

Interactive comment on “Consistent increase in dimethyl sulphide (DMS) in response to high CO₂ in five shipboard bioassays from contrasting NW European waters” by F. E. Hopkins and S. D. Archer

Anonymous Referee #2

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GENERAL COMMENTS.

The authors present data from 5, 4-litre volume, bioassay or incubation experiments conducted onboard ship in June-July 2011. The seawater samples for these experiments were collected (stainless steel frame, CTD and rosette) at 5 geographically different sites around the UK with variable waters. The carbon chemistry of the water was manipulated by addition of sodium bicarbonate and hydrochloric acid to enable incubation at ambient, 550,750 and 1000uatm CO₂ levels. The main take home messages are that when compared with the control DMS concentration increased and DMSPt

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decreased when CO₂ was increased. This is opposite to most (though not all) of the results presented in the literature to date. DMSp production rates and DMS consumption and turnover rates were also measured but the data gave no consistent response with CO₂ concentration.

Overall this is a well written paper with relatively few minor errors. Plus points are the geographical coverage of the data set and the inclusion of data for DMSp production rates and DMS production and consumption rates. Negatives include a lack of clarity in aspects of the methodology and some unjustified or poorly justified assumptions. Revision of these aspects should lead to a good paper matching the publication criteria of Biogeosciences.

SPECIFIC COMMENTS.

The following issues need to be addressed and properly justified in the text ahead of full publication:

- 1) The abstract is not very informative nor quantitative – please add more detail about where the study was done and how, what was measured and quote specific data.
- 2) Much is made of the level of replication e.g. abstract line 5, 2272 line 23. However, this relates to the experiments conducted by all the scientists onboard the ship and it is an over-statement for the experiments presented in this manuscript which have a standard 3 replicates. Please correct.
- 3) Simulating the real world in an incubation experiment is challenging and all approaches have limitations. For ocean acidification the insurmountable issue is timescale; most natural waters will show a gradual shift in pH and CO₂ concentration over future decades rather than a sudden shift from ambient CO₂ to a higher level. The authors state their experiments assess only the short term response (2271 line 16), but it could be argued that this approach is equivalent to looking at a ‘shock response’ rather than an acclimation response –again it is a question of timescale. Please reword

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this section.

4) The authors argue that short term perturbation experiments are 'superior' to the 3-4 week enclosed mesocosm approach without any mention of the 'bottle effect' that was well documented in the literature more than a decade ago. This effect was amongst the strong reasons for adopting the mesocosm approach. The 2 references below are examples of molecular studies that document shifts in bacterial population composition on similar time-scales to those in the Hopkins and Archer experiments. Lebaron, P., Servais, P., Troussellier, M., Courties, C., Muyzer, G., Bernard, L., Schäfer, H., Pukall, P., Stackebrandt, E., Guindulain, T., and J. Vives-Rego. (2001) Microbial community dynamics in Mediterranean nutrient-enriched mesocosms: changes in abundances, activity, and composition., *FEMS Microbiology Ecology*, 34: 255 - 266 Schäfer H, Bernard L, Courties C, Lebaron P, Servais P, Pukall P, Stackebrandt E, Troussellier M, Guindulain T, Vives-Rego J, Muyzer G (2001) Microbial community dynamics in Mediterranean nutrient-enriched seawater mesocosms: changes in the genetic diversity of bacterial populations , *FEMS Microbiology Ecology*, 34: 243 - 253

5) The major conclusion that DMS concentration consistently increased when CO₂ was increased relative to the ambient controls (2268 line 10) whereas DMSPt decreased when CO₂ was increased. The biggest problem this dataset presents concerns the control data. The data for the time zero time-points were for water collected in a completely separate water collection cast to those for the subsequent 2 time points (top of page 2273). I can see the practicalities leading to this decision, but seawater is very heterogeneous in both space and time. This puts the validity of the all-important 1st data point in serious doubt. The 2nd cast was presumably taken at the same location but post-dawn and with analytical samples taken directly from the Niskin bottle and so not treated in an identical way in terms of handling, decanting into bottles etc. The authors need to convince the reader that this is an acceptable approach and optimally to prove it with data.

6) Also concerning the control/T0/initial conditions data, there is a mismatch between
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the E01 and E02 DMSPt data in Table 1 (59.6 and 25.9 nmol l⁻¹) and Figure 2 (~25 and 60). Perhaps the data in either table or figure have been switched?

7) For detail of the method for measuring the DMSP synthesis and production rates the reader is referred to Archer et al 2013 (2274). I checked this and noticed that the isotope fractionation factor used is that derived from studies on cultures of *Emiliana huxleyi*. Here the data are for variable mixed populations and the assumptions behind this deserve the addition of a few lines in the discussion. Likewise, the K_s values determined were highly variable but a mean value was used (2277). As these were not measured at each site the implications need to be worked through better in the main paper discussion section.

8) The light incubation conditions are poorly described. Please give more detail on the LED panels (2272 line 19). What wavelengths of light were covered? Give the in situ light conditions in Table 1 for comparison and say whether there any attempt to adjust the light to the ambient conditions (excepting UV)? The discussion section page 2284 is misleading given that polycarbonate bottles generally cut out UV light. Why should the response to elevated CO₂ be comparable to the response to UV?

9) There is no discussion at all on grazing effects – which is surprising given that the 2nd author has published on this topic in the context of DMS and DMSP. This means there is an inherent but unwritten assumption that grazing is the same across all CO₂ treatments, but what evidence can the authors offer to corroborate this?

10) I agree that the data could be suggestive of algal processes, exudation and lyase activity being part of the responses seen in bulk DMS and DMSP concentration, but neither is proven here. The wording should be removed from the abstract and the wording in the main body of the paper needs to be toned down to a more suitable level to prevent others quoting this as an experimental finding. These parameters are very difficult to isolate experimentally and the authors should offer specific suggestions for future research concerning these processes.

OTHER COMMENTS:

The following paper is relevant and should be mentioned: Lee, P.A. et al. 2009. Effects of increased pCO₂ and temperature on the North Atlantic spring bloom III Dimethylsulphoniopropionate. Marine Ecology Progress Series 388: 41-49 doi:10.3354/meps08135.

2269 line 3 – the Stefels et al 2000 paper concerns the overflow and not the antioxidant hypothesis.

2269 first full paragraph – the work of Todd et al on the wide variety of DMSP-dependent DMS release pathways ('lyase') deserves a mention.

2271 line 9 – say how DMSP responded.

2272 – state whether water was 200 um filtered or not.

2272 was the pH maintained throughout the timecourse of the experiments. Add data to Figure 2.

2272 line 15 – give a full description of the septa lids.

2275 – States that system sensitivity and drift were monitored, but it isn't clear whether there was no drift or whether data were adjusted accordingly.

2280 line 11 – there is no taxonomic data so this can't be proven.

2286 line 27 do you mean consumption kinetics were saturated?

2287 line 8 do you mean Figure 6 there is no Figure 7.

2287 it is a pity to limit the discussion to E01 and E04!

Table 1 please add nitrate and ammonium data as well as TON.

Figure 2 the curves fitted to the data here are a bit misleading e.g. for site E03 DMSPt at ambient CO₂, the concentrations at 48 and 96 h are very similar but the curve gives

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the impression of an increase.

Figure 5 legend mentions asterisks denoting a significant difference from ambient bioassays but none can be seen in the figure.

Figure 6 legend mentions mean values open circles but there are none on the figure.

Figure S1 please provide a legend to indicate which profile is for which site.

Figure S2 the kinetic curve is only provided for KE3, add the curves for the other 2 sites as well.

Interactive comment on Biogeosciences Discuss., 11, 2267, 2014.

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