Biogeosciences Discuss., 6, C1099–C1103, 2009
www.biogeosciences-discuss.net/6/C1099/2009/

Biogeosciences Discussions

Interactive comment on "Influence of elevated CO_2 concentrations on cell division and nitrogen fixation rates in the bloom-forming cyanobacterium *Nodularia spumigena*" *by* J. Czerny et al.

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Received and published: 9 July 2009

Thank you very much for the many constructive remarks on our manuscript that helped us to improve it substantially.

1. P4281 L15-16: the references cited are scarce and several other papers could be cited both for cultures and natural communities.

We added a review paper on CO2 effects on cultured microalgae by Rost et al. (2008) and an interesting study by Tortell et al. (2002b) on carbon uptake mechanisms in

natural plankton assemblages.

2. Do Nodularia blooms occur in other forms than dense aggregates? If not, can the authors reconcile their statement that CO2/O2 conditions within these dense blooms might significantly alter CO2 related physiological effects with their experimental results based on unaggregated filaments?

Nodularia is found in the Baltic Sea only during the summer month, while it rests in the form of special spores called akinetes during winter. Single cells evolving from akinets in the next summer become filaments that accumulate at sufficient density to form aggregates. The most carbon and nitrogen production probably happens during blooms when biomasses are high and aggregates dominate the picture. In this study we found a strong effect of pH on the physiology of dispersed Nodularia filaments. Microprobe measurements inside natural Nodularia aggregations by Helle Ploug (1994), on the other hand, showed strong diurnal pH variations ranging from 7 in darkness to over 9 during illumination. Therefore, we explicitly qualify our results as being valid for single filaments during a pre-bloom situation. We can not make any statement on weather there is an effect of seawater pH on growth, carbon or nitrogen fixation in aggregated Nodularia nor which direction it could have.

3. -Please specify the volume in which the semi-continuous cultures were grown in?

The cultures (and pre-cultures) were grown in 1 L Schott-Duran glass bottles, which contain about 1.25 L water when they are filled without headspace. The exact volume is now specified in the revised version of the manuscript.

4. -The authors chose to perform acid addition for CO2 control of their experiments. Two papers were recently published in the same journal exposing the potential issues with this method (Shi et al. 2009, Gattuso et al. 2009). The authors might want to justify their choice of CO2 control as this has been a controversial issue lately.

The choice of the TAlk approach for CO2 manipulation is now justified in the

manuscript. We decided to cite Schulz et al. (2009) as it best reflects our opinion to this controversial issue.

5. P4283 L 25: this sentence is a bit ackward in its formulation and I don't understand how the pre-acclimatation was done precisely. Please make this section a bit more explicit.

The sentence was rephrased in the revised version of the manuscript.

6. P4284 L17: TAlk and DIC were measured after filtration of the media: Is filtration and air bubbles not a problem for DIC measurements?

The filtration was done bubble-free by means of a filter cartridge and a peristaltic pump. Bottles were filled from the bottom to the top in the manner that seawater samples for gas measurements are taken. A total of about 30 bottles were filled without headspace of which 20 were used for the experiment and the pre-cultures and some of the rest were used for TAlk and DIC measurements. The results of all bottles measured were remarkably consistent. A section describing the filtration technique and the DIC and TAlk measurement at the beginning is now included in the revised version of the manuscript.

7. P4287 L21: why assume that the data is normal? There are some very simple tests to run to check for this, some are even available on line. I suggest the authors verify this assertion with the appropriate normality test.

Now data were tested for normal distribution with the Kolmogorov-Smirnov test. They are all normally distributed. The sentence was erased from the manuscript.

8. I found the discussion overall very interesting and my main concern is the justification for experiments on mixed unaggregated filaments. At the end of the discussion the authors conclude that the adverse effect of CO2 on cell division might delay the formation of the large surface bloom of Nodularia, which may be in turn outcompeted by other species. But this comes a bit late as the reader wonders throughout the article

whether this experiment can be extrapolated to in situ conditions. I would state in the introduction (P4283, first paragraph) more clearly that this experiment allows to investigate pre-bloom conditions of Nodularia, when filaments are scarce and unaggregated.

We tried to clarify that the results can be applied to dispersed filaments in a well mixed pre-bloom situation. The pH/[CO2] related physiology of Nodularia, however, is described quite well by the results of this study as the setup provides very constant controlled conditions for the cells. With the same physiological equipment Nodularia has to meet the strongly varying chemical demands inside aggregations. We agree that a cross check of this study using a setup where filaments are allowed to aggregate appears to be the logical next step to assess the full ecological relevance of our results. However, for reasons outlined below a culture experiment of this kind is likely to deliver ambiguous results that may prove difficult to be extrapolated to in situ conditions. A: The quantitative sampling methods used here are prone to errors when the biomass contribution is as patchy as in an aggregated Nodularia culture. B: The microclimate conditions a Nodularia culture is exposed to in a stagnating culture bottle are not comparable to natural microclimate conditions. C: Aggregations formed by cultured Nodularia show little similarities to natural aggregations and may differ strongly in there physical properties. D: The microclimate inside an aggregation is characterised by strong chemical inhomogeneity (Ploug, 2008) that makes it impossible to correlated measured variables to a chemical parameter. We are currently working on an alternative approach to elucidate the role of microenvironments in the interaction of filamentous cyanobacteria with their physico-chemical surrounding. A shorter version of this discussion is now included in the manuscript. The statement that there might be a change in species composition was based on data on Anabeana by Franz et al. (2009), that showed a similar response to elevated [CO2]. However, Anabaena reacted with a lower sensitivity and partly different trends in elemental composition of the formed biomass. We specified that in the discussion and cited a publication by Tortell et al. (2002a) presenting similar results for eukaryotic plankton.

Interactive comment on Biogeosciences Discuss., 6, 4279, 2009.