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Interactive comment on "Dissolved organic carbon lability and stable isotope shifts during microbial decomposition in a tropical river system" *by* N. Geeraert et al.

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We would like to thank Referee #2 for the constructive feedback. The comments are reiterated below, each time followed by our reply.

Major comments and remarks:

REF: At current, I found part of the results and discussion section ('removal mechanism and origin of DOC') not to be well based on the data provided. This concerns primarily the discussion of the C3/C4 vegetation differences and how these would affect the lability of DOC. I believe that the authors need to provide more evidence here, or to

cut back on their interpretation and conclusion. In this context, I wonder if there is at all any data that objectively suggests any type of relationship between stream dDOC, d13C and the C3/C4 landcover data?

REPLY: We launch the hypothesis that the shift in stable isotope ratios we observed during microbial decomposition is related to differences in the contributions of C3 and C4 vegetation, because this shift appears to be observed only in such mixed systems. This can indeed not be unequivocally demonstrated by our limited dataset. Therefore, we have reformulated the abstract by explicitly mentioning it as a hypothesis. In the Conclusions section, we emphasized that there was a different decomposition rate between DOC with heavy and light isotopes, without stating that the difference is a consequence of the mixed vegetation.

REF: Statistical methods are explained within the combined results and discussion section. I strongly suggest describing these in the methods section. At current, there are results of the stats presented, without the reader knowing what stats methods were actually used.

REPLY: We added a paragraph about the statistical methods in the methods section, stating the statistical program and the applied statistical tests.

REF: I generally prefer to have separate results and discussion sections. However, I acknowledge that this may only be my personal preference and the authors have prepared the manuscript now in the given format. Therefore, I will leave it up to the editor to decide, whether separating results and discussion is feasible and will increase the quality of the manuscript.

REPLY: In initial versions of the manuscript which had separate results and discussion sections, the results had to be repeated before interpreting them in the discussion section. To avoid this redundancy, we have decided to combine those sections.

Minor comments:

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REF: P12764, L4: I was a surprised that it is stated that 'microbial consumption can take place in the entire water column'. Whereas this statement is, as such, true, there is no mentioning of the important role of the benthic system, which may also host microbial biofilms that can greatly enhance heterotroph activity. I suggest to add a sentence or two on this topic. A reference could be (Battin et al. 2003), but there are other good ones as well.

REPLY: We added a sentence, mentioning their importance for the DOC dynamics. We refer to Battin et al. (2003) and Romani et al. (2004), whereby the latter one is more relevant for larger river systems such as the Tana River. However, as the importance of the benthic compartment is very dependent on the characteristics of the river, we did not go further in detail.

REF: P12765, L14-18. It may help the reader to understand, which parts of the catchment can be considered humid and which arid (or semi-arid). This aspect may be also important for the question of how the landscape contributes to stream DOC.

REPLY: We added the geographical directions (northwestern and eastern, respectively) to the description of the study area, together with the link to the map (Fig. 1).

REF: L21: interesting approach this mixing model for the landscape C3/C4 proportions. Maybe it would help to guide the reader to why the authors apply this model. A sentence like the following could be added: "To investigate the possible effect of vegetation cover on DOC isotopic composition. . . we estimated C3/C4 vegetation coverage". However, before writing this, the authors may need to clarify the necessity of this vegetation cover data for the study for themselves.

REPLY: We have changed the first sentence of the paragraph in order to clarify that the vegetation can affect the riverine organic C.

REF: L25: Interestingly these d13C numbers are pretty close to those named as 'typical numbers' for d13C numbers of CO2 in soil (-23‰ and -9‰ for C3 and C4 plants,

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respectively) named by Clark and Fritz (1997). May be worth to note this somewhere here.

REPLY: The values we are using were those used by Still and Powell (2010) to convert the maps with %C3 and %C4 vegetation cover to maps representing the averaged vegetation δ13C. This has been reformulated more clearly in the manuscript.

REF: P12770, L5: please reconsider the presentation of statistical results. Were all assumptions for a t-test (normality, homoscedasticity) met here?

REPLY: We changed throughout the manuscript to the non-parametric paired Wilcoxon rank test because the normality assumption was not always met. This did not affect the interpretations about the parameters being significantly different or not.

REF: L25: as not all readers may be so familiar with the selective photochemical oxidation, I suggest adding the reference that the authors cite in the introduction.

REPLY: The reference to Opsahl and Zepp (2001) has been added.

REF: P12771, L2: The idea of selective decomposition is truly interesting. Maybe the statement that 'isotopically heavier carbohydrates were preferentially decomposed' could be evaluated and discussed a bit more. Also, it is unclear to me, based on which data the authors come to this conclusion. Please provide more detail.

REPLY: This idea is a hypothesis, since our data show the opposite of what has been observed during photochemical oxidation, i.e. an increase in δ13C due to preferential decomposition of 13C-depleted lignin relative to the 13C-enriched carbohydrates (Opsahl and Zepp, 2001; Introduction P.12764 L. 23-25). This has been rephrased by stating that it is a hypothesis, which is countered in the remainder of the paragraph as this isotope shift was not observed in other river systems, with the exception of other mixed C3/C4 catchment systems.

REF: In some older literature one can read that 'bacteria prefer to metabolize the isotopically light organics and oxidizers [. . .]' (Clark and Fritz 1997), as it is easier to break

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12C-H bonds than 13C-H (or C-2H). This is generally assumed to cause the opposite effect as the one described above. So here is truly an interesting aspect to explore. But first, the reader needs some more evidence for a relationship of a DOC source and the C3/C4 story.

REPLY: It is plausible that bacteria prefer the compounds with isotopically light C, even though this has not resulted in an isotopic shift in riverine DOC in other rivers, except two other tropical rivers. We are not able to provide evidence of the linkage with C3/C4-vegetation, but we think our observations elucidated a pattern that can guide further investigations.

REF: P12772, L1: Even if I have not been to the Tana River, I am not sure these are all the potential sources of DOC to this system. You may also consider i) additions of leaf litter from riparian vegetation that can enhance POC, but also DOC for example through leaching or ii) any human activities, such as sewer inflows that may also contain organic matter. On the contrary, groundwater appears to me like an unlikely source of DOC to the river, as this is commonly considered to be low or very low in DOC, but often high in pCO2. Also, this point comes back to my first main comment.

REPLY: We expanded the list of sources by mentioning human activities, even though those are unlikely to provide significant amounts of DOC as the population density is very low and, to our knowledge, there are hardly any continuous sewage inputs. We expanded the source of ground water to 'groundwater and subsurface water inputs through leaching of DOC from leaf litter', as this is more how we interpreted the groundwater, even though it was not accurately formulated.

REF: Figure 2 and 3: They appear a bit redundant, as they show almost the same thing. I wonder if these could be combined or if one of them could be removed(?).

REPLY: Although the figures are indeed constructed in a similar way, the messages they should convey are different. In Figure 2, the focus is on the contrast between the two methods (filtered vs. unfiltered). In Figure 3, we wanted to emphasize the strong

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decrease in concentration and δ13C. The inclusion of the data of 2014 is important, because the hydrological conditions were different from 2013 (flooded vs. non-flooded).

REF: Figure 4: First part of caption reads strange. It's the percentage of change of the initial . . .

REPLY: The caption has been changed to: "Relative change (in %) between the initial concentration (day 0) and final concentration (day 8) in function of the initial concentration of the DOC.

REF: Figure 5 and associated results (p12770, L12-16): I believe this is a typical example, where the use of a simple regression based on least squares fitting is not a good choice. The authors acknowledge this, as they present two such regression models. However, two regressions don't make much sense here. Instead the authors should reconsider their approach and use one of the commonly used 'robust regressions' to account for the two possible outliers.

REPLY: We have reconsidered the approach, as the second regression curve indeed doesn't provide more information than the first one. We now have a robust linear regression by using an M-estimator.

References:

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Opsahl, S. P. and Zepp, R.G.: Photochemically-induced alteration of stable carbon isotope ratios d13C in terrigenous dissolved organic carbon, Geophys. Res. Lett., 28, 2417–2420, 2001.

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Interactive comment on Biogeosciences Discuss., 12, 12761, 2015.