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Interactive comment on "Inhibition of nitrogenase by oxygen in marine cyanobacteria controls the global nitrogen and oxygen cycles" *by* I. Berman-Frank et al.

Anonymous Referee #3

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This manuscript is essentially a rehash of previously-published discussions about the "oxygen problem" as a constraint on N2 fixing potential of many diazotrophs, including heterocystous and non-heterocystous cyanobacteria, i.e. Trichodesmium. The explanations for and arguments pertaining to this problem have been made many times before, and I might add, in much more through and well-supported ways. There is nothing new here, except for maybe the rewording of somewhat ironic (pardon the pun) paradox that photosynthetic oxygen evolution (which cyanobacteria "invented") has turned out to be a contraint on the extent and magnitude of N2 fixing potential among the cyanobacteria. This has been more thoroughly articulated by others, including Gallon (1992), Paerl (1990) and Paerl and Zehr (2000). In fact, Paerl (1994) experimentally showed the incompatibility of these processes at high rates of photosynthesis (i.e. O2

evolution) in Trichodesmium 101 well over 10 years ago.

While some interesting molecular response mechanisms to varying ambient O2 levels were discussed, these have also been presented and discussed far more comprehensively and thoroughly by Zehr et al. (1997). Again, there's nothing new of major significance presented in the current manuscript.

Lastly, all the evolutionary arguments presented here have been made before (see the references cited below).

In summary, this manuscript adds little, if anything, to our understanding of the physiological, ecological, evolutionary and biogeochemical implications of the "oxygen problem". In addition, the authors left out key manuscripts and chapters that previously presented and discussed this issue. I do not recommend publication or for that matter further discussion of this manuscript.

Some specific comments on the manuscript:

P. 262, Abstract. First sentence: Where is the evidence that "N2 fixation supplies the vast majority of biologically accessible inorganic nitrogen to nutrient-poor aquatic ecosystems"? This seems like a gross and unsupportable generalization. There are certain regions of the worlds oceans where N2 fixation is a significant new N source (i.e. Baltic Sea, parts of the Gulf Stream and Kurishio Current), but these are only a fraction of the Earth's "nutrient-poor aquatic ecosystems".

P. 262, Introduction, line 1. The statement that "Only a small fraction of prokaryotic organisms from the bacterial and archaeal domains can procure and utilize atmospheric nitrogen by reducing it to ammonia" is naïve and incorrect. The process of N2 fixation is actually fairly broadly distributed among different phylogenetic and physiological groups. In fact, this is probably one reason why the genes encoding for it are so highly conserved in the first place.

P. 263, lines 16 and 17: the part of this sentence "restrictions on nitrogen" makes no

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sense. Something appears to be missing here. Also in line 18, it isn't clear what "real-world" means (as opposed to "unreal world"?). Lines 23-24: Trichodesmium 101 was initially isolated from North Carolina Atlantic coastal waters (see Prufert-Bebout et al. 1993). It is not known whether this strain "contributes significantly to nitrogen fixation in the tropical and subtropical oceans". This statement is one of numerous examples of gross overgeneralization and unsubstantiated armwaiving that seem to characterize this manuscript. These types of statements are both factually incorrect and add little relevant information.

P. 267, line 6: What does "chronically crippled nitrogenase" mean? Nitrogenase could be functioning at sub-optimal rates due to several (interactive) limiting factors over moderate to long time scales (i.e. days to weeks) and this may not be linked to the O2 problem. Fe and/or P limitation may be operating over such time scales, or high degrees of mixing and smaller scale turbulence may lead to sub-optimal rates of N2 fixation.

Line 22: What does "efficiency" mean here? This may not be an "inefficiency" problem at all, if we consider the interactive effects of multiple potential limiting factors (i.e. P, Fe, organic C, turbulence).

Additional references of relevance (not cited in manuscript).

Paerl, H. W. 1990. Physiological ecology and regulation of N2 fixation in natural waters. Adv. Microb. Ecol. 11:305 344.

Paerl, H. W. 1994. Spatial segregation of CO2 fixation in Trichodesmium sp.: Linkage to N2 fixation potential. J. Phycol. 30:790 799.

Paerl, H. W. and J. P. Zehr. 2000. Nitrogen Fixation. Pp. 387-426., In, D. Kirchman [Ed.], Microbial Ecology of the Oceans, Academic Press, New York.

Prufert Bebout, L. E., H. W. Paerl and C. Lassen. 1993. Growth, nitrogen fixation and spectral attenuation in cultivated Trichodesmium. Appl. Environ. Microbiol. 59:1350 1359.

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