Review of Biogeosciences Manuscript
Bg-2011-400

Title: A molecular perspective on the ageing of marine dissolved organic matter

Authors: R. Flerus, B.P. Koch, O.J. Lechtenfeld, S.L. McCallister, P. Schmitt-Kopplin, R. Benner, K. Kaiser, G. Kattner

General Comments

This manuscript presents ultrahigh resolution mass spectral data on 137 waters collected from the Eastern Atlantic Ocean. Organic carbon concentrations, amino sugar yields, and radiocarbon measurements are reported. Certain peaks in the mass spectra were shown to correlate linearly with the $\Delta^{14}C$ values (either positively or negatively), and a degradation index was developed based on this data. The science and data interpretation in this manuscript is of high quality, and the novelty of the data analysis routine is particularly unique. While many FTICR-MS studies do an overall characterization of solid phase extracted dissolved organic matter, this manuscript uses the molecular-level information in a new way that relates specific peaks with other bulk measurements, which is far less common. The large sample set allows for this extensive study to be particularly useful, and I think this manuscript could make a particular impact on the DOM community.

Detailed Comments

1) Page 2, Lines 21-22: "All other compounds should persist partly thermohaline circulation." This sentence reads quite strangely to me, and I think a preposition is missing somewhere. Perhaps adding 'during' or 'throughout' is better? "All other compounds should persist partly during/throughout thermohaline circulation." This sentence is also in the caption to Figure 10 and needs updating there as well.

2) Page 3, Line 15: "modern" refractory DOM seems to be an oxymoron. Do you mean younger, refractory DOM? An additional explanation to this sentence and the Jiao et al. citation would be useful here.

3) Page 5, Lines 8-11: The reference given for the water sampling details is a manuscript that is in preparation. A few more sentences describing the sample stations would be useful for readers.

4) Page 6, Lines 4-5: How much ultra-pure water was used to re-dissolve the 50 uL of dried extract? Add this to the experimental information.

5) Page 7, Lines 12-14: Section 2.4 states that the mass spectra are evaluated at 200-500 m/z, but the spectra are internally calibrated at 340-540 m/z, with only 5 peaks. This is surprising, as internal calibration is typically performed throughout the entire m/z range being analyzed, using

at least 1 calibrant peak every 30-50 m/z units. Why is the calibration list used so short? How can the authors be confident that 0.5 ppm is a sufficient limit for formula assignments?

6) Page 7, Line 18: Why do you only use a m/z range of 200-500? There is quite a lot of data at 500-600 m/z (according to Fig. 1). If you calibrated to a higher m/z range, the accuracy would be sufficient for molecular formula assignment. The S/N at 500-600 appears to be about the same as 250-300 m/z, which was used during data analysis.

7) Page 8, Line 3: I understand that the peak magnitudes were summed for each mass spectrum, but I do not understand the intensity threshold described as 100 ppm. From what I can gather, this is similar to a S/N threshold, but calculated by the authors, rather than using the Bruker software. Is that correct? If so, what exactly is this 100 ppm, and how is it calculated? What S/N threshold is used in the Bruker software to construct the mass lists, prior to summing the peak magnitudes? Because this parameter is used throughout the text, the calculation and justification for its use should be more clear. I do agree with using normalized peak magnitudes (as described by the authors at lines 8-11 on this page).

8) Page 8, Lines 24-25: Regarding Figure 3, why are there no values for the y-axes shown on the plots? The authors state how much magnitude the summed peaks account for in all the spectra ($66 \pm 1.4\%$), so why aren't values shown on the y-axis?

9) Page 9, Line 4: I believe Table 1a should just be Table 1.

10) Page 10, Line 10: For Figure 6, it may be useful to put a line on the van Krevelen plot at H/C=1.0. The y-axis only shows 0 and 2.5, and I think having a line at 1.0 would be beneficial to readers.

11) Page 10, Lines 26-28: The authors give H/C ratios for young DOM as 1.27 and 1.25 for aged DOM. Are these 2 values statistically different from each other? If you compare the magnitude-averaged H/C ratio for all 137 samples, is a difference of 0.02 actually statistically different?

12) Page 10, Line 29: For Figure 7, how are the dotted lines actually calculated? It is not clear from the figure caption, and these lines are not discussed in the text. I think they could easily be deleted from the figure to avoid confusion, since the authors don't actually discuss them in the text.

13) Page 11, Lines 10-11: The authors state that CRAM is obviously a significant contributor to SPE-DOM. This is not obvious. This statement is based only on the location of points in the van Krevelen diagram, which is an area of overlapping functionality that is likely due to any number of structural isomers of formulas that could be assigned to a variety of biomolecular compound classes.

14) Page 11, Line 20: For Table 2, I think the caption should be 'Formulas utilized for magnitude summation in order to calculate the degradation index' or something more along those lines. The wording of the current caption is confusing.

15) Page 11, Degradation Index: The authors give the equation but do not justify it. How was the calculation developed? Because this index is an important part of the manuscript, the authors should do a better job of describing the index and rationalizing the calculation.

16) Page 11, Line 22: The authors state that the index can differ due to the instrument. Does that mean which specific FTICR-MS was used? What are the differences between the instruments utilized by the authors (magnetic field, manufacturer, operator, installation time-frame, etc.)?

17) Page 12, Lines 1-2: The authors give degradation index values in the range of 14.6-40.1, yet the coloring scale on Figure 8 appears to be more narrow (18-28). What is the reason for this discrepancy?

Recommendations

Accept to Biogeosciences after suitable revisions have been made, with special attention to detailed comments #5, #7, and especially #15.

END OF REVIEW