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## ***Interactive comment on “Stable carbon isotope biogeochemistry of lakes along a trophic gradient” by A. de Kluijver et al.***

**A. de Kluijver et al.**

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We thank the reviewers for the time to review the manuscript and the comments that helped us to improve the manuscript. The suggestions and comments (s)he raised are answered below.

Reviewer 1: Referee: This paper summarizes efforts to characterize a series of lakes of various trophic states in terms of carbon utilization and C-stable isotope composition. The paper is reasonably well written and organized; presenting an interesting dataset. The authors have attempted to develop a detailed dataset of the compartmentalization of C (concentration and isotopically) in variety of pools in the lakes studied. However, the evaluation of these data is not clearly presented such that the discussion and conclusions drawn are not readily interpretable. I would urge the authors to re-write the

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discussion in a form that conveys the “story” of the carbon in their systems in clear and concise fashion.

Reply: The reviewer is not very specific in which sections have to be rewritten. However, we will incorporate the suggestions made below. Our cross-system carbon isotope study contains some explorative aspects besides question-oriented approaches. The results section is organized by type of analysis, while the discussion sections address not only topics of current interest (isotopes and lake metabolism, the isotope composition of allochthonous material and phytoplankton), but also a discussion on the isotope differences among various organic pools (carbohydrates and lipids).

Referee: 1) The site descriptions lack any information on the geologic setting. The title of the article contains “biogeochemistry” but the paper gives very little “geo” background. Are there sedimentary exposures (carbonate minerals) near the field sites that can influence the C pools?

Reply: Both research areas were substantially impacted by the same glacial episode ca. 12 000 ybp and are covered by thick till from the same source areas (Grimley 2000). There are no exposures of carbonate minerals in the area that could influence C pools. This information will be added to the MS.

Referee: What are the  $\delta^{13}\text{C}$  values of surrounding soils, vegetation, sediments, etc.? The website links only have water quality data and no specific site information.

Reply: In this region, the soil and lake sediments should broadly reflect the vegetation type and/or forest composition (Collins et al 2000; Dzurec et al 1985). The  $\delta^{13}\text{C}$  of vegetation has been analyzed and is part of the MS.

Referee: 2) What time of day were the samples collected? Did the authors take diel effects into account? (e.g., Pokrovsky and Shirokova, (2013) *Water Res.* 47, 922-932; Ziegler and Fogel, (2003) *Biogeochemistry*, 64, 25-52.

Reply: Water samples were taken between 10h-16h, a period of the day that yields

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relatively stable water chemistry readings in these lakes. This information will be added to the MS.

Referee: 3) Do any of these lakes have anoxic zones near the bottom? Methanogenesis from disproportionation of acetate produces isotopically light CH<sub>4</sub> and enriched CO<sub>2</sub>. This can have a significant impact on the d<sup>13</sup>C of both DIC and DOC.

Reply: Anoxic hypolimnia are rare in these lakes, either due to low nutrients or polymixis. The fatty acid composition in seston did not indicate methanogenesis, i.e. we did not find fatty acids typically occurring in methanotrophic bacteria nor <sup>13</sup>C-depleted fatty acids. This will be discussed in the revised MS.

Referee: 4) Water samples were all collected at one depth and this may not be the best representation of the overall biogeochemistry of the lakes being investigated. The authors should consider Gammons et al. (2013) App. Geochem. 36, 57-69 for a further examination of the changes in DIC and d<sup>13</sup>C-DIC with depth in an oligotrophic lake.

Reply: Samples were taken at a position that has been determined in past analyses to broadly reflect the average spatial conditions in the lakes. This will be added to the discussion

Referee: 5) Line 20: is it mercuric or mercurous chloride?

Reply: it's mercury chloride or mercuric chloride

Referee: 6) Lines 12 & 13: field rather than laboratory pH should be used since this variable can change due to degassing of CO<sub>2</sub> in samples that are oversaturated.

Reply: We had better trust in lab than field pH and pH was analysed within the same day after sampling. The overall results did not differ much.

Referee: 7) Line 6: "simply" states: : :: : :: : ...

Reply: This will be changed

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Referee: 8) Line 26: a “relatively” larger: : .:

Reply: This will be changed

References Grimley, D. A. (2000). Glacial and nonglacial sediment contributions to Wisconsin Episode loess in the central United States. Geological Society of America Bulletin, 112(10), 1475-1495.

Collins, H. P., Elliott, E. T., Paustian, K., Bundy, L. G., Dick, W. A., Huggins, D. R., ... & Paul, E. A. (2000). Soil carbon pools and fluxes in long-term corn belt agroecosystems. Soil Biology and Biochemistry, 32(2), 157-168.

Dzurec, R. S., Boutton, T. W., Caldwell, M. M., & Smith, B. N. (1985). Carbon isotope ratios of soil organic matter and their use in assessing community composition changes in Curlew Valley, Utah. Oecologia, 66(1), 17-24.

Reviewer 2: Authors have analyzed or recalculated isotopic composition of carbons pools (DIC, DOC, POC, Phytoplankton, Zooplankton, Bacteria, detritic fraction) of 22 North American lakes displaying various trophic status. They try to link isotopic composition to allochthonous vs. autochthonous contribution to carbon pool and to trophic status. I have found this paper very good, well written, bringing new insight on carbon metabolism and cycle in lakes. I like the multi-approach concerning stable isotopes methodology (bulk measurement, GC and LC-Irms measurement, calculation of sources contribution using mixing model). I am impressed by the amount and the quality of the isotopic data. I do not have any major comment on this ms and recommend its publication like this. Very minor comment: Accordingly to Fry (2006), I do not like the use of the term “isotopic signature”. For me, a signature is invariable and always allow to distinguish between two things (persons for examples). It is not the case of stable isotopes composition which are variable and, unfortunately, are not always unique to a pool/sources,etc.

Reply: Thank you, we will replace signatures by composition.

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