Ocean Science Discussions

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Comment on os-2021-82

Anonymous Referee #1

Referee comment on "Salinity as a key control on the diazotrophic community composition in the southern Baltic Sea" by Christian Furbo Reeder et al., Ocean Sci. Discuss., https://doi.org/10.5194/os-2021-82-RC1, 2021

The study contains an interesting data set with UCYN-A including both genes and potential N2 fixing transcripts in the southern Baltic Proper. However, the study was conducted a bit off season as compared to expected and the manuscript lacks an argumentation to why this study is needed? Was it late in the season on purpose and if so why?

I think the combination of genes and transcripts is interesting, but I wonder if the transcripts can be somehow quantitative. Maybe you can present also transcripts per gene copies so its normalized to abundance? I think this particular data is what is novel with this study and should be lifted in the manuscript and aims. Especially UCYN-A has not been widely studied in the Baltic Sea and not this late in the season.

I am a little bit confused by the use of the bubble method here for N2 fixation as I thought this was not used any more due to the risk of underestimation (e.g., White 2012) and recent studies in the Baltic Sea has used dilution method (e.g., Klawonn et al. 2016). I think this should be discussed in the manuscript and not lifted as an advantage as it is now. Can it be that rates were underestimated?

The statistics needs some work in the paper, better describe the PCA in the text and why you have two graphs presenting what seems like contradicting results. As it is now it is difficult to tell apart what is correlated to what since all arrows has the same colors and point in different directions in A and B. Maybe it is better to run an NMDS analysis for stations and taxonomic groups correlated with environmental factors (as arrows on top), and only one panel? With significant arrows in one color and non-significant in another.

I suggest that the manuscript need major revision before being considered for publication.

Detailed comments

Lines 7-8, change to "decreased salinity due to altered precipitation related to climate change"?

Line 8, clarify N2 by including nitrogen (N2) fixing?

Line 17, Nodularia spumigena?

Lines 18 and 19, gene copies? Cells? Filaments?

Line 20, are you sure they are N2 fixers or should you say potential before?

Line 20, I think statistical testing here is redundant. Either say what test or just skip it and start the sentence from "salinity was identified".

Line 22, similarly significant? Either it is significant or not?

Line 38, change to severely affects?

Line 44, is this how the reference should look like?

Line 46, remove the comma after both?

Line 66, Anabaena/Dolichospermum if often referred to as spp. since its not only one species. You should state Klawonn et al. 2016 or similar study here when saying these three carry out the N2 fixation.

Line 68, maybe also mention rates from newer studies such as Klawonn et al. 2016.

Line 69, I don't understand this huge deviation between what the fix and consume, the 35% in Ploug et al. would not explain that big difference in rates.

Line 72, missing period.

Line 81, what unit?

Lin 82, how does it affect the nitrogenase activity?

Line 86, abbreviate to N. spumigena after first mentioned. I think you should broaden the range, Aphanizomenon is extremely common in the Baltic Proper with salinities around 5-6, with higher densities than the Bothnian Sea.

Line 87, change "speak for" to indicate?

Line 89, include Southern Baltic Sea?

Line 90, why did you choose a low productive season?

Line 91, what do you mean by controls?

Lines 89-92, I think you should revise this aims section as you need to argue for why you choose the low productive season and also that you actually look at genes and transcripts which is really cool. That salinity affects the diazotrophic community composition is already known, but it is novel that you here look at other diazotrophic taxa than the heterocystous ones, I think this should somehow be told here too.

Line 97, change on to using and remove "to the Baltic Sea"?

Line 102, both ammonium, nitrate, and nitrite?

Line 110, change to fixed?

Lines 112-113, revise so that it is clear that the 0.5-1 L was filtered onto the membrane filters and change filter to plural. On all stations and depths?

Line 115, it is not clear to me how these samples were collected, all from the 0.5-1 L filtered samples?

Line 123, one from each station? Please clarify how you came up with 15. Which depth?

Lines 132-135, why these random depths?

Line 149, the statistics section is usually found at the end of the method section?

Line 150, I think you need to provide more details on the PCA here, what are the different components in the figure? What is the difference between figure A and B?

Line 155, why these stations and which depths?

Lines 156-157, I do not fully agree with this since lately this method has not been used due to underestimation risks (e.g., Klawonn et al. 2016).

Line 157, top-filled?

Line 160, did you measure this or is it based on calculations? What is this in moles?

Line 162, were they incubated for the same time? Any labelled T0 samples collected to ensure free of contamination?

Line 168, feels like a few words are missing, the typical Baltic Sea gradient I guess?

Line 170-171, this is very low temperatures for Nodularia.

Line 178, lower than what? Expected?

Line 180, you refer to DIN in the methods? Please be consistent.

Lines 184-185, higher than the report or as compared to other locations?

Line 195, or temperature?

Line 196, that does more sound like they have more N than C not N depletion?

Line 199, you have another order in the methods, with N2 fixation and community, consider keeping it consistent?

Lin 203, what is your detection limit?

Line 204, did this occur together with high Chl a concentration or cyanobacteria abundance?

Line 208, lower the "2".

Line 208, maybe expand here connect with lines 212-218? Maybe move the isotope data to the niche discussion?

Lin 209, stick to past tense, "was supported".

Line 216, abbreviate to N. spumigena.

Line 217, low light when? As compared to what?

Line 219, if you state low rates, maybe give examples of high rates from the area or put in some kind of perspective.

Line 222, maybe mention where the rest of the N might come from?

Line 236, it is very common in the Baltic Proper, starting early in the season, with salinity of 5-6 (e.g., Svedén et al. 2015 in FEMS) and dominating N2 fixation (Klawonn et al. 2016). Saying that it prefers as low as 0-2 is not really true here.

Line 238, I think you rather mean Dolichospermum? Although it used to be called Anabaena it has been referred to as Dolichospermum since 2009 (Wacklin) and then you can also refer to Olofsson et al. 2020 and Klawonn et al. 2016 here. You refer to it in line 66.

Lines 423-427, how does gene copies relates to cell numbers?

Line 261, check the reference layout.

Line 276, how can genes and transcript be related to cell numbers?

Line 298, do you mean Olofsson et al. 2020 here? Karlberg and Wulff was a laboratory study. Or if you refer to several studies in the sentence then include them, now it sounds like Karlberg and Wulff has done everything in the sentence including modeling.

Line 313, maybe specify where this freshening would be beneficial to Nodularia? Since freshening in the northern Baltic sea will have an opposite effect since its already very low.

Line 324, maybe include south here?

Line 725, Olofsson et al. 2021 is missing from the reference list?

Figure 1, maybe have the three figs in a row in the same size instead? Why is it called last? Can you mark those stations where N2 fixation were measured? And explain in the legend.

Figure 2, maybe include 20°E as well since this on the map in Fig. 1? Number of samples?

Figure 3, "pres [db]" need to be better described. This figure feels a bit redundant, maybe move to supplementary? Number of samples?

Figure 4, the text on the axes is far too small. Also the information within the graph-window. A lot of the discussion part of the text in the legend should be in the manuscript instead. Why is it limited to those stations? Number of samples?

Figure 5, consider stretch the x-axis so that the numbers is visible, now it almost overlaps. I would suggest that you write out carbon fixation in the legend, now it looks like it say Figure 5 C. This legend also consists of much results that should go into the manuscript instead. Why limited to those stations? Number of samples?

Figure 6, the text is too small to be able to read, can you make it larger? Number of samples? Move to supplementary since you already have so many figures?

Figure 7, move to supplementary since you already have so many figures?

Figure 8, consider using colors instead, now its hard to distinguish. Can you maybe make also a graph with transcripts per gene copies so its normalized to abundance? Number of samples?

Figure 9, what is components one, two, and three? I don't see the differences between the figures and what has been done? Also how can for example N2 fixation and Nodularia point in different directions on B while being in the same directions in A? What do you mean by the center of each station?

Table 1, how were these numbers extracted? For example, 0.36 and 0.37 mmol N m-2 d-1 in the table is much lower than reported in Olofsson et al. 2021 figure 5? Provide ranges when you have, Klawonn et al. 2016 for example also have many measurements so you should be able to provide a range or mean value with SD. Why does some have ranges and some means and some just one value? Also use the same number of significant digits where you can, Rinne et al. for example has an absurd number of digits?

Table 2, is these mean values for those studies? Provide more details in the legend. There should be more studies providing this for the Baltic Sea? Are they from the same season? Species?