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Interactive comment on "Sterol preservation in hypersaline microbial mats" *by* Yan Shen et al.

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Shen et al present a paper that is very similar in set-up as a paper of the same first authors published in 2017, where they analyse the sterol distribution in a microbial mat from a hypersaline lake on Christmas Island (Kirimati) - the main difference is that they now targeted another lake - which was previously investigated by the same group (Blumenberg et al 2015) who published hypy-GCMS results. This paper presents free sterols and some fatty acid data from the same microbial mat. The data is interesting and gives some additional insight in the lipid biomarker composition of these hypersaline mats, as a modern analog to ancient stromatolites.

I have some concerns that i like to be addressed before accepting as a final paper: * The use of TMCS in methanol to methylate fatty acids is rather uncommon. Please provide a reference where this method and its efficiency is described. * How were the 13C

contents of the sterols and FAs corrected for the added derivatizing groups? * I wonder if pyrolysis GC-MS is the best way to assess if steroids make it into the 'kerogen' (residue after extraction) fraction of recent material. I am not a py-GCMS expert but pyrolysis at 560'C is a rather high temperature where most organic molecules will break down to smaller pieces; any remaining intact compounds or larger fragments will be low in concentration. The fact that some hopanoids (fragments) may indicate that they are simply more abundant, while any remaining steroids could be below detection limit (i.e. below the background of the 213+215+217 trace). Absence of evidence is not the same as evidence of absence. Information about the reference kerogen of the Green River shale is lacking (e.g. sample amount, or relative amount of hopanes-steranes in Green River bitumen) so this comparison does not tell very much. More critically, Blumenberg (2015) could report hopane/sterane ratios for the same mat, which means steranes were present throughout – although they did appear to decrease with depth (at least compared to the hopanes). I would have the same critique to the already published paper about lake 22 (Shen 2018) when it concerns the use of py-GCMS to investigate the presence of steranes in the 'proto-kerogen'. * The paper cannot really be read without also consulting a more comprehensive hypy-GCMS- biomarker analysis published by Blumenberg (2015), who finds hopane/sterane ratios of 20-100 – thus no surprise there are no sterols found by py-GCMS while hopanes do show a trace.

* Why did the authors not measure (or present) free hopanoids? Or for that matter a more comprehensive biomarker study (i.e. a free extractable lipid version of the Blumenberg paper). This would give the paper much more body, constraining it to only steroids feels very limited. I strongly recommend expanding the paper in this way.

* The 14C dating of carbonates on a Coral atoll has a large risk of a reservoir effect (the coral is likely from the mid-Holocene sea level high stand thus several 1000 years old). Indeed a 14C age of just -239 years indicates a fraction modern of just over 1, i.e. a mixture of post-bomb atmospheric CO2 and an ancient source. The downcore increase in age does make sense, but one cannot assign any exact ages to the mat

material – for this, one needs to date plant macrofossils. I realise the results were published already by Blumenberg et al but they can only be interpreted as deeper = older. * When looking at the depth profiles of the sterols, I do not see a clear decrease with depth, except the large difference between the surface layer and layers below the phototrophic active part. Layer 5 and layer 2 have the same concentration per g TOC, and layer 3 and 6 the same expressed per g dry mat. * Comparison with lake 22 (Shen 2018): What is different between the two lakes is a halite-gypsum crust on top of lake 22 – which must impair oxygen flux to the upper layer. Yet, lake 22 shows considerably higher steroid concentrations than this lake 2. This may explain the absence of higher sterol abundances in the upper layer (i.e. absence of eukaryotic sterol-producing photosynthetic organisms in lake 22 but rather a 'fossil' signal starting already in layer 1 below the halite crust. The higher sterol conc. in lake 22 may simply be a higher contribution from terrestrial vegetation, but as the authors state it can also be a lower degradation because of ultrahigh salinity. Coprostanol found in lake 22 could be derived from the abundant land crabs on Kirimati, which live around the lakes and eat the local vegetation (and each other - personal observation in 2005). * I agree with the final conclusion that different microbial mats, like lake 22 and lake 2, generate different fossils records, because of their different limnic/environmental properties. * I also agree that the data confirm the hypothesis that microbial mats do not preserve original photosynthetic lipids from the upper very well, and that this signal is overprinted by heterotrophic organisms. However marine or lake sediments do have the same 'problem': organic matter degradation on heterotrophy within (anoxic) sediments, i.e. a diagenetic overprint. * That said, these are not really very new insights, the added value compared to the Blumenberg 2015 and Shen 2018 papers is marginal. * I do not agree with the conclusion that steroids are not preserved, in my opinion the authors have not used the right method to investigate this. Blumenberg (2015) found steranes after hypy.

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