

Review of the manuscript No. BG 2017-198, title “The effect of light on N<sub>2</sub> fixation and net nitrogen release of field *Trichodesmium*” by Lu et al., 2017

Lu et al. have made isotopically enriched on-board incubation experiments to understand the role of light on N<sub>2</sub> fixation. They further present estimates of nitrogen release from the diazotrophs and infer that light also plays a role on nitrogen release. Most of their findings are not new, have been known for some years now. The effect of light on nitrogen release seems interesting, but it is difficult to understand the way it's presented. I have provided several comments below that might improve the manuscript.

Major comments:

1. Diazotroph derived nitrogen (DDN) release increases with increase in light intensity at S0320 (Fig. 5 a) but there was no variation at the other two stations (Fig. 5b,c). However, % of the total NF release always decreases. On the other hand, N<sub>2</sub> fixation increases with increase in light intensity and saturates at some point (Fig. 4a). Put all these pieces of information together, it appears that it becomes difficult to say what role light play in DDN release. Diazotrophs would release N anyways, so what is the role light (they would release even if put them in absolute dark). Therefore, the discussion provided in the section 4.3 is not convincing.

2. Were all the incubations samples at each station taken from the same Niskin Bottle? I believe not because of bottle capacity. As we know the sample (especially when the cell abundance is high) in different Niskin bottles could not be homogeneous although taken from the same depth and the same CTD. If the cell abundances were different in different light incubations to start with, then the rates would be different because of cells and not because of light. So it would be helpful if authors provide the biogeochemical data (at least in supp info) for each Niskin that is used for different light incubations.

3. How does the “average” intensity of light estimated. Were the light measurements continuous or monitored n times during the day?

4. While changing the light conditions, some density filters were used. Was the wavelength, which these filters block, was also estimated? Do they block the same fraction wavelength for all wavelength?

5. POC:PON ratio could be close to the Redfield ratio but the Carbon uptake:N<sub>2</sub> fixation ratios (Fig. 4b) are surprising. CF:NF ratios can be upto three order magnitude higher even in tricho bloom conditions, where highest N<sub>2</sub> fixation rates were measured (Gandhi et al., 2011). In not very active N fixation regions, this ratio could be even higher. This is simply because most of photoautotroph fix C but not all can N<sub>2</sub>. So C fixation to N (NO<sub>3</sub>+NH<sub>4</sub>+Urea+N<sub>2</sub> fixation) uptake ratios would be close to Redfield. I

would suggest the authors to look for the hypotheses presented on page 11 (lines 18-23) and explanations at several other places.

6. Provide an estimate of fraction of released DON and released inorganic N (ammonium) uptake by non-diazotrophs.

#### Minor Comments:

Page 1: Title should be revised as “field Trichodesmium” reads a bit awkward. I would suggest: “The effect of light on N<sub>2</sub> fixation and net nitrogen release in a field study”

Page 3, line 8: Light is an ultimate source of energy for everything not only to photoautotrophs. Revised this sentence.

Page 4, line 9: Not most but only some NF rates have used <sup>15</sup>N techniques, most have used Acetylene reduction assay, see Table 3 in (Singh et al., 2013), Table 5 in (Capone et al., 2005) and Table 4 in (Benavides and Voss, 2015).

Line 10: “The <sup>15</sup>N.....into account”. <sup>15</sup>N enrichment is taken into account as can be seen the equation (6) in (Montoya et al., 1996): the AN<sub>2</sub> takes care of the enrichment. I think authors mean the released <sup>15</sup>N-TDN during the incubation is not taken into the account and hence the underestimation.

Line 16-17: Contribution of N<sub>2</sub> fixation to export production can be upto 92% during Trichodesmium bloom (Gandhi et al., 2011; Kumar et al., 2017)

Line 21: “reports” should be replaced by “has reported”

Page 5: Line 5: Could 4000 cells/L be called bloom?

Line 16: Were the nutrient samples filtered through 0.2 µm filter? Were these measured at both the start and end of the incubations?

Line 22 and elsewhere: Reference format should be same throughout

Page 6, line 1: were should be replaced by was

Line 2: delete a

Line 5: put space after 40

Line 11: micron symbol throughout should be used rather than u

Line 22: (Mohr et al., 2010) is the original reference

Line 22: Were incubation done single, duplicates or triplicates?

Page 7, line 4: Were <sup>13</sup>C and <sup>15</sup>N<sub>2</sub> added in the same bottles?

Line 7: Perhaps 0.7 µm pore size should be mentioned.

Line 8: It would be a surprise if the authors were able to filter 4.5 L water on single 25 mm GFF filter?

Line 19: There is no  $I_k$  in equation (1)

Page 7, line 12: replace classical by typical

Line 14: How the average value of PAR calculated?

Line 18: 34.6 salinity is not really high. It is normal in open oceans

Line 23: "thus .....in all the experiments" can be deleted as preceding part of the sentence implicitly states the same.

Page 9, line 2: how many samples were taken to obtain the standard deviation and mean

Line 3: "two order of magnitude" is not quite true.

Line 4: "Detail" should be replaced by "detailed"

Line 6: biomass should be replaced by abundance or cells

Line 18: If at  $t=0$ , POC was same in all the light experiments, then how does POC decrease with light within 24 hrs so rapidly. With this logic, POC concentration will be drastically different during the evening and in the morning in the ocean.

Page 10, line 8: replace "was decreasing" by "decreased"

Line 12: Define this mentioned fraction. Is it the ratio of  $^{15}\text{N}$  TDN uptake by non-diazotrophs and total production of  $^{15}\text{N}$  TDN by diazotrophs. Or is it the ratio of  $^{15}\text{N}$  TDN uptake by non-diazotrophs and total ( $^{15}\text{N} + ^{14}\text{N}$ ) uptake by non-diazotrophs.

Line 19: "locates" does not read properly. Revise the sentence.

Line 21: Replace "strong of" by "strong"

Line 24: (Gandhi et al., 2011; Kumar et al., 2017) could also be proper citations here.

Page 11, line 2: 15-40 m is confusing here. Does it mean Trichos are more abundance in 15-40 m compared to that in 1-15 m?

Line 14: this section (including the hypothesis presented) should be revised as suggested in the major comments.

Page 1, line 6: "sever" should be replaced by "severe"

Lines 9-13: Not clear what the authors want to convey in this sentence

Page 13, line 16: (Montoya et al., 1996) is the original reference

Line 16: Why do the used a different technique may lead to higher DDN release?

Page 14, line 4: Replace "recently" by "recent"

Table 1: Also provide P\* values (as expressed by (Deutsch et al., 2007)) in a column

Table 2: Also provide the fraction of diazotrophic biomass to the total phytoplankton biomass

Fig. 1: Why is there so much fluctuations (variation) within minutes in PAR values? Were the conditions cloudy during incubations?

Fig. 4: Either use CF/NF or PP/NF. Be consistent.

Supp Table 1: Normally enrichment in  $^{13}\text{C}$  is much more than  $^{15}\text{N}$ . How much was  $^{13}\text{C}$  added, and how much would the approximate theoretical  $^{13}\text{C}$  enrichment at  $t=0$ ?

#### References:

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- Deutsch, C., Sarmiento, J.L., Sigman, D.M., Gruber, N., Dunne, J.P., 2007. Spatial coupling of nitrogen inputs and losses in the ocean. *Nature* 445, 163–167.
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- Singh, A., Lomas, M., Bates, N., 2013. Revisiting  $\text{N}_2$  fixation in the North Atlantic Ocean: Significance of deviations from the Redfield Ratio, atmospheric deposition and climate variability. *Deep Sea Res. Part II Top. Stud. Oceanogr.* 93, 148–158.