

# ***Interactive comment on “Use of flow cytometry and stable isotope analysis to determine phytoplankton uptake of wastewater derived ammonium in a nutrient-rich river” by Calla M. Schmidt et al.***

## **Anonymous Referee #1**

Received and published: 8 August 2017

General comment :

This study aims to trace the dissolved inorganic nitrogen source primarily used by phytoplankton in a river impacted by an heavy anthropogenic nutrient (ammonium) enrichment. The authors report an interesting dataset of stable nitrogen isotope ratio measurement in several inorganic and organic nitrogen pool. Moreover, they used a novel and elegant method (combination of flow cytometry cell sorting with stable isotope analysis) in order to distinguish (healthy) phytoplankton cells from the bulk particulate organic matter. The manuscript is well-written and the results reported by the

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authors are appropriately discussed, overall I greatly enjoyed reading this manuscript.

Specific comment :

\*p4, line 26 : No results from a  $^{15}\text{N}$ -labeled nutrient uptake experiment are described in this manuscript, hence I would suggest to remove this from the material & method section.

\*p5, line 1-5 : Please, provide more information about the methodologies used to measure  $\text{NO}_2^-$ ,  $\text{NO}_3^-$  and  $\text{NH}_4^+$  concentration ( the chemistry behind).

\*p5, line 6 : I don't understand to what "70 $\mu\text{m}$ " is related. As you certainly know, nominal pore size of the GFF filters is 0.7  $\mu\text{m}$ . typo ?

\*p8, line 6-19 : I would suggest to plot the  $\text{d}^{15}\text{N}$ - $\text{NO}_3^-$  and  $\text{d}^{15}\text{N}$ - $\text{NH}_4^+$  data in a way similar to figure 3 (ie. Data plotted against travel time). These data are interesting, but it is difficult to visualize the trend when looking at table 1 only.

\*p9, line 25 : typo : fluorescence, and not florescence.

\*p10, line 2 : Higher importance of labile POM mineralization in the +EFF parcel seems indeed plausible. It is a bit unfortunate that you did not measure heterotrophic bacteria abundance, but did you measure dissolved oxygen concentration, or any other variable related to ecosystem metabolism (for instance, community respiration) ? They might be helpful to directly put in evidence a putative higher importance of heterotrophic metabolism in the +EFF parcels.

\*p10, line 16 and below : I understood reading your paper that the diatom "health status" was decreasing downstream, then could it be hypothesized that a change in the composition of the phytoplankton assemblage downstream of the location where the effluent enter the Sacramento river explains the gradual increase in the contribution of  $\text{NH}_4^+$  ? Did you look at the phytoplankton composition (and assess its variability) at several location during the travel of the two parcels of water downstream ?

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\*p10, line 16 and below : Beside  $\text{NH}_4^+$  and  $\text{NO}_3^-$ ,  $\text{N}_2$  fixation could also be a significant N source in systems where cyanobacteria are abundant. Do you know what was the contribution of cyanobacteria to the phytoplankton assemblage ? Could you explain why you rule out any contribution of  $\text{N}_2$  fixation as a N source in the Sacramento river ?

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