

Dear Marcel,

Thank you for approving the manuscript and the comments. I also carefully read it again.

Sincerely

Valerie

Line 21: technically I think it are phospholipid derived fatty acids. It might be good to make that clear at least once or twice.

Yes, I introduced this line 21 and 69.

Line 60-61: ...increasing number of studies report the importance of chemolithoautotrophy in groundwater...

Changes done

Line 66: "Intact polar lipids, mainly phospholipids, are important constituents of bacterial and eukaryotic cell membranes and consist of a polar head group linked to a glycerol backbone with two fatty acids esterified to it." Or something similar. The lipids are not in the membrane as fatty acids, but as apolar tails of an intact polar lipid. You have to break the ester bonds to free them, hence the phospholipid derived fatty acids. And it excludes Archaea since they make completely different lipids.

Changes done

Line 77: in general autotrophs are ... In the discussion you do mention the reversed TCA cycle and heavy isotopic composition. Would it makes sense here to use something like "typical RuBisCO carbon fixation"?

I preferred to use heterotrophs versus autotrophs since here we theoretically don't have photoautotrophs

Line 83: despite PLFAs being widely used ...

Changes done

Line 84: microbial communities

Changes done

Line 85: limitations of PLFA based studies

Changes done

Line 85-87: The big risk is that so many micro-organisms have never been studied in "pure" culture and we do not know what they make. So I agree with your statement, but it might be even a bit more tricky than that.

And particularly in groundwaters...

Line 94-100: yes, and glycolipids are not only storage lipids, there are also functional glycolipids. So even if the separation would be perfect you would still not necessarily separate structural from storage lipids.

Yes, both DNA and PLFA studies have weakness. I do not think the ideal marker exists. I hope by such a combining approach to overcome some of those problems.

Line 98-99: PLFA fractions
Changes done

Line 178: define FAME, this is now in line 188?
Changes done

Line 184: remove the , after and
Changes done

Line 188: see comment on line 178.
Changes done

Line 283: were instead of was
Change done

Line 338-343: ? I found this confusing, the explanation for PC3 is missing, but there are three “separations”?

The main grouping is along PC1 and PC2 which separated the wells according the water chemistry in three groups. PC3 is not relevant for the discussion since it may separate the wells according the sampling dates. But, this has to be confirmed with more data points. I rewrote this part.

Line 342: either there should be a . after 5.3 or it should be “along”. (see also previous comment).
Change done

Line 362 and 363: I assume with increasing O₂ concentrations. Perhaps it would be good to actually say that especially in line 363.
I am not sure to understand this comment. I rewrote the sentence.

Line 377: 13C-enriched, more positive and therefore 13C-enriched at least compared to the more negative values associated to Annamox.
Change done

Line 473: eukaryotes such as microalgae etc.
Change done

Line 485: limited in food, for photoautotrophic micro-eukaryotes it might be nutrients, but they also need light.

Photoautotrophic organisms are really rare in groundwater. DNA showed some cyanobacteria but they are likely introduced from surface.

Line 593: you use evidence quite a few times, why not show or sometime suggest. Evidence used in this way feels weird.

I replaced evidence by show.