Biogeosciences Discuss., 10, C6126–C6129, 2013
www.biogeosciences-discuss.net/10/C6126/2013/

Biogeosciences
Discussions
Open Access

Interactive comment on "Acquisition of intact polar lipids from the Prymnesiophyte *Phaeocystis globosa* by its lytic virus PgV-07T" *by* D. S. Maat et al.

Anonymous Referee #2

Received and published: 26 October 2013

The manuscript by Maat et al. focuses on lipid changes in phytoplankton during algal virus-host interaction of the globally relevant Prymnesiophyte Phaeocystis globosa with its lytic virus PgV-07T. The authors compared the membrane lipid composition of non-infected and infected algal biomass, and of the purified virus. The overall similarity of the lipid pattern of infected and non-infected cultures together with the lack of characteristic viral membrane glycosphingolipids (vGSL) which were found in a previous study of the Prymnesiophyte E. huxleyi and its virus EhV-86 led the authors to the conclusion that the virus-host interactions vary considerably depending on the involved algal host.

General comments:

The manuscript is well written and the topic is certainly interesting to the readers of Biogeosciences. However, I agree with the assessment of referee #1, that the results of this study are only of limited significance because only one additional virus-algal host system has been analyzed. In light of this, I do not agree with the general conclusion that "the absence of viral sphingolipids as shown in the current study might be a more general feature" (page 11718, line 3). We have one system producing vGSLs and another that does not, so I suggest to not jump to early conclusions until a variety of other species have been studied.

My major point for improvement regards the reported IPL data that is currently quite lean. All conclusions of this study are based on the comparison of IPL compositions of the different experiments and even small changes might be significant. The current manuscript will thus greatly benefit from a more detailed description of the IPL composition, which also directly translates into increased relevance for future studies of other viral-algae systems where robust data for comparison is mandatory. With little additional work, the improvements outlined below will strengthen the manuscript considerably and raise the impact from "smallest publishable unit" to "good research paper":

(1) A more detailed description of the detection limit for the individual IPL classes. Some IPLs were below detection limit, where is the cutoff – what is significant?

(2) The use of response factors for quantification of relative IPL distribution, which is also directly relevant to a question raised by referee #1. It is known, that different ionization efficiency of different IPL classes can lead to apparent increase/decrease of the relative proportion of certain IPL groups when not considered. Quite often IPL standards are not commercially available and can thus not be used for quantification. In this case, however, all the required IPL standards to determine the relative response are available in the lab (P11712, lines 8-12).

(3) Most importantly, a more detailed description of the variation of fatty acids within

each IPL class is necessary. The base peak plots in Fig. 4 show clear evidence of in-class variation of fatty acids in the IPLs, especially for the phosphocholine lipids. As the authors report (page 11714, line 23), the saturation state within IPL classes can be affected by viral infection (Evans et al., 2009). I strongly suggest that such data be included in the revised version of the manuscript. The good news is, that a lot of the required data are already available from the MS2/MS3 fragmentation pattern already acquired and that only four additional analyses in negative ionization mode are necessary for the remaining IPL species. Alternatively, as bare minimum, the authors could include information about the total number of carbon atoms and double bonds combined in both fatty acid chains; this information is available in the MS full scan data. The former option, however, is more desirable because it allows direct comparison with existing and future fatty acid data ontained by gas chromatography. As a result of the modifications, Table 1 needs to be updated to include the relative composition of different IPL groups in % and the additional in-class fatty acid data.

Detailed comments

P09, line 24: what was the filter pore size?

P09, line 7: I believe it should be “4100 x g”

P10: line 4: filter pore size?

P12, line 12: here the description of response factor and detection limit determination can be inserted.

P13, line 12-16: there are clear differences in Fig. 4 not only regarding the abundance, but also regarding the fatty acid pattern (see general comment above). This needs to be addressed.

P13, line 22: state the detection limit

P14, line 5-6: the relative abundance of polar headgroups might be similar, but what about the fatty acids (see above).

P14, line 22-paragraph end: here the authors suggest that the fatty acid composition within each IPL class might be different and they state the relevance of unsaturations (Evans et al., 2009). Since fatty acid composition data has already been generated or can be recorded with little extra work, it should be included in the discussion here (see general comment above).

Section 4.2: the discussion of similarity of IPL patterns of the algal host and the virus would also benefit greatly from a more detailed comparison of the fatty acid composition. I am sure, that different IPL sources within the cells (cytoplasm, cell membrane, endoplasmatic reticulum, chloroplasts etc) might lead to different fatty acid compositions – this should be considered in the discussion. I wonder, if there might be some carry-over of lipids from the host lysate to the purified virus solution during the separation outlined in section 2.4. Is it possible, that some of the lipids found in the virus might be derived from incomplete separation? This could be tested by adding a synthetic standard to the lysate before isolation of the virus and subsequent testing for the presence of the synthetic compound in the virus extract. Has this been considered? Could fatty acid patterns help to exclude this possibility, too?

P17, line 8-11: maybe some IPL-group-fatty acid compositions are specific for the infected culture or the virus?

P18, line 3-8: see general comment above. We don’t have enough data to generalize.

Fig. 3: it occurs to me that some of the structures are negatively charged while others are positively charged or in a neutral zwitterionic state. Perhaps this can be made consistent (all with a net neutral charge).

Fig. 4: can the figure be expanded a bit?

Interactive comment on Biogeosciences Discuss., 10, 11705, 2013.