

Interactive comment on “Direct uptake of organic carbon by grass roots and allocation in leaves and phytoliths: ¹³C labeling evidence” by A. Alexandre et al.

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The manuscript “Direct uptake of organic carbon by grass roots and allocation in leaves and phytoliths: ¹³C labeling evidence” describes a hydroponic experiment investigating the uptake of ¹³C and ¹⁵N from three dual-labeled amino acids (L-Alanine, Phenylalanine, and Methionine) and ¹³C from ¹³C-labeled D-Alanine in tall fescue grass over 14 days. I find the reported data has significant shortcomings, which makes it unacceptable for publication (see below). I therefore recommend that the manuscript is rejected.

General comments: (A) The authors present an experiment where the control is un-
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replicated and the treatment has only two replicates. My experience with isotope-labeling studies is that even under very controlled conditions, there will always be variation among replicates. Therefore I am very surprised that the authors choose to conduct a study only with two replicates. The data presented by the authors gives the reader no chance to evaluate whether the conclusions are based on a coincidence or there are in fact differences between bulk plant and phytoliths as stated. The lack of replicates is a major weakness of the study.

(B) The authors refers in several places to “old soil C” and “microbial metabolites” (e.g. p. 19752 l. 8; p. 19754 l. 1; p. 19768 l. 9), but there is no justification given how the amino acids used make a fair representation of old soil C or microbial metabolites. Usually it is found that amino acids cycle very fast in soil - in the range of hours – therefore it is very surprised to see amino acids linked to “old soil C”. I disagree with the author’s indication in the conclusion that their findings support that plants take up old soil C and “store” it in Si-precipitates – in fact their data would then show that even young soil C is stored this way.

(C) The experiment that was carried studied the uptake of labeled amino acids in grass for 14 days. The authors state that they sealed of the labeling solution from the grass shoot – to avoid uptake of labeled CO₂ by photosynthesis, but the authors make no justification of the extent of intact amino acid uptake from the labeling solution. Both ¹⁵N and ¹³C can be taken up in their inorganic forms. In the results section the authors state that 4.5% of supplied ¹³C and 46.9% of supplied ¹⁵N was absorbed by the roots. This must mean that the majority of ¹⁵N was taken up in mineral form from the solution, which must mean that inorganic ¹³C (bicarbonate, carbonate) was also present in the labeling solution and hence could have contributed to the uptake of ¹³C by the grass.

(D) The authors state that they distinguish between labeling in plant amino acids and in phytoliths, but when I read the method-section it seems to me that analysis of plant amino acids and of phytoliths are two parallel analysis, which implies that in principle it could be the same isotope-labeling the authors measure in the two pool (i.e. the

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pools are overlapping). The authors describes ^{13}C enrichment in phytoliths, but make no attempt to determine what type of compounds are precipitated along with Si – in principle it could be ^{13}C in other organic forms than the added amino acids or even inorganic ^{13}C . Yet, the authors state that the amino acids are bound in phytoliths – they do not show this.

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