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Interactive comment on "Millennial-age GDGTs in forested mineral soils: ^{14}C-based evidence for stabilization of microbial necromass" *by* Hannah Gies et al.

Anonymous Referee #2

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Gies et al have developed a method to isolate and prepare GDGTs for radiocarbon analysis. In their study, they present 14C data for isoprenoid and branched GDGTs in two well studied soil profiles in Switzerland. They find that GDGTs have an increasingly longer turnover time with depth, and suggest that GDGTs are protected against degradation through associations with mineral surfaces. Their data provide evidence for the contribution of microbial necromass to the pool of stabilized carbon in (deep) soils.

The data and the paper are well suited to be finally published in Biogeosciences. However, I have some comments, questions, and suggestions.

General comments: I miss some basic background information in the introduction. Details on the sources/producers of GDGTs in soils, as well as their metabolism is crucial for this paper, but these aspects are only marginally addressed in the current version. It is not so clear on what level the 14C composition of iso and branched GDGTs will be interpreted: as markers for Archaea and Bacteria, or on a more specific level (e.g. Acidobacteria in case of the branched GDGTs)? The introduction should at least summarize what is known about the producing organism(s) of GDGTs. For the branched GDGTs, at least mention that Ia has been found in specific Acidobacteria, although it has not been excluded that other phyla may produce brGDGTs as well (Sinninghe Damsté et al., 2011, 2014, 2018). Also mention the (presumed) metabolism (for example Pancost and Sinninghe Damsté, 2003; Oppermann et al., 2010; Weijers et al., 2010). Add similar information for the isoGDGTs. They are merely introduced as Archaea, but this is of course a very broad term. Do they have the same producers as in the marine environment? What is their role in soils? Again Sinninghe Damsté (et al., 2012) has done some work on this. I found part of this missing information in the discussion (section 4.2), but I really think this should be part of the introduction already.

In addition, there are a couple of studies that provide estimates of the turnover of branched GDGTs in soils. Although they have not used 14C dating, these studies are currently not mentioned in the introduction. Check Weijers et al., 2010; Peterse et al., 2010; Huguet et al 2013, 2017, who propose turnover rates of years to decades for branched GDGTs in surface soils, each following a different approach.

Finally, the implications of the results for both soil organic carbon cycling as well as for paleoclimate reconstruction have not been given sufficient attention. The authors mention that there are implications for the use of GDGTs as 'tracers and proxies', but it is not clear what those will be exactly. MBT'5me and CBT' ratio values are reported in the results, but these indices are nowhere explained, and the values are not discussed. How will the turnover rates estimated in this study influence GDGT-based paleoclimate reconstructions? One of the few implications that is mentioned in

the implications section (4.3), is that old brGDGTs (older than the depositional age) in sedimentary archives reported in the literature confirm a soil source of brGDGTs, as the old age may be explained by intermittent storage during transport, or export of deeper soil material. This is actually in contrast with the overwhelming evidence for the production of brGDGTs in aquatic systems (rivers, lakes, and the coastal marine environment, see references in detailed comment below, as well as De Jonge et al., 2014 for in situ production in rivers and Sinninghe Damsté 2016 for in situ production in coastal settings), and the offsets in brGDGT signals between e.g. lake sediment and catchment soils (for example, see Tierney and Russell 2009; Miller et al., 2018; Guo et al., 2020). The aspect of in situ production and thus a contribution of aquatic sourced GDGTs (both branched and isoprenoid) has not been mentioned in the manuscript, and should be addressed in the implication section (and introduction, where appropriate).

Detailed and textual comments: P2L54-55: This sentence seems incomplete. P3L56: soils are considered a major source of brGDGTs to aquatic systems: I am not convinced that this is still widely believed. Over the past few years, several studies have provided evidence for a primarily in situ, hence aquatic source of brGDGTs in lakes (e.g. Tierney and Russell, 2009; Sinninghe Damsté et al., 2009; Tierney et al., 2010; Weber et al., 2015, 2018; Miller et al., 2018; Russell et al., 2018; van Bree et al., 2020). P3, section 2.1: also add the amount of samples included in this study. It later appears that one sample from every soil horizon has been analyzed. Provide this information here. P3L82-85: this sentence is very hard to read. It even seems like there may be some words missing? P4L94: . . .is impractical, WE focused instead on. . . P4L96: Adding to my earlier comment: the authors should better introduce the (supposed) sources of the GDGTs in soils in their introduction, and indicate here on what level their putative biological precursors and formation pathways are considered common. For brGDGTs, this may be true on the level of 'bacteria', but there are indications that the 5-methyl and 6-methyl isomers are produced by different subdivisions of the phylum Acidobacteria (Sinninghe Damsté et al., 2018). Given that the authors have pooled the 5-methyl and 6-methyl brGDGTs in this study, this is important information to add.

The same is true for isoGDGTs, of course. P4L124: Was the purity check done on full scan, or using selected ion monitoring as mentioned in Hopmans et al., 2016? In the latter case, how can you assure that there were no potentially co-eluting compounds present? P5 section 2.4: can you give a slightly more elaborate description of the different pools? What kind of compounds do you expect, or are assumed to be part of the fast cycling and passive pools, respectively? P6L164-168: Given the uncertainties associated with the sample size, and the minimal sample sizes mentioned here (at least 20 ugC for samples with a radiocarbon age <1800) and at least 50 ugC for samples older than 6000) compared to the relatively small sample size used in this study (at least 15 ugC), I think it is important to include the sample sizes of each sample in a (supplementary) data table. P7L179: The MBT'5me and CBT' ratios have not been defined. P7L199: Do you mean C28 fatty acid? P8L212: what do you mean with 'light fraction'? P8L212-213: Can you add the range of the estimated turnover times, for reference? How do the turnover times based on the two different approaches compare? P8 section 4.1: I think that the motivation to pool all GDGTs for radiocarbon measurements comes a bit late, as the pooling is a passed station. What if the literature had pointed out that each GDGT was likely to have another isotopic composition? How would you then interpret the pooled results? I feel like this section on the presumed shared metabolism should be moved to the introduction, as I am sure that this information has influenced the approach of this study rather than formed the 'coincidental happy fit' as presented now. P9 section 4.2: same comment here. The metabolism of the GDGTs, and its consequences for their 14C composition, should be better described in the introduction. P10L281: Thaumarchaeota L283: replace studies by studied L301: How do you match the previously reported turnover rates of GDGTs in surface soils of years to decades to centennia to even millennia, as you find here? L323: as mentioned earlier, I am not convinced that brGDGTs systematically trace soil OC given the evidence for in situ production in aquatic systems (rivers and lakes), or the loss of the soil signal upon entering a river (Zell et al., 2013, 2014; Guo et al., 2020). L335-337: Is there any way we can test the protection of GDGTs through association

with mineral surfaces?

General: Damsté et al should be Sinninghe Damsté et al.

Figures: The panels in the figures are almost consistently plotted the other way around than how they are discussed in the text. For example, the Beatenberg profile is described first in section 2.1, but GDGT concentrations are given in panel B of Figure 3. Similarly, results from the Beatenberg profile are presented first in the text (3.3), but are visualized in panel B of figure 4. Can the panels in the figures be turned around to avoid confusion?

References: van Bree et al., 2020 Biogeosciences Discussions De Jonge et al., 2014 GCA – Yenisei river Guo et al., 2020 Biogeosciences Hopmans et al., 2016 Org Geochem. Huguet et al., 2013 GCA 105, 294-315 Huguet et al., 2017 GCA 203 103-116 Miller et al., 2018 Climate of the Past Oppermann et al., 2010 GCA Pancost and Sinninghe Damsté, 2003 Chem Geol. Peterse et al., 2010 OG Russell et al., Org Geochem Sinninghe Damsté et al 2009 GCA Sinninghe Damsté et al 2011 AEM Sinninghe Damsté et al 2012 AEM Sinninghe Damsté et al 2014 AEM Sinninghe Damste, 2016 GCA Sinninghe Damsté et al 2018 Org Geochem Tierney and Russell, 2009 OG Tierney et al., 2010 GCA Weber et al, 2015 GCA Weber et al, 2018 PNAS Weijers et al., 2010 Biogeosciences Zell et al 2013 L&O Zell et al 2014 Biogeosciences

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