

## *Interactive comment on* "Carbon turnover in cell compartments and microbial groups in soil" *by* Anna Gunina et al.

## Anna Gunina et al.

guninaann@gmail.com

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## # Thank you for the study evaluations.

1. Hypothesis 1 could not really be tested given the duration and intervals of sampling during the experiment. Glucose gets processed, incorporated, lost and recycled into PLFA already in the first two days (e.g. Ziegler 2005) and this can vary with soil and environmental conditions. Starting measurements after 3 days leaves us without any information of when the peak of uptake took place (thus when time zero for decomposition started) and when recycling started which would matter for trying to estimating turnover. Then on the other end, 50 day was not sufficient time for the aminosugars to finish building up and start decomposing, as the authors discuss. Perhaps the data can be used to answer a different question. From Figure 2, we don't know how good

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the model fits were (it would be important given there were three points and large error bars).

# Thank you for the comment. The time 0 was not taken into account, due to several reasons: i) microorganisms uptake glucose from soil solution very fast according to lab observations, (reported times are in the second to minutes range (Hill et al., 2008), however it can be slower for the field conditions. So, due to part of the glucose still can be in soil solution the 13C incorporation into cytosol will still increase, whereas on day 3 the complete uptake of glucose can definitely occur. ii) Due to glucose is taken up first into the cytosol pool and incorporation into other pools is delayed, that it would not be possible to have the t0 for other pools. Moreover, in our experiment we were focus on the 2nd phase of glucose utilization and not on the first (uptake phase). Figure 2: we agree that this is not big amounts of points, but curve fitting was done for 4 replications for each time point. Due to this parameter is related to the big uncertainty not a lot of conclusions are made base on that parameter. The most important is that this is only approach, and also that the 13C incorporation decrease or increase over time for the pools which can be clearly seen based on the 3 points. Moreover our experiment was done in the field and much larger variations can be expected compare to lab experiment were the soil is homogenized before the experiment (before the application of tracers).

2. Hypothesis 2 is about differential incorporation by bacteria and fungi (who incorporates it faster). Again, missing the first three day is pretty critical (Ziegler 2005 clearly shows this). There are already many experiments that have assessed "initial" incorportation of 13C into biomarker lipids and we wouldn't then need 50 days incubation for this. Also, I am surprised that there was no effect of time on the composition of the PLFA as 50 days is quite a long time for microbes and PLFA profiles tend to be more sensitive to time than any other driver, and, C depletion in 50 days of an incubation would be substantial.

# Thank you for the comment. Yes, we agree that the time is very important in the labeling experiments, especially if the labile substances are applied. However, our ex-

periment was done in the field (with 1.5 kg of soil and changes of the condition between day and night), and microorganisms can take up glucose much slower compare to lab experiments. In this case measurement of 13C in the PLFAs on the 1st or 2nd day will probably not show the highest incorporation. That is why we did not make the 1st point 1 day after 13C incorporation. Concerning the initial incorporation: most of the experiments were done in the lab, whereas our experiment was done in the field, were processes goes much slower. The data on total remaining 13C after 50 days of experiment are clearly show this: around 70% of 13C derived glucose remained, showing slow rates of processes in the field experiment. Moreover, we were focused on the 2nd phase of glucose utilization and not on the first. The structure of PLFAs stayed quite stable during the field experiment because of the following: i) soil was plowed two week before the experiment and microorganisms were already adopted to the changed conditions of limiting C and ii) low concentration of glucose were applied, which did not promote microbial growth or changes of microbial community structure.

3. Hypothesis 3 is hard to follow. I tried but was not able to understand it.

# Thank you for the comment. We have deleted hypothesis 3.

4. In the Introduction, the paragraph comprising Lines 96-108 is a very convoluted and hard to follow, however, it refers to the main rational for carrying out this study.

# This paragraphs will be improved.

5. I don't understand why there was not an attempt to estimate ks for the PLFAs. That would be the main purpose of this approach, in my view. Also, how can the VAM be building up 13C, if they are mycorrizhae? This probably suggest the marker was indicating a saprotroph, not mycorrhizae, which is known to happen in soils.

# The decomposition constants can be estimated in our study, however, only for the groups where 13C enrichment decreased. However, we aware that these values can not be obtained for all groups and thus, we can not make quality comparison. Yes, we

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agree that 16:1w5 can belong to saprotrophic fungi. This will be corrected in the text.

6. About the aminosugars, what we still don't know and doesn't get explored and discussed, yet, it would be the most interesting is: which builds up more per unit of C assimilated (this would be an indication of which may contribute more to SOC building), or in other words a measure of enrichment/recovery.

# Thank you for the comment. We can not estimate how much aminosugars were build per unit of C assimilated, because it is not possible to separate amino sugars of living microorganisms from one accumulated in SOM in composition of microbial residues. Only what we did, we estimated enrichment of amino sugars by 13C on the day 3: " To estimate the 13C enrichment into amino sugars of living cells, we first calculated the amount of amino sugars in the living MB pool based on the fatty acids content. Assuming that PLFAs are present only in living biomass, and that the ratio of fatty acids to amino sugars in living biomass is about 0.23 (Lengeler et al., 1999), we estimated the amount of amino sugars in living MB to be 0.20  $\mu$ mol g-1 soil fatty acids/0.23 = 0.87  $\mu$ mol g-1 soil. The estimated percentage of amino sugars in living biomass from the total amino sugar pool was 0.87/7.70 (total AS ( $\mu$ mol g-1 soil))\*100 = 11%. This estimate agrees with that of Amelung et al. (2001a) and Glaser et al. (2004), who reported that the amount of amino sugars in living biomass is one to two orders of magnitude lower than in the total amino-sugar pool. We calculated the 13C enrichment in amino sugars for the first sampling point, assuming that all replaced C is still contained within living MB after three days of glucose C utilisation. Total tracer incorporation into amino sugars consisted of 0.00071  $\mu$ mol glucose derived 13C in amino sugars g-1 soil/0.87 (µmol amino sugars g-1 soil)\*7 (mean amount of C atoms in amino sugars)\*100 = 0.57% of the C pool. Comparison of these data with the 13C enrichment into PLFAs and the cytosol allowed us to conclude that the replacement of the amino sugar C with glucose derived 13C in living biomass is two-fold slower than the replacement in PLFAs, and faster than in the cytosolic pool". This is presented in the discussion section.

7. Glucose is likely to behave differently to other C substrates, therefore I would restrict the title and Discussion to glucose (not carbon).

# Title was corrected: Glucose C turnover in cell compartments and microbial groups in soil. The discussion will also be corrected and only glucose C will be discussed.

8. The rationale for the difference found in turnover time between the cytosol and PLFA is really not convincing and not well supported. Both active and dormant organisms have a membrane and if they're not active they wouldn't be picking up the 13C.

# Thank you for the comment. The idea of the work was estimate C turnover time in various cell microbial pools: cytosol, PLFAs and amino sugars. Cytosol is always assume as the most labile pool and supposed that C here should have the fastest turnover. However, in our work it is clearly seen that is it not the case, and actually C turnover times here are much slower that in structural cell pools (such as PLFAs). Our data on 13C pool enrichment include information as about dormand as about active microorganisms (due to calculation was done relatively to the total C pool), and clearly show that more 13C is incorporated into cell membranes than in cytosol and also that C turnover is slower in cytosol, than in PLFAs. Concerning dormancy: even a dormant microorganisms repair their membranes and, thus, have a C turnover in them, whereas microorganisms can be survived without DNA reparation.

9. The explanation for why more 13C was in the bacterial than non-bacterial lipids completely ignores anything about their ecology or physiology.

# Thank you for the comment. The goal of the study was not to investigate the ecology or physiology of microorganisms, and for that purpose only low concentrations of glucose were applied. We were mainly concentrated on the turnover times of C in various cell pools and worked with microbial community which was already formed in the soil before we started the experiment.

10.Describing what fatty acids were more or less abundant (section 3.3.) is not re-

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ally informative as these data don't reflect absolute abundances and these patterns of abundance are more or less the same for a lot of soils.

# Thank you for the comment. We will reduce this section.

11. I would replace the term 'incorporation' with 'recovery' which seems to be what they calculated.

# Thank you for the comment. This will be changed in all entire text.

12.I find it surprising that soil moisture would be "essentially constant" across 50 days.

# The data for soil moisture are the following:  $25.7\pm1.2$  (3 days),  $23.3\pm1.3$  (10 days),  $21.4\pm0.7$  (50 days). This information will be added into the material and method section. Due to we did not sample the soil directly after the rain, the moisture variations were low.

Minor methods comments The rational for the amount of C added is not presented.

# The C was added in the amount to prevent any priming effects, as well as growth of microorganisms due to glucose application. The concentration was chosen to trace the natural pool of glucose in soil solution (Fischer et al., 2007), rather than stimulate the activity of microorganisms.

Not clear if the field collected soil column had or not vegetation.

# The columns had no vegetation by the collecting time, as well as when the 13C glucose was applied. This will be added into materials and methods section.

I don't understand the "assignment of fatty acids to distinct microbial groups by factor analysis".

# Factor analysis was used as classification method, with the main purpose to split the microorganisms which belong to one group (according to literature) into subgroups which behave differently in the soil. Based on the analysis, fatty acids which were loaded into one factor with the same sign (+ or -) and were related to the one bacterial or fungal group, were summed up together. In the end of the analysis only groups of PLFAs related to ether bacteria or fungi were presented. The information about which fatty acids belong to which microbial groups was taken from the literature (Zelles 1997).

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