Author Response to Interactive comment on "Interrelationships among soil nitrogen transformation rates, functional gene abundance and soil properties in a tropical forest with exogenous N inputs" by *Yanxia Nie et al.*

Yanxia Nie and co-authors
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Response to Anonymous Referee #2

Reviewer comment: Due to the complexity of nitrogen cycling in terrestrial ecosystems, it deserves to explore how elevated nitrogen deposition affects soil N transformations in the N-rich soil of tropical forests. Overall, this manuscript was well written and easy to read, but the current version is suffering from some critical defects.

Response: Thanks for the positive evaluation to our work! We carefully revised the manuscript based on your suggestions. Our point-by-point responses to your comments are listed below. Hope you would find these revisions satisfactory.

Reviewer comment: First, this study measured the net mineralization and nitrification, completely different from gross mineralization and nitrification. To this point, the title of this study is not appropriate, because net mineralization and nitrification actually include the balance of various transformation processes such as ammoniation and immobilization, which conceals real nitrogen transformation processes.

Response: Agreed. Net mineralization and nitrification rates essentially measures the net temporal changes in the pool size of inorganic N (NO_3^- and NH_4^+) contents within the incubation period (in our case, 30 days). The limitation of field-assessed net rates can not disentangle the detailed gross transformation rates actually happening simultaneously. We therefore specified the N transformation rates as 'net N transformation rates' in the title and throughout the manuscript during this revision. In another study from our lab, Han et al. (2018) reported the responses of gross rates to N additions (Science of the Total Environment, 626: 1175-1187). We mentioned some of their results in our discussions.

Reviewer comment: Second, the descriptions in Methods are not detailed and thus affect understanding of the results. For example, the descriptions about the specific time for nitrogen addition and sampling soil cores for net mineralization were unclear. Considering net mineralization is the difference of ammonium concentrations between 30 days, the time for nitrogen addition and the sampling of two soil cores is very important. If the sampling of second soil cores was just after nitrogen addition, mineralization could be overestimated because added N contributed to increase in soil ammonium concentrations.

Response: N additions were applied on the 24th of each month from September 2014 through October 2016. The incubations were carried out 9 times in September 2014, December 2014, March 2015, June 2015, September 2015, December 2015, March 2016, June 2016, and September 2016. Each incubation was started a couple of days before the N addition date and lasted for 30 days. We have provided these methodological details in the revised manuscript as the reviewer suggested.

Reviewer comment: Third, it is well known that nitrogen addition will lead to soil acidification. However, this study did not separate from inorganic nitrogen input from its acidification (also see Fig. 6). This strongly reduces the importance of this study, e.g. both low pH and higher inorganic nitrogen concentrations can show negative effects on nitrogen transformations.

Response: In the 2-year study period, we monitored the changes in both soil pH and inorganic N (NH_4^+-N and NO_3^--N) contents after N additions and analyzed the relationship between these important factors and net N transformation rates. No significant relationship was found between them in the dry season (Table 1a). However, in the wet season, the net N transformation rates (R_m and R_n) had significantly positive correlations with NO_3^--N content, but had significantly negative relationships with soil pH and NH_4^+-N contents (Table 1b and Fig. 5). Since changes in pH actually was induced by the nitrogen additions, we were therefore not able to separate the N addition effects from the acidification effects with our experimental design (only N input was manipulated). Further studies manipulating both soil acidification and N addition at the same time might be helpful in teasing out the two kinds of effects.

Reviewer comment: Fourth, it is very good to include the measurements of N-related functional gene abundance, but it is a pity that N-related functional gene abundance was not related with the specific nitrogen transformation processes. As a result, it is difficult to make a microbial mechanism explanation for net mineralization and nitrification. Before the manuscript is accepted to publish, the above issues should be well clarified.

Response: Yes, we only found N_2O emission exhibited a significantly relationship with *nosZ* gene abundance in this study. We did not find a significant relationship between the AOA abundance and net N mineralization rates. The main reason may be that net N mineralization rate actually measures the net temporal changes in inorganic N pool sizes, which are governed by several specific gross input and output rates such as gross mineralization and immobilization. It is possible that the functional genes abundance may have closer relationships with the gross N transformation rates. Some of such relationships have been reported in a recent study by Han et al. (2018) using soil samples taken from the same experimental plots as in this study. We have added these descriptions in the revised discussion to further explore the relationships between functional gene abundance and N transformation rates.