTSP regardless of the degree of oligotrophy, …"

Page 12, lines 22-3: change for "… NO_3^- input by turbulence always represented a minor contribution to the N budget."

Page 13, line 16: replace for "… and a clear dominance of …"

Page 14, lines 9-10: Add a period after “in oligotrophic open ocean regions.” Start a new sentence with “To date, few qPCR *nifH* data from sediment traps are available…”.
Perhaps the proportion of dead versus live zooplankton could be estimated from the flask not filled with formaldehyde collected on the fifth day of sampling and used for diazotroph quantification.

Page 15, line 4: change for: “… in different oligotrophic regions of the ocean, for instance, the SPG …”

Additional references:

Cornell, S., Randell, A. and Jickells, T., 1995. Atmospheric inputs of dissolved organic nitrogen to the oceans. *Nature*, *376*(6537), pp.243-246.

Dabundo, R., Lehmann, M.F., Treibergs, L., Tobias, C.R., Altabet, M.A., Moisander, P.H. and Granger, J., 2014. The contamination of commercial $^{15}N_2$ gas stocks with ^{15}N–labeled nitrate and ammonium and consequences for nitrogen fixation measurements. *PloS one*, *9*(10), p.e110335.

Knapp, A.N., DiFiore, P.J., Deutsch, C., Sigman, D.M. and Lipschultz, F., 2008. Nitrate isotopic composition between Bermuda and Puerto Rico: Implications for N_2 fixation in the Atlantic Ocean. *Global Biogeochemical Cycles*, *22*(3).