

## ***Interactive comment on “Multi-isotope labelling ( $^{13}\text{C}$ , $^{18}\text{O}$ , $^2\text{H}$ ) of fresh assimilates to trace organic matter dynamics in the plant-soil system” by M. S. Studer et al.***

**Anonymous Referee #1**

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Multi-isotope labelling ( $^{13}\text{C}$ ,  $^{18}\text{O}$ ,  $^2\text{H}$ ) of fresh assimilates to trace organic matter dynamics in the plant-soil system

by

M. S. Studer, R. T. W. Siegwolf, M. Leuenberger, and S. Abiven MI.

General comments The paper deals with atmospheric multi isotope ( $^{13}\text{C}$ ,  $^{18}\text{O}$ , and  $^2\text{H}$ ) labeling of plant tissues followed by isotope detection of different plant compartments and soil. Generally, the paper is an example of "making simple things complicated". Instead, good science should make complex (complicated) things easily understandable.

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It would have been wiser to deal with isotope exchange and partitioning of the applied isotopes and proper discussion of their sources and fate rather than complicating the issue with not straight-forward models.

With respect to oxygen and hydrogen (water), the three water sources you have is the label (atmosphere), the soil and xylem. However, with respect to oxygen, a further source must be considered: From uptaken  $\text{CO}_2$  during photosynthesis. Thus, most of the oxygen of primary photosynthetates comes from  $\text{CO}_2$  and not from uptaken water. Therefore, the concept of the three labels is questionable as these sources are not linked to each other. While most of the  $\text{CO}_2$  goes into the photosynthetates, most of the water vapor goes into the plant water with subsequent or simultaneous isotope exchange without uptake.

The concepts of "maximum label strength" and labeled isotope ratios ( $^{18}\text{O}/^{13}\text{C}$ ,  $^2\text{H}/^{13}\text{C}$ ) are ambiguous and make simple things (isotope labeling with  $^{13}\text{C}$ ,  $^{18}\text{O}$  and  $^2\text{H}$ ) more complicated than it is. There are many more specific comments, which are given below.

To accept the paper for final publication in "Biogeosciences" I strongly suggest to simplify the whole story and to remove ambiguous concepts / data.

Specific comments P15913 L 12: Replace "molecular formula" by "molecular composition".

P15913 L15: You should include and refer here Fig. 3a.

P15913 L18: This "logic" is ambiguous as such can be done only using dual-labeled compounds e.g.  $^{13}\text{C}$  and  $^{15}\text{N}$ -labeled amino acids and detection of both isotopes in amino acids.

P15913 L22: Delete "of high specificity" as different organic molecules might be labeled with different isotopes in different plant organs.

P15913 L23: I disagree as your approach does not help for the detection of labeled plant

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material in (bulk) soil. Therefore, the whole argumentation has to be deleted.

P15913 L29: Write "plant compartments... and soil (organic matter?)".

P15914 L18: How can a leaf area of 6.5 m<sup>2</sup> correspond to only 3 g leaf (and stem) biomass? P15914 L24: What do you mean by "hermetical"? Is it only separated from the gases or also from the intrplant matter fluxes? The whole procedure is unclear. A schematic of the labeling chamber would probably clarify a lot.

P15915 L8: If pots are "hermetically" sealed, how can they be aerated?

P15915 L18: If air humidity and temperature is high, why water vapor should be taken up? Instead, transpiration of plants should be high.

P15915 L27: Unclear what that study has to do with this one. Either delete this sentence or explain the added values of the new study in comparison to the previous one.

P15916 L10: This is already discussion and should be moved there.

P15917 L12: Please give exact conditions of oxidation and pyrolysis (temperature, catalysts etc.). Please explain why 18O analysis was not undertaken by TC/EA.

P15917 L21: Unclear what you did and why.

P15917 L24: Precision of working standards is not of interest. Instead, please give accuracy (how did you do calibration ?) and precision of real (your) samples. How soil samples were treated and analysed?

P15918 L19: Three isotopes (13C, 18O, and 2H)!

P15918 L26: Please explain each single abbreviation in eqn. 1. Alternatively, delete eqn. 1 as you do not need it (see eqn 2).

P15919 L9: What about water already in the plant?

P15919 L10: Why don't you call it simply "soil water"? Why couldn't you simply use the isotope signature of your soil water for this variable?

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P15919 L12: What you measure is in the plant!

P15919 L19: Why don't you simply call it "atmospheric water"? Why can you not simply take the isotope signature of your labelling atmosphere for this variable?

P15919 L8: Not clear what you mean by "maximum label strenght". Therefore, I suggest deleting this section or referencing it to the maximum isotope labels.

P15919 L3: It is useless and wrong to give a d13C value for 10at% CO<sub>2</sub>. Please delete.

P15925 L3: Why back diffusion? Delete "back". The same applies for the rest of the paragraph.

P15925 L27: Can you exclude simple isotope exchange without water uptake by this approach?

P15926 L10: The three water sources you have is from the label (atmosphere), from the soil and from xylem. However, with respect to oxygen, a further source must be considered: From uptaken CO<sub>2</sub> during photosynthesis. Thus, most of the oxygen of primary photosynthetates comes from CO<sub>2</sub> and not from uptaken water. Therefore, the concept of the three labels is questionable as these sources are not linked to each other. While most of the CO<sub>2</sub> goes into the photosynthetates, most of the water vapor goes into the plant water with subsequent isotope exchange.

P15927 L6ff.: Tracing organic matter with this approach is not possible given the facts mentioned in the previous comment.

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