

Interactive comment on “Novel hydrocarbon-utilizing soil mycobacteria synthesize unique mycocerosic acids at a Sicilian everlasting fire” by Nadine T. Smit et al.

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This is a very interesting study and well written manuscript. In addition to the comments raised by the reviewers, I would also like to add a few more, very minor comments for you to consider:

Line 14: Could you please add more examples here instead of stating “various habitats” of mycobacteria. I see that this information is provided in the main text of the manuscript, but thought it would be good to add some of this information here to show that these organisms have a high diversity.

C1

Line 25 and elsewhere: Better add full name (Fuoco di Censo instead of Censo)?

Line 99-109: Could you please note why you use BDE for extraction and saponification instead of other methods? Using BDE is a great method to prevent decomposition of some lipids, but then you undertake base hydrolysis? Did you also make a comparison of your results by looking at intact polar lipids/PLFAs as well? For the saponification step, is there a reason why you adjusted the pH to 5 and not lower than that?

Line 118: polar compounds

Lines 197-198: Are you able to you add an approximate number to show how much/percentage the uncultured mycobacteria represent?

Line 211: However instead of although?

Lines 217-219: Could you please add more details about these “explorative searches”? Or even a reference to provide more information about the synthesis of biohopanoids in mycobacteria?

Line 244: consider changing to methyl group (twice in this line)

Lines 250-251: M⁺ with superscript “+” and remove “of m/z”, also add respectively at end of sentence.

Line 274: Biomarkers

Line 333: Could you note briefly why the d13C values of methane and ethane are quite enriched in these seeps? Even if the background information is surely provided in the cited study and known to many readers of your manuscript, I think it could be useful for the reader that is not very familiar with this, because methane is commonly highly depleted in other settings.

Line 387: Not necessarily limited to this seep/site. Investigating various other, similar settings would also provide more insight into the abundance and community composition of mycobacteria and their lipid composition (relative amounts and stable isotope

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composition), as well as to better understand how these lipids help mycobacteria to adapt to these special environments.

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