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Interactive comment on “Influence of elevated CO₂ concentrations on cell division and nitrogen fixation rates in the bloom-forming cyanobacterium *Nodularia spumigena*” by J. Czerny et al.

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This is a well-written short communication that makes the important point that not all diazotrophic cyanobacteria will react to increasing pCO₂ with increases in nitrogen fixation and growth rates. In fact, the response of this important bloom forming species from the Baltic Sea is just the opposite, with decreased cell division and N₂ fixation rates as pCO₂ goes up. The fact that all N₂-fixing cyanobacteria won't experience the same “CO₂ fertilization” effect that has been reported for *Trichodesmium* and *Crocospaera* shouldn't really be a big surprise, in view of the diversity of CO₂

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responses that have been documented for various eukaryotic algal groups and for non-diazotrophic picocyanobacteria. Nevertheless, this is an important point to emphasize, and the paper therefore makes a significant contribution to the growing literature in this field. Here are my specific comments: Abstract: The statement that the surface ocean absorbs $\frac{1}{4}$ of current CO₂ emissions is not referenced (naturally, since references aren't typically included in an abstract), but to my knowledge this is still a relatively uncertain number and somewhat controversial. Maybe it would be better to start out by simply saying "As CO₂ emitted to the atmosphere from human activities dissolves in seawater, it reacts..."? Introduction on page 4282: Isn't Nodularia just one member of a multiple species cyanobacterial consortium that blooms in the Baltic? For readers not intimately acquainted with Baltic cyanobacterial blooms, perhaps this could be mentioned here, along with some of the other co-occurring groups (Anabaena is mentioned in this regard in the discussion). Is Nodularia spumigena the dominant one most of the time? This paragraph implies this but doesn't come right out and say it. Page 4283: The reason that the cultures were grown in a manner intended to avoid aggregation is well described and justified. Clearly though, as they imply here and in the discussion, a dense surface aggregation might have a completely different response to changing pCO₂ due to "microclimate effects". Since both aggregated and dispersed growth seem to be features of this organism's life cycle, it would be especially interesting to do the same experiments with both. At any rate, it is obviously necessary to qualify all of the results obtained here as applying specifically to homogeneously mixed cells, and recognize that the story for aggregated surface blooms might be quite different. M&M, page 4283: For a species that often blooms right at the surface, 85 $\mu\text{mol photons m}^{-2} \text{ sec}^{-1}$ seems like a fairly low irradiance. Is anything known about saturating light levels for growth of this isolate, and is it possible that the cultures were light-limited to a greater or lesser degree? M&M, page 4284: The authors chose to manipulate pCO₂ using acid/base additions rather than bubbling. This is fine, but this text says that TALK was measured at the start of the experiment, whereas the data and legend in Table 1 say it was measured at the end but not the beginning. This should be clarified, and

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perhaps something included in the text to specifically recognize that TALK in acid manipulated seawater does not realistically mimic TALK in gas-equilibrated seawater. It is also not completely clear whether any attempt was made to maintain the initial target pCO₂ levels through further acid additions during the 7 day growth period of the experiment. It seems that perhaps this was not the case, and pCO₂ was allowed to vary as the culture grew, but this is not entirely obvious from the text here. Results, page 4288: A minor comment- I would suggest that the chlorophyll a results in Fig 1b be referred to on the previous page, when presenting the rest of figure 1, rather than here. Results, page 4288: The rates of cellular carbon and production calculated from cell quotas and growth rates need to be qualified as being net production rates. Calculations from changes in cell number can't account for carbon lost to respiration and exudation, of course. Results, page 4288: If cell carbon and phosphorus quotas increase by a third or so at high pCO₂, but cell volume is unchanged, doesn't this imply a quite substantial increase in cellular density? How else can you have cells of the same size, but containing a lot more C and P? This result is an odd one- are there precedents in the literature for this? Maybe this puzzling observation deserves some consideration in the discussion section. Discussion, page 4290: The text here says the "accumulation of cellular nitrogen was less pronounced". Actually, there was no significant increase in the cellular N quota at all, correct? Discussion, page 4290: I like the explanation that this stoichiometry effect could be due to reduced N transfer from the heterocysts to the vegetative cells, and a good case for the possible pH sensitivity of this process is made on the next page. However, I can think of another possible explanation for increases in C and P but not in N in the cells. Could there have been an enhanced loss of fixed N as exuded material (ammonium or possibly DON) at high pCO₂? Some of our results from Hutchins et al 2007 suggested this for Trichodesmium, at least indirectly through comparisons of N fixation measured by acetylene reduction ("gross" rates) and ¹⁵N assimilation ("net" rates). This seems like a possible alternate explanation to the hypothesis presented here about altered N transfer from heterocysts. Discussion, page 4292: Again, this whole extended discussion about the significance of aggregation

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makes it apparent that a study comparing CO₂ effects on aggregated and dispersed cells is needed to really ascertain the overall ecological and environmental implications of their results. Discussion, page 4294: The obvious question arises, are the trends observed in this study general among heterocystous cyanobacteria? Apparently the authors have a paper in prep on *Anabaena*, of which they say somewhat obscurely “there will be a different reaction to rising CO₂”. Without stealing the whole story from this upcoming paper, can’t the authors here come out and say whether *Anabaena* is stimulated by higher pCO₂ or not? It is not too useful to readers to simply imply coyly that there is another response among similar species, without even generally indicating what it is. If *Anabaena* does show a different (positive?) reaction to increasing pCO₂, doesn’t this potentially argue against the pH effects on heterocyst/vegetative N transfer model they discuss extensively earlier in the text? If this model is correct, wouldn’t all heterocystous cyanobacteria exhibit the same response?

In general, my comments are fairly minor and the paper is in good shape for final publication with only relatively minor revisions.

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