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Interactive comment on "C_5 glycolipids of heterocystous cyanobacteria track symbiont abundance in the diatom *Hemiaulus hauckii* across the tropical north Atlantic" *by* Nicole J. Bale et al.

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Thank you for your initial pre-review comments. Indeed the lack of figures does not make for an auspicious start to the reviewing process. We are unsure what happened to the figures and contacted the journal, who suggested we uploaded the full document with figure below, as we have now done.

Regarding the quantification method we have applied in this study. As described in the method section, our samples contained a short-chain glycolipid standard, n-dodecyl-

β-D glucopyranoside, which is indeed a glycolipid with a hexose head group rather than a pentose one. In the majority of the studies of heterocyst-forming cyanobacteria that we have carried out, hexose glycolipids were the target compound. Hence N-dodecyl-b-D-glucopyranoside was developed for use as an internal standard as it is a commercially available analogue of the naturally occurring hexose glycolipids, yet contains a carbon chain that is not observed in nature. Furthermore we were previously able to isolate a hexose glycolipid, 1-(O-hexose)-3,25-hexacosanediol from cultures of free-living heterocyst-forming cyanobacteria (Bale, 2017) using preparative HPLC in order to determine a RRF between it and the internal standard. It is not currently possible to isolate enough of a naturally occurring pentose-glycolipid due to the limitations of culturing diatom-diazotroph associations. However, we do not expect massive differences in ionization efficiency between a hexose and a pentose glycolipid. Rather the lack of alcohols on the chain is a more likely factor driving any difference. The RRF of the internal standard and the hexacosanediol is therefore probably fairly realistic and, at this point, due to the lack of standards, our approach is the most realistic estimate of quantities there is right now. We will make it clear in any revised version of the manuscript that our quantification of the pentose-glycolipids is semi-absolute, as we used a similar, but not identical compound. But as the trends in the natural glycolipid concentration would not change with a different RRF, the conclusions in our manuscript regarding their suitability as biomarkers for diatom-diazotroph associations stand.

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