

Interactive comment on “Density fractions versus size separates: does physical fractionation isolate functional soil compartments?” by C. Moni et al.

Anonymous Referee #2

Received and published: 14 September 2012

The paper aims at evaluating physical fractionation procedures of soil organic matter (SOM) with regards to their ability to isolate functionally different but unique (non composite) soil organic matter pools with a homogenous decay behaviour. For their evaluation of physical fractionation procedures the authors select two fractionation procedures (I) a fairly new aggregate density fractionation (ADF) and (II) a traditional and widely used particle size-density fractionation (PSDF). The two physical fractionation procedures were applied to two Cambisol soils under managed beech forest, one in France and another in Germany and samples were collected 8 resp. 12 years after application of ^{15}N enriched labelled litter. Fractions are characterized by their C- and N-contents, C/N ratio, $\delta^{13}\text{C}$ - and $\delta^{15}\text{N}$ -values as proxies for the degree of microbial processing. Fractionation procedures are compared and characterized through a standardized principal component analysis (PCA) to identify two independent variables

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that account for the majority of the data variability. To visualize dynamics of ^{15}N label incorporation, a contour map representing excess ^{15}N was applied in the plane defined by the main variables. As conclusion the authors suggest an improved ‘economized’ fractionation procedure that ‘only retrieves meaningful fractions’.

The presented dataset and especially the statistical analysis are very interesting, innovative and valuable.

However the overall approach is inconsistent: A clear structure is missing in the paper which should be a guide to evaluate the implications of the applications of different fractionation protocols. While some methods are introduced with dense information other methods are only mentioned (e.g. fractionation by particle size versus aggregate density fractionation). Some parts of the introduction and the discussion are very detailed with lots of complex issues but little clearly transparent context.

The intention why the authors selected two specific ‘standard methods’ for their examination despite they addressed in the introduction methodical limits do not become clear. Please state more clearly your objectives and your hypotheses! The points ‘lack of knowledge’, ‘hypothesis’ and ‘new findings’ just do not come out clearly enough. Shortening and focusing of the text would help greatly.

The application of the two standard methods led to already well known results (Chapter 3.1 to 3.5). Chapter 3.2 demonstrates that the obtained fractions are not pure and statistical analysis was applied to evaluate ‘mixed’ fractions. This part is interesting, new and informative and the potential of such statistical analysis and possible applications could be better highlighted and discussed in the manuscript! Variables used for PCA are proxies for the degree of microbial processing and I regret that no unique indicators could be used. Further, following the dynamic of ^{15}N label incorporation basing on the relative enrichment ($E^{15}\text{N}$) can be biased in the field by ‘dilution’ effects. The dynamic should instead be based on the mass-losses or mass increases of ^{15}N . The discussion and the evaluation should take these issues into account and should

be done more cautiously.

The proposed new fractionation procedure seems promising but no generalization should be made by the examination because only one soil type (Cambisol) was used and serious discussions in view on different soil properties (e.g. different textures, different carbonate/dolomite concentrations...) and of comparable examinations in the literature are missing. Density cut-offs and intensities of dispersions should be tested and discussed on a broader set of soils.

Specific comments:

In Chapter 1.2, Chapter 1.3 and Chapter 2 important details as well as references are missing that are necessary for a critical evaluation:

Chapter 1.2: Description of advantages/disadvantages of the fractionation procedure by size: please add references!

Chapter 1.3: Generally: For each method add a statement with regards to the potential usefulness of the method. Aggregate density fractionation: Methodological points of critique are not mentioned and discussed. E.g. medium densities do not separate 'true aggregates' but also light organo-mineral complexes and/or very small-sized particles.

Chapter 1.4: Please state more clearly your objectives. Why did you select the two fractionation procedures (although you described in detail their limitation)? What do you intend?

Chapter 2.1: "highly enriched 15N litter": Please tell in more detail. Please summarize the results by Hatton et al. (2012) that are necessary to understand the experiment.

Chapter 2.2.1: "Assuming an average particle size density of 2.44 g cm⁻³": Please explain in more detail and add references. Describe in more detail and more objectively how fractions coarser than >6 μ m were separated: "repeatedly soil fractions were gently swirled in..." : How often and how long and how did you shake the solution? How much time was needed for sedimentation. . .

C3991

Chapter 2.2.2: Please add a short description of the method proposed by Sollins et al (2006) and the usefulness of the method. What did you intend?

Important literature is missing. E.g.: Kaiser, K., Guggenberger, G., 2007. Distribution of hydrous aluminium and iron compounds over density fractions depends on organic matter load and ultrasonic dispersion. *Geoderma*, 140: 140-146. Cerli, C., Celi, L., Kalbitz, K., Guggenberger, G. and Kaiser, K., 2012. Separation of light and heavy organic matter fractions in soil - Testing for proper density cut-off and dispersion level. *Geoderma*, 170(0): 403-416.

Interactive comment on Biogeosciences Discuss., 9, 8405, 2012.

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