Response to reviewer #1. The reviewer's comments are presented in italics, and our responses are shown as plain text. We have responded to all of the reviewer's comments.

Drake et al. present a study on the role of plant exudates and their elemental composition (C and N) on growth and activities of microorganisms living into plant rhizosphere. Microbial activities are characterized by measurement of microbial respiration and enzymatic activities involved in the degradation of labile and recalcitrant organic matters. For their study they adopted an elegant approach combining theoretical modeling and experimental test in field conditions. Their results point on the importance of stoichiometric constrains of rhizosphere micro-organisms and show that the simultaneous exudation of C and N in rhizosphere triggers a larger stimulation of microbial activities and growth than the exudation of C compounds alone. This microbial stimulation may then release N from SOM for plant uptake. Thus, these results can partly explain (from a life strategy point of view) why plants generally exude some N-rich compounds (like amino acids) though they are often N-limited. Thus, these findings are timely and of interest for communities of ecologists and biogeochemists. That being said I have also got some important reserves on the experimental plan, results and their interpretation.

The reviewer has summarized our manuscript quite nicely. We appreciate his or her careful reading and synthesis, and responding to the reviewer's reservations has improved and clarified our manuscript.

First, the experimental plan lacks of a +N control. Thus, it is possible to know whether the impact of "C + N treatment" on microbial activities is due to the delivery of N or both elements (C and N). The delivery of N alone could explain the observed decrease in activity of enzymes involved in decomposition of recalcitrant SOM (i.e. Bowdenet al., 2004; Sierra and Nygren, 2005; van Groenigen et al., 2006) and the observed stimulation of microbial growth and activity of enzymes involved in decomposition of labile OM (see the prolific literature on N limitation of soil decomposers). The problem of lack of N control should at least be discussed.

The reviewer is correct- we did not include a +N treatment in our experimental design. We included four treatments: a disturbance control, a +water control, a +C treatment, and a +C and N treatment. We did not include a +N treatment partly in response to monetary constraints on the number of peristaltic pumps we could acquire to deliver the exudate solutions, but the primary reason for our decision was that a +N exudation treatment is not biologically relevant. That is, roots do not exude N in isolation; N is only exuded in combination with C. Most studies of root exudation have only been concerned with C compounds and have not considered N exudation (e.g., Jones 1998, Ryan et al. 2001, Bertin et al. 2003, Farrar et al. 2003, Phillips et al. 2011). Additionally, model simulations showed no response of microbial biomass, respiration, or enzyme activity to additions that contained only N (data not shown), as microbes were always C limited prior to exudate delivery in the model scenarios (positive rates of N mineralization are indicative of C limitation, Fig. 2m-p). Thus, given limited resources, we elected not to include a +N treatment.

The reviewer references literature concerning microbial responses to long-term N addition studies simulating atmospheric N deposition. There is a large body of literature demonstrating that surface additions of inorganic N often reduce microbial biomass, soil respiration, exo-enzyme activities, and decomposition rates in temperate forests, leading to the accumulation of soil organic matter (SOM), particularly in the organic horizon (Saiya-Cork et al. 2002, Wallenstein et al. 2006a, Treseder 2008, Janssens et al. 2010, Thomas et al. 2012). A recent review largely attributed these effects to a reduction in belowground C allocation by forest trees, which reduced microbial biomass through a loss of priming (Janssens et al. 2010). The reviewer is correct that our observations of reduced lignolytic exo-enzyme activity in response to +C and N are consistent with this literature, suggesting that our observations may be driven by N alone. However, the balance of our observations are not consistent with this interpretation; we observed increases in many indices of microbial activity in response to the +C and N treatment, including microbial biomass, respiration, and labile exo-enzyme activities, while the literature on surface N additions largely show reductions in these variables (references above; but see Saiya-Cork et al. 2002). Thus, we suggest that our interpretation of the microbial response to +C and N is supported more strongly by the observed data. Additionally, we added very little N relative to this literature on N fertilization. Studies of the microbial response to N fertilization have added an average total load of ~1000 kg N ha^{-1} (Treseder 2008); we estimate that we added < 0.1% of this amount. **We have added a paragraph to the discussion to address these points, as suggested by the reviewer.**

Second, authors only discuss the idea that soil microorganisms needs C AND N to growth and secrete enzymes (that is, stoichiometric contrains of microorganisms) signifying that the delivery of N in rhizosphere by plants is essential for an efficient stimulation of rhizosphere microorganisms that could mineralize soil organic nitrogen releasing soluble N for plants. However, there are numerous studies that also show that low nutrient availability favors microbial mining of N in recalcitrant SOM (i.e. Fontaine et al 2004, Carney et al, 2007). These studies appear to be in contradiction with your model and experimental results, but it might be not the case. Try to reconcile these results by proposing a "global theory" in the discussion section; for example it is possible soil microorganisms need a minimum N availability for growing and producing enzymes, which you could explain your results. However, when nutrient availability exceeds a threshold the supplemental delivery suppresses the mining activity of soil microorganisms and stimulates microbial communities that do not degrade SOM (Fontaine et al., 2003).

The literature cited by the reviewer demonstrates that priming of SOM decomposition is a process that actually occurs in ecosystems, and is not simply an apparent effect of increased microbial turnover (Fontaine et al. 2003, Fontaine et al. 2004a, Fontaine et al. 2004b, Carney et al. 2007). Notably, these studies do not address the source of the N used for the production of microbial biomass and exo-enzymes, although more recent literature directly supports the reviewer's statement that "low N availability favors microbial mining of N in recalcitrant SOM" (Fontaine et al. 2011). **We have extended a paragraph of the discussion,** as per the reviewer's suggestion, which includes the idea of a threshold of N availability, where N availability constrains the production of enzymes and biomass at very low N, while at moderate to high N availability the microbial community composition or function shifts toward the acquisition of C from more labile sources (i.e., not SOM priming). We modify this threshold idea slightly to link our manuscript and the literature; it is possible that priming of SOM at low soil N availability is made possible by root N inputs. That is, stoichiometric constraints at low N would preclude a microbial response to C exudates, but the exudation of small amounts of N allow for the production of exo-enzymes and thus enable the priming observed in the literature (Fontaine et al. 2003, Fontaine et al. 2004a, Fontaine et al. 2004b, Carney et al. 2007, Fontaine et al. 2011).

Concerning the writing, the discussion section of the manuscript is clearer than the abstract and introduction section. Try to re-inject some sentences from the discussion to the introduction and abstract to better present your ideas.

We have taken this suggestion, particularly at the end of the abstract and the latter two paragraphs of the introduction. We have incorporated the language from the discussion into the introduction regarding root N inputs as potential drivers of microbial function.

For example, line 5 in abstract what do you mean by "the causal role of exudation"? I guess what you mean but the redaction should be improved to be clear (i.e. disentangle rhizosphere processes, isolate exudation effect).

We have removed the phrase "the causal role of exudation" and replaced it with "the effects of exudate chemicals in isolation from other rhizosphere processes (e.g., hydraulic changes, exchange of signalling molecules, and interactions between roots and mycorrhizal associates)." **More generally, we have re-framed the manuscript to focus on the stoichiometric controls of microbial response to exudates, with less focus on the "causal role of exudation", as per the comments of both reviewers.**

Line 13-15 in abstract: this part is important and should be limpid since you are presenting the two main hypothesizes of your manuscript.

We agree, and we have modified the introduction to focus on how N availability may constrain microbial response to C exudates, incorporating some aspects of the discussion section as suggested earlier.

What do you mean by "exudation alone"? Here it seems that the objective of your manuscript is to isolate the effect of exudation from effects of other rhiposhere processes, but you do not even mention these other processes and you do not explain how do you isolate specifically the exudation effect.

We have added mention of other rhizosphere processes to the abstract, and explained these other processes in more detail in the introduction. We specify how we specifically isolated the exudation effect at the end of the introduction: "by adding chemicals often found in root exudates to intact soil in the field, we have isolate the effect of these chemicals from other rhizosphere processes (e.g., hydraulic changes, exchange of signalling molecules, and interactions between roots and mycorrhizal associates)."

Some sentences of the manuscript seem banal. For example at the end of abstract "This study supports a cause-and-effect relationship between root exudation and enhanced microbial activity: : :". Really I think that your manuscript contains more important messages than this *and that nobody doubts on the cause-and-effect relationship between root exudation and enhanced microbial activity in rhzosphere.*

We have modified the final sentence of the abstract to focus on potential N constraints on the microbial response to C exudates, rather than focusing on the direct cause-and-effect relationship between root exudates and microbial activity. Specifically, we now end the abstract with the idea that both reviewers found most interesting: “Together, the model and field experiment suggest that C-containing exudates can induce microbial N limitation and constrain enzyme synthesis; root N inputs can lift this constraint. Thus, this study suggests that exudate stoichiometry is an important and underappreciated driver of microbial activity in rhizosphere soils.”

Specific points
P6903 L1-7 and in other parts of the manuscript where you present your two hypothesizes. Presentation of first hypothesis is not clear: what do you mean by “exudation alone”. I am not sure that you isolated the effect of exudation from effect of other rhizopshere processes since exudate mimics were applied into plant rhizosphere where the other rhizopshere processes can proceed as well (which was a good idea from my opinion). Still in the presentation of first hypothesis: what do you mean exactly when you mention “exudate mimics”? Do these exudate mimics include C only or C and N ? It is not clear.

In response to these comments and the comments of the second reviewer, we have removed the original presentation of two hypotheses and condensed them into a single, general, and more biologically interesting hypothesis. **We now hypothesize that “The ability of soil microbes to utilize root exudates for the synthesis of additional biomass and exo-enzymes is constrained by N availability- roots may elicit a larger rhizosphere response by exuding N as well as C”.** This hypothesis more clearly addresses the central scientific concept of our manuscript and the issue both reviewers found compelling and novel.

P6905 Indication of soil pH is useful

We added soil pH to the site description. These soils were moderately acidic, with pH values (1:1 soil:water extractions) of ~4.0 (Bowden et al. 1998, Brzostek and Finzi 2011).

P6906 Could you indicate whether microlysimiters were inserted in a soil zone where tree roots were present (precise root biomass present in soil where lysimiters were inserted)? It is important to understand whether exudates mimics were incorporated nearby plant rhizosphere.

The microlysimiters were inserted into the soil in random locations within an intact closed-canopy forest, so there certainly were roots present. We measured the root biomass collected with the small soil cores and there was no difference across treatments (ANOVA, $p > 0.1$), but these very small cores do not give a precise quantitative measure of root biomass that we are comfortable reporting in the manuscript. Extensive measurements of fine root biomass have previously been reported for this specific site within Harvard Forest at ~300 g C m^{-2} (Gaudinski et al. 2000), which is relatively high root biomass. This information has been added to the methods section of the manuscript.

P6909 L8-14 The method for measuring soil proteolytic enzymes is not enough clear. I know that papers describing the method have been published elsewhere but it is important to understand principle of the method.

We have expanded the methodology related to the measurement of soil proteolytic enzyme activity, as requested. The principal of the method is that proteins are amino acid polymers, and the reaction governing the rate of protein breakdown into bioavailable forms is depolymerisation into amino acid monomers. This method measures the concentration of free amino acids before and after a 4-hour incubation; the change in free amino acids reflects the rate of protein depolymerisation. A small amount of toluene is included in the incubation to inhibit microbial uptake of amino acids, which otherwise would invalidate the use of this method.

P6910 1-10 The model do not consider a plant compartment. For the comparison of model predictions and experimental results, how did you take into account effects of living plants on soil functioning (i.e. plant exudation, plant N uptake etc). You should discuss these questions and present main assumptions of the model.

The reviewer is correct- the model does not explicitly include a plant compartment. We explicitly model exudation by live roots with the additions of dissolved organic matter (DOM) as explained in the methods. We included a first-order equation to remove dissolved inorganic N (DIN) from the available soil pool as explained in the methods and supplementary material; this removal of DIN reflects the aggregate activity of multiple processes, including root DIN uptake but also leaching, nitrification and denitrification. Thus we have modelled some plant processes, but certainly not all important plant processes.

It would be important to include a specific plant component if we were working at a higher level of biological organization (e.g., ecosystem scale biogeochemistry), as stand-level fine root crop and turnover would influence SOM substrate availability and the proportion of rhizosphere soil to bulk soil. However, our modeling exercise is focused on microbial responses to exudates, which is a mechanism that operates on a smaller scale of inference (e.g., a cm^3 of soil). It would be interesting to incorporate this model of microbial physiology and decomposition (Schimel and Weintraub 2003) into an ecosystem-scale biogeochemical model (e.g., CENTURY, PnET) to examine the broader consequences of exudation and priming on ecosystem functioning, but that is outside the scope of this current project. **We have expanded our model description in the methods to explicitly address the issues of spatial scale and the assumptions that we have made regarding plant activities.**

P6912 When possible, it would be nice to interpret predictions of the model in light of your knowledge of model structure and assumptions. Why does this model yield such results?

We agree, and we have addressed some aspects of the model structure and assumptions in the discussion section (P15, L30). Within the model results section (near to where the reviewer's comment was pointed) we have explicitly explained the model behaviour that is most applicable to this manuscript: "The addition of C exudates frequently pushed microbes into N limitation; thus the simultaneous exudation of N allowed for the synthesis of additional microbial biomass and exo-enzymes, which have low C:N ratios and thus high N requirements."

P6915 L5-18 All these citations confirm that the cause-and-effect relationship between root exudation and enhanced microbial activity in rhizosphere is not new. My objective is not to diminish the importance of your study but to help you to insist on what is more original : the stoichiometric contrains of rhizosphere microorganisms and the role of plant N exudation on microbial activities.

We agree, and we have focused the revised manuscript on the stoichiometric control of microbial response to exudation (see above), rather than the more general “direct response” of microbes to exudation chemicals, which both reviewers viewed as an underwhelming framework for the manuscript.

P6916 L9-13 This sentence is too general since the delivery of exudate mimics increased activity of enzymes that decompose labile substrate (generally N poor substrates) and decreased activity of enzymes that decompose recalcitrant SOM. What is the model prediction about the type of enzymes (enzymes degrading recalcitrant substrates versus enzymes degrading labile substrates) stimulated by the delivery of exudate mimics?

The model only simulates a single enzyme and a single substrate pool and is thus unable to address issues related to recalcitrant vs. labile substrates. Modifying the model to incorporate recalcitrant vs. labile substrates would require substantial time for model development, and is unfortunately outside the scope of this manuscript. Such an effort is underway in our subsequent research. We also note that protein and amino sugars are abundant in soil and relatively N-rich but labile. Thus it is not always the case labile SOM contains little to no N.

P6917 L15-18 The reduction of activity of enzymes decomposing recalcitrant substrates after delivery of exudate mimics (C and N) could be due to an effect of N alone (see references in Generral comment). This is the reason why I think that the experimental plan lacks of a +N control to be able to clearly understand the role of plant exudates.

We have addressed this issue (as explained above) by adding a discussion paragraph related to the extensive literature on atmospheric N deposition. Briefly, the reduction in lignolytic enzyme activities is the only aspect of our study that is consistent with the literature that N additions can reduce SOM decomposition (reviewed by Janssens et al. 2010). The bulk of the measured responses are consistent with a larger microbial response to C and N exudates relative to C exudates alone, including higher microbial biomass, rates of microbial respiration, and labile exo-enzyme activity, all of which are opposite of the general response to N deposition (Wallenstein et al. 2006b, Treseder 2008, Janssens et al. 2010). Thus we have maintained our interpretation of the data. However, we believe that the new discussion paragraph detailing these issues has improved and clarified the manuscript.

Good luck

We thank the reviewer for his or her insightful comments and criticisms.

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