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Biogeosciences Discussions

Interactive comment on "An unknown respiration pathway substantially contributes to soil CO_2 emissions" *by* V. Maire et al.

Anonymous Referee #2

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With this work S. Fontaine and his co-authors challenge the broadly accepted understanding that respiration is carried out by endo-enzymes inside cells, through a series of well thought and generally well executed laboratory experiments. The strength of this work is in its logic (and pretty comprehensive) structure. With a sequence of experiments, the authors test/demonstrate: 1) the existence of an extracellular oxidative respiration pathway, that they name Exomet; 2) the stabilizing role of the soil matrix on Exomet; 3) the relative contribution of Exomet to total soil respiration and 4) the resistance of Exomet to factors that affect cellular respiration (i.e., high temperature, autoclaving and chloroform fumigation). The validity of this work is based on the realized sterility of the soils and water samples. The authors made a significant effort in proofing this true and convincing data on the sterility are also reported in the Supporting material. The ms is generally well written.

The results from this work, given the significance (16-48%) of the measured exomet relative to total heterotrophic soil respiration for the analyzed soils, call for new research to text exomet in more soils – they tested 4 - but also to better understand generally the role of enzyme stabilization in soil processes. In my opinion, this is a very interesting and paradigm-shifting work that deserves publication.

However, I do have a few comments/suggestions that will require some revision:

1) Authors should expand the discussion, in particular by providing some more in depth interpretation of the observed results. For example, the potential controls of Exomet are not at all discussed. The authors found that different enzymes had a different degree of stabilization (Why? What are the controls?), as well as that soils differing in land use and clay content showed different Exomet, with the lowest been found in the sandy soil under cropping management (again, why?). I understand that the data are limited and that there is a high risk for speculation, but I suggest that the authors point out a few treads that new research should follow.

2) I do not follow the basic assumptions behind the experiment to quantify the relative contribution of soluble and soil-immobilized enzymes to total enzyme activity (P8670 section 2.3.3). The authors assume that the activity after 5 minutes it's only from soluble enzymes, why? I understand that it will take longer for the enzymes to stabilize, but that would just say that it is from the total enzymes still all in solution. After that time, the enzymes remaining in solution decline, while those which stabilize remain active. My concern is further demonstrated by the fact that the initial activity of soluble enzymes is in fact higher than the total (Fig. 2) which obviously does not make sense. Also, if the soluble enzyme activity is not quantified at subsequent times, where does the dynamic shown in the figure comes from? The authors should reconsider their interpretation of this experiment or, if I missed something – which I think I did -, do a better job at clarifying the assumptions and procedure used.

3) The level of CO2 reached in the microcosms is very high for lab incubations, often

exceeding 10% (Fig 3). This may have inhibited CO_2 diffusion (and possibly production) from soils to the atmosphere in the microcosms where CO_2 was accumulated in the headspace (where it was trapped in soda it should have not affected CO_2 efflux). In fact the authors observed in one case that the O_2 had limited respiration. This inhibition would, if happened, actually represented an underestimation of Exomet. However, the authors need to discuss this potential problem in their analyses.

Specific comments:

P8666 L2: Delete soil names and change in "The five soils presented textures ranging from . . ."

P8666 L4: add "(Table 1)" after crops.

P8666 L4: Replace here and throughout the text "the soil of" with "the soil from"

P8667 L12: Add reference for Biuret method

P8667 L13: U MDH – this is for specialists – clarify

P8667 L29: The experimental units are defined "microcosms", but what are they: jars (as for one of the following exp) ?, vials? How big? Air tight? Dark, clear? The authors need to provide a clear description of the physical structure of the microcosms.

P8667 L29: add the sentence: ", following the methods described in section 2.6." after incubation

P8668 L13: Why for this experiment incubation was at 20C, while all others were carried out at 30C?

P8668 L22: Add the sentence "repeating the above experiment but only . . ." between "content" and "using"

P8670 L24: Add reference for the calculation of half life.

P8671 L18: Correct "Rl=" with "Ri=" in the second equation.

P8671 L21: Indeed it is simple algebra, but for clarity the authors could add after Rx "Thus, k can be obtained from . . ..".

P8673 L15: add the sentence: ", following the methods described in section 2.6." after measured

P8674 L2: Was titration done manually or automatically, please specify, and in the latter case add the model of the instrument used for titration.

P8674 L3: To my knowledge CO_2 in air is measured by either IRGA or by Gas Chromatography (with an appropriate detector). What do the authors mean by "Gas spectrometry"? Please clarify the method and provide the model for the specific instrument used.

P8674 L21: Why native stabilized enzyme do not benefit from glucose additions? Are they not C-limited? This result have several potential implication for soil C cycling in the natural environmental and the authors should do a better job at highlighting this result and discuss it.

P8675 L21: Correct "incbation" with "incubation"

P8675 L24: Add "sterile" to read "from sterile control soil.."

P8676 L14: See general comment (1) above. This is a large variation – the authors should discuss it and speculate on some possible explanations.

P8676 L23: Provide actual CO_2 values besides percentages.

P8677 L2: Again, see general comment (1) above. The authors should discuss the variability observed between soils and speculate on some possible explanations. A more general discussion may also fit well at P8678 L5, before "Finally".

Table 1: Order the soils by a criterion (e.g. alphabetic, land use, clay content), any would do as long as there is one. Soro is not spelt correctly. It is a Danish name and the correct spelling is "Sorø", please correct. Delete "(-)" after pH. The SI unit for CEC,

is centimol which is written “cmol+” and not “Cmol”, please correct.

Fig.3 To my knowledge if CO_2 is given as a concentration, % or ppm are used as units. Thus I suggest deleting “atm” from the y-axis labels.

Interactive comment on Biogeosciences Discuss., 9, 8663, 2012.