#### **General comments**

This study used <sup>15</sup>N and <sup>13</sup>C labeling coupled with cell sorting by flow cytometry and nanoSIMS analyses to look at the transfer of <sup>15</sup>N-labeled diazotroph-derived N (DDN) to the dissolved pool and non-diazotrophs. The authors used a microcosm experiment to compare the transfer efficiency of <sup>15</sup>N-labeled DDN for different diazotroph groups: *Trichodesmium erythraeum, Crocosphaera watsonii* and *Cyanothece* sp.

I found these results extremely interesting and timely. They elucidate the fate of fixed N from N<sub>2</sub>-fixation as well as the specific role of different diazotrophic communities in the ocean. The paper is clear and well written. I recommend publication after minor revisions (see below).

# **Specific comments:**

## **Abstract:**

Page 3, line 28: "...heterotrophic bacteria followed phytoplankton...". Do they mean: "heterotrophic bacteria followed by phytoplankton"?

## Introduction

Page 5, line 8: Bourbonnais et al. (2009) also observed low  $\delta^{15}$ N-PON in sediment traps (most likely from  $N_2$  fixation in surface waters) in the subtropical northeast Atlantic and should also be cited here.

Page 5, line 16: I do not believe that UCYN-C (unicellular cyanobacterial <u>Group C</u>) is defined previously.

#### **Materials and methods:**

Page 9, line 12: I think a few lines should be added regarding the potential contamination of  $^{15}N_2$  gas by  $^{15}N$ -labeled  $NO_3$ ,  $NO_2$  and  $NH_4$ , that could lead to overestimation of  $N_2$  fixation rates, as reported by Dabundo et al. (2014). Although two batch syntheses of the Cambridge Isotopes gas were determined to contain only trace concentrations of  $^{15}N$   $NH_4$ ,  $NO_2$  and  $NO_3$ , I am curious to know if the authors verified the purity of the  $^{15}N_2$  gas used before their experiment.

Page 9, lines 18-19: How and how long was the bag (15N2 bubble) shaken?

Page 12, lines 19-21: <sup>15</sup>N depleted NO<sub>3</sub> (likely from N<sub>2</sub> fixation) was observed in the subtropical north Atlantic Ocean (see Knapp et al., 2008; Bourbonnais et al., 2009). Since nitrification can occur in the euphotic zone (Yool et al., 2007), it is thus possible for part of the labeled <sup>15</sup>N pool to be transferred to the NOx pool (particularly NO<sub>3</sub> ), which could then be rapidly assimilated. I agree that the contribution from NH<sub>4</sub> \* should be more

significant, and that this mechanism would be more important at lower irradiance deeper in the water column, but I think this point should, at least, be discussed.

#### **Results:**

Page 18, lines 21: why would the DDN be higher (at least double) in the control treatment?

#### Discussion and conclusions

Page 20, lines 18-26: This whole paragraph is a repetition of the introduction. I would remove.

Page 21, line 10: Can these rates be compared with the one in Garcia et al. (2007)?  $N_2$  fixation rates using methods prior to the one developed by Mohr et al. (2010) tend to be underestimation, whereas rates calculated with contaminated gas stocks (Dabundo et al., 2014) tend to be overestimation.

Page 23, lines 15-23: Can the authors explain what may cause the significant differences in DD<sup>15</sup>N transfer in the UCYN treatments observed in their study compared to Bonnet et al. (2015a)?

Page 24, lines 19-22: This is also in agreement with the observation of a recalcitrant DON pool by Knapp et al., 2005 and Bourbonnais et al. (2009) in the subtropical Atlantic, on the basis of the concentration of DON its  $\delta^{15}$ N in surface water.

Page 25, line 4: What were the [DON] and [NH<sub>4</sub><sup>+</sup>] concentrations prior to the incubations?

Page 26, starting line: Bonnet et al. (2015b) is currently in review, making it difficult to evaluate this part of the discussion. Please update.

## **Tables and Figures**

Table 2: DON, NH<sub>4</sub><sup>+</sup>, NO<sub>2</sub><sup>-</sup> and NO<sub>3</sub><sup>-</sup> concentrations should also be included in the table for the different treatments.

Figure 5. Please show significant linear regressions, with r<sup>2</sup> and p-value.

Figure 6: This figure is too small. The font for x- and y-axis should be increased as well as the size of the overall figure.

## **Technical corrections**

Page 5, line 4: replace the second "which" by "and".

Page 11, line 16: replace "chlrorphyll" by "chlorophyll"

Page 17, line 10: replace "to" by "the"

## **References:**

Bourbonnais, A., M. F. Lehmann, J. J. Waniek, and D. E. Schulz-Bull (2009), Nitrate isotope anomalies reflect N<sub>2</sub> fixation in the Azores Front region (subtropical NE Atlantic), *J. Geophys. Res.*, 114, C03003, doi:10.1029/2007JC004617.

Dabundo, R., M. F. Lehmann, L. Treibergs, C. R. Tobias, M. A. Altabet, P. H. Moisander, and J. Granger (2014), The contamination of commercial <sup>15</sup>N<sub>2</sub> gas stocks with <sup>15</sup>N-labeled nitrate and ammonium and consequences for nitrogen fixation measuremetns, PLOS one, 9, 10, e110335.

Knapp, A.N., P.J. DiFiore, C. Deutsch, D.M. Sigman, and F. Lipschultz (2008), Nitrate isotopic composition between Bermuda and Puerto Rico Implications for N<sub>2</sub> fixation in the Atlantic Ocean, *Global Biogeochem. Cy.*, 22, GB3014.

Yool, A., A. P. Martin, C. Fernández, and D. R. Clark (2007), The significance of nitrification for oceanic new production, *Nature*, 447, 999-1002.