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Interactive comment on "Increasing addition of autochthonous to allochthonous carbon in nutrient-rich aquatic systems stimulates carbon consumption but does not alter bacterial community composition" *by* K. Attermeyer et al.

Anonymous Referee #2

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bg-2013-386 Increasing addition of autochthonous to allochthonous carbon in nutrient-rich aquatic systems stimulates carbon consumption but does not alter bacterial community composition Attermeyer et al

The authors describe a main experiment where two sources of DOC (tree leaf and phytoplankton leachates) were added to lake water in different ratios to study the effects on bacterial respiration, growth and community structure in lake water. There are also two additional experiments described in the supplement to test the effects of pulsed additional of leaf DOC on bacterial carbon processing and light induced DOC

degradation. DOC cycling in lakes is an important issue in the global carbon cycling and utilization of allochthonous DOC in the microbial foodweb currently receives a lot of attention. DOC was characterized by a chromatographic method (LC-OCD-OND) , bacterial communities were studied by DGGE and PLFA analysis and 13C ratios of DIC and PLFA were used to estimate respiration and assimilation based on the two DOC sources. The main findings are that: 1. allochthotous (leaf) DOC was more available for bacteria then autochthonous (phytoplankton) DOC and supported higher growth efficiencies. 2. In mixed incubations, increased addition of phytoplankton DOC to leaf DOC increased degradation. Phytoplankton DOC was preferentially assimilated and leaf DOC was respired more. 3. Single DOC sources gave similar bacterial community structure, but mixtures showed a shift with increasing phytoplankton DOC. And they conclude that chemical quality rather than source of DOC determines bacterial DOC turnover. I found that this conclusion is rather an open door and their results may not be directly translated to lakes as they are based on the DOC sources used which may not represent the natural DOC sources (see below). In lakes, the main sources terrestrial and phytoplankton have specific DOC compositions, which is thought to determine their fate in lakes. So, there is a direct relation between source and composition or quality.

There are issues with the two sources of DOC were used: namely leached leaf litter representing allochthonous DOC and ‘’algal’ hydrolysate for autochthonous DOC. There is relatively little information on the procedures used to make the two sources. It is for instance not stated if the leafs were green fresh or brown senescent, which may have important implications for the composition of the leached DOC. Also, the methods used to extract DOC from the algae are poorly described/lack information and look rather severe (a combination of dissolution in distilled water, autoclave, and ultrasonication). The methods description for algal DOC should be improved: why a mixture of 2 cultures and what was the ratio between these two cultures; why was the treatment so harsh and different from the leaf DOC extraction; how much of the biomass was extracted as DOC; how much material was used per volume of water. It also seems to me

that this harsh treatment will not likely extract DOC of similar composition as released by natural phytoplankton, which may hamper the translation of the results to the natural environment. For instance, the phytoplankton DOC contained a high amount of aromatic HS (humic substances), which seems rather unusual for natural autochthonous DOC from phytoplankton. And, DOC leaf was more available to bacteria than DOC phytoplankton in single source incubations, which is not the case for natural source materials. Given that the composition of the two DOC sources may not be representative natural autochthonous and allochthonous DOC inputs into lakes, it is difficult to tell to what extend the results can be directly translated to bacterial carbon cycling in lakes. The authors should fully acknowledge this in the paper.

DOC algae is used for the autochthonous DOC, but this is a bit odd as it was extracted from a mixed green algae and a cyanobacterium culture. Clearly a cyanobacterium is not an algae, and it should be renamed to DOC phytoplankton or similar. There are other issues with acronyms: HS, LMWS, HMWS are only explained in the supplement, and BR and BP (page 14279) are not explained anywhere as far as I could tell.

Bacterial community structure was determined by both PLFA and DGGE analysis. Both show that community structure was similar when individual DOC sources were used and that a shift was only observed with mixed DOC. This last observation makes sense, but the similarity in community structure between single source incubations is difficult to explain given the difference in composition between leaf and phytoplankton DOC. Also, what was the initial community composition of the lake water inoculum? Was this determined and was it similar to the single source incubations? It seems that at least the PLFA data are available based on the Fig 4 y-axis. Finally, 4 data points are shown for the leaf DOC incubations, whereas triplicate incubations were used?

I felt that the discussion lacks clear structure and various issues are discussed repetitive. The authors should have another go at the structure of the discussion to improve clarity. Also, a substantial part of the discussion and conclusions appears to be based of non-significant effects. Why? This should be treated with a lot of care eg. this should

not make it to the abstract.

These publications also studied the effects of DOC source on bacterial community structure and may be included: Roika et al. 2012 Aquatic Sciences 74: 513 Jones et al 2009 Environm. Microbiol. 11: 2463 And Kritzenberg et al 2005 Aquatic. Microbial. Ecol. 38: 103 studied the relationship between DOC source and growth efficiency.

Specific comments. * The abstract is rather long and contains a lot of introduction * Page 14264 line 15: . . .., which is related to rising CO2. . . * Page 14265 line 7: . . .., the effect of DOC quantity. . .. * 2.1: please move the third paragraph up one paragraph * Page 14269 line 12: this paragraph fits as a sub-chapter (Calculations or similar) after 2.5, as 2.5 describes some of the analysis used in the formulas. * Page 14269 line 25: Keeling plots are specifically used for atmospheric concentrations; this should be described as an isotopic mixing model. * Inoculum is typically used for microbial additions and not for DOC (heading 3.1). * Page 14272 line 4: . . . a better comparison between treatments, ... * Page 14273 first paragraph: this is difficult to follow and repeats the data shown in the figure. Please discuss in terms of major findings. * Page 14273 line 25: Only bacterial and total PLFA are discussed. However incubations were done with a day/night cycle; were any phytoplankton derived poly-unsaturated PLFA detected? And how would this influence the results. * Page 14274 line 4: . . .. efficiency (BGE) was . . .. * Page 14274 line 4: Please state for how many incubations respiration could not be estimated. * Page 14275 line 8: Delete “To answer this inquiry,”. * Page 14275 line 24: If differences were not significant, why are the data discussed extensively here and why are some of the conclusions based on these data? See also page 14279 line 14 and page 14280 line 19. * Page 14276 line 13: Delete the However. * Page 14277 line 21: FAME should be changed to PLFA throughout the manuscript. Bacteria do not contains FAME but PLFA. * Page 14278 line 11. I don’t get this sentence: Degradation of DOC supported high bacteria biomass, which in turn led to higher degradation of DOC? It is one or the other. * Page 14278 line 20. . . .. Besides nutrient limitation, ... * Page 14279 line 1: . . .. the question remains . . .. *

Page 14280 line 24: a higher influence? Should read a larger influence and explain to which this is compared * Fig 1. The DOC amounts in the hatched bar are difficult to read * Fig 2. Molecularity should read Molecular weight

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Interactive comment on Biogeosciences Discuss., 10, 14261, 2013.