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Interactive comment on "Distribution of ammonia oxidizers in relation to vegetation characteristics in the Qilian Mountains, northwestern China" *by* X. S. Tai et al.

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Dear Dr. Jia,

Thank you very much for handling of our manuscript and thanks a lot to the anonymous referee #1 for his/her wonderful comment. Our replies summarized from the discussions of all the authors are shown as follows.

1. In page 5125, alpine meadow covered, the "covered" should be as covering. In page 5129, the sentence "Nutrient poor soils are usually indicated by . . .dissolved N poor." need to be polished.

Thank you very much for your good suggestions. In p. 5125, l. 28, the word "covered" has been changed to "covering" in the revised MS. In p. 5129, l. 10-12, the sentence "Nutrient poor soils are usually indicated by . . . dissolved N poor." has been replaced by "It has been found that DON (dissolved organic nitrogen) comprises the majority of TDN (total dissolved nitrogen) pool which is indicative of alpine ecosystems (Hill et al., 2011, Nature Climate Change)".

2. Materials and methods; from Fig.1 we known the sampling sites cover a large area, but why the size of sampling site was not described in this MS? Moreover, how the author made sure three quadrats could represent the different types of alpine meadow? As we all known, the spatial variability is huge for plant or soil in a mountain area. And which level of soil was taken for this study?

Designing of sampling referred to the previous similar studies (Yuan et al., 2013, FEMS Microbiol. Ecol.; Xiong et al., 2014, FEMS Microbiol. Ecol.). The sampling sites located in a typical valley along an altitude gradients in the upper reaches of the Heihe River (Fig. 1). Each site covered an altitudinal span of 200 m while it is 10 " to 30 " in longitudinal extent (Table 1). The size of each quadrat was 1 $\times$ 1 m2 (Wang et al., 2003, Plant Ecol.; Wu et al., 2011, Acta Ecologica Sinica). The chosen quadrats typically represented each sampling site, as well as the different types of alpine meadow. Soil samples were taken from the depth of 20 cm of each profile. These information have been added in section 2.1 of the revised MS.

3. Materials and methods; only fifty clones of each library were picked up for sequencing. Is it enough to represent the diversity? Had the author done the rarefaction curves analysis? Moreover, they might have sequenced 1500 clones for AOA and AOB according to the author's description. However, only 175 sequences of amoA genes were deposited in genbank. In my opinion, all sequenced clones should be submitted to genbank.

The rarefaction curves and coverage of the clone libraries were applied to prove that

fifty clones of each library was enough to represent the amoA diversity and have been added in the supplemental materials. The sequences of the amoA OTUs were deposited into GenBank instead of all of the sequence data. If needed, we can deposit all of the data into the bank.

4. Materials and methods; nitrification rate is an important indicator for ammonia oxidizing enzyme activity. However, the data was not present in this MS.

Thank you very much for the good suggestion. We believe that detection of nitrification rate will be plus to prove the conclusions drawn from this research. In this study, we have focused on diversity, abundance and community composition of ammonia oxidizers. Otherwise, we plan to proceed field manipulation experiments including enzyme activity measurement in the future works.

5. Results; in page 5130, line 4-10, "Group I.1a. . .. . .", the long sentence was not suitable in here or to be concised. Fig. 4 and Fig. 5 were bad at presentation. The phylogenetic trees of amoA in supplemental materials were better. But both of them should be added the access number information in the phylogenetic trees.

Thanks a lot for the helpful suggestion. In p. 5130, l. 4-10, the long sentence "Group I.1a . . .. . ." has been simplified as "Group I.1a, Group I.1a-associated, Group I.1b and ThAOA are typical thaumarchaeotic groups associated with ammonia oxidation.". In the revised MS, Fig. 4 and Fig. 5 have been replaced by the phylogenetic trees in the supplemental materials. Besides, all of the amoA sequences were added with the access numbers.

6. For Q-PCR analysis of amoA genes, the author had listed the standard curve of amoA gene Q-PCR and their R2, efficiency. However, from the supplemental materials and Fig. 2, we known the copy numbers of bacterial and archaeal were very low. So I am not sure these data really represent the AOA and AOB populations. Sometimes even the negative control could be detected a signal as high as the data present in this study. Please attach the Q-PCR and their melting curve graphs in the supplemental

materials.

Q-PCR analyses of amoA have been conducted in duplicate to make sure that the results were reliable. Results of Q-PCR including the melting curves have been attached in the supplemental materials.

7. Discussions; in this part, the author always stayed in a descriptive model, and didn't convincingly explain the distribution pattern of AOB and AOA in five different alpine meadow.

Thank you very much for your excellent suggestion. The discussion part has been rewritten according to the suggestion in the revised MS.

Thanks again for the excellent suggestions provided by the anonymous referee.

Yours sincerely, Guangxiu Liu

Interactive comment on Biogeosciences Discuss., 11, 5123, 2014.