

Interactive comment on “Abiotic versus biotic controls on soil nitrogen cycling in drylands along a 3200 km transect” by Dongwei Liu et al.

Dongwei Liu et al.

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Dear editor,

We would like to extend our grateful thanks to reviewer1 for his/her constructive comments and suggestions to our manuscript. In the revised version of this manuscript, we will rethink our data carefully and fix many confusing sentences that the reviewer had pointed out. In addition, we will also make further efforts to improve the English writing (including taking specific comments from another reviewer) by asking a native speaker to correct our revised manuscript before resubmission. Please find our line-by-line response to the review comments below.

General comments

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Overall, this manuscript provides an insightful data set that I believe will be of interest to anyone interested in aridland biogeochemistry. The large geographic scale and compound-specific isotopic analysis are especially valuable and the conclusions reached seem valid. Generally, the discussion of the mechanisms driving the observed patterns is thorough. There are some issues that need addressing, primarily in the discussion where several important processes have not been raised (mainly NO production), and there are several mechanisms that do not make sense (perhaps partly as a result of unclear English). This section would benefit from revision. Generally, the figures are clear and informative. Editing for English language is necessary prior to publication.

Reply: Thank you very much for your appreciation of our study. The comments from the reviewer are really helpful. In the revised version of the manuscript, we will rewrite part of discussion section as suggested by the reviewer. We will improve general writing by asking a native speaker to correct our revised manuscript before submission.

Specific comments:

1) Methods: Is it just a coincidence that there is a gap in sampling sites around 100 mm MAP?

Reply: You are correct that it is just a coincidence that a gap of MAP seems appear around 100 mm. Our sampling sites were well-distributed at the distance of about 100 km between two adjacent sites. The threshold happened between site 15 (MAP = 102 mm) and site 16 (MAP = 142 mm). Please see Figure 1 in the main text. The "unintended gap" serves nicely for us to break the entire gradient into 1) arid zone and 2) semiarid zone for data synthesis and discussion.

2) Discussion: What about loss of NO during nitrification? NO can be the dominant trace gas emitted from arid soils, and would explain loss of ammonium without subsequent appearance of nitrate. For process see Firestone Davidson (1989) For arid land NO production see Homyak et al (2016, PNAS) and Soper et al (2016, Global Bio-

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geochemical Cycles). This process belongs on Figure 8! A discussion of the isotopic consequences of this process should also be included.

Reply: Thank you for your suggestion. We agree with you that NO emission during N transformation is really important in arid soils. In the revised manuscript, we will add details about this NO losses in the section of 'The losses of nitrate and ammonium'. For example, 'The concurrence of abiotic and biotic processes governed gaseous N efflux, contributing to the remove and 15N enrichment of soil inorganic N; for example, studies had reported that nitric oxide (NO) was the dominate emissions in some drylands (Homyak et al., 2016; Soper et al., 2016). Chemodenitrification is an abiotic process, in which the reduction of NO_3^- or NO_2^- to NO and N_2O is coupled to the oxidation of reduced metals (e.g. Fe (II)) and humic substances (Medinets et al., 2015; Zhu-Barker et al., 2015). Ample soil NO_3^- was present in some arid zone soils in this study (Fig. 2c), meanwhile our companion work also observed higher available Fe in arid zone soils (Luo et al., 2016). Chemodenitrification therefore can occur when soil NO_3^- contacts with metal (e.g. Fe (II)) minerals (Zhu-Barker et al., 2015). In addition, the most important reaction of chemodenitrification is the formation of NO via nitrous acid (HNO_2 (aqueous phase), HONO (gas phase)) decomposition (Medinets et al., 2015). In the dry soils, nitrifiers can remain active in thin water films and results in higher potential nitrification rates when conditions are right, e.g., after pulse of rain (Sullivan et al., 2012). Nitrite originated from soil nitrification may be more likely to be decomposed to NO via chemodenitrification than oxidized to nitrate (Homyak et al., 2016), explaining the loss and 15N enrichment in soil ammonium. Alternatively, nitrifier denitrification can also serve as an important mechanism for NO emission by the reduction of soil NO_2^- (Homyak et al., 2016).'

In Figure 8, we will also try to compare and contrast the major soil N pools and N transformations (ammonification, nitrification, denitrification, among others) between mainly abiotic driven arid zone soils and biotic dominated semi-arid zone soils. Specific losses of N, including NO, N_2O , and N_2 , are not shown; instead, discussed in the text.

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3) The foliar ^{15}N reflecting ^{15}N of NH_4^+ in the soil- this could also reflect shifting plant physiology across the significant precipitation gradient, rather than just plant source preference for ammonium. Many aspects of plant internal N cycling likely shift as a function of water availability and would influence foliar ^{15}N . This should at least be acknowledged as an alternative explanation. Also, ammonium shows a larger range of isotopic values along the transect than nitrate, making it less likely that plant ^{15}N would correlate with nitrate ^{15}N anyway.

Reply: Thank you for your suggestion. In the revised manuscript, we will add plant physiology as an alternative reason for the changing pattern of foliar $\delta^{15}\text{N}$. Seen from the data of foliar ^{15}N and inorganic ^{15}N , foliar ^{15}N range (-2.1 8.7‰) was basically smaller than that of ammonium (-9 16‰) and nitrate (1.6 13.3‰). Significant ^{15}N range for soil NO_3^- did exist but did not correlate to the plant ^{15}N .

4) Deposition- I think you need to be clear about the difference between wet versus dry deposition (with different isotopic signatures) and how you might expect them to change along the transect. Are there any measures of deposition anywhere on the transect you could mention?

Reply: Good point. In our companion work along this transect (Wang et al. 2014), we reported that rates of bulk N deposition (wet + dry) were increasing from west to east along this transect; data were estimated from a published paper (Lelieveld and Dentener 2000). We expect that dry deposition could be higher than wet deposition in the arid zone soils, and that in the semiarid zone soils the contribution of wet deposition will increase significantly. In previous studies, higher $\delta^{15}\text{N}$ values in dry deposition than in wet deposition had been reported for nitrate (e.g., by 1 to 3 permil in the northeastern US) (Garten 1996, Elliott et al. 2009) and ammonium (up to 33 permil) (Heaton et al. 1997).

Line 225- nitrate could also be removed from the soil by biological uptake. This also has potential to be a fractionating process (although evidence for fractionation by directly

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by plants under field conditions is limited, mycorrhizal fractionation is likely). You posit later that plant uptake of nitrate is low, but this may not necessarily be the case.

Reply: Thank you. We will add plant and microbial uptake of NO_3^- in the revision. For example, 'Nitrate can be removed from the ecosystem via denitrification, leaching, and also plant and microbial uptake.' This study area is highly N-limited according to previous N manipulation experiment. Plant would take in both 15N and 14N in N limited areas (Craine et al. 2015). So, the fractionation effect during the plant N uptake could be low.

Line 235- increasing compared to what? This is important.

Reply: The sentence will be modified as 'In addition, a preliminary study using a 15N -labelled incubation experiment also showed that the potential N_2 loss rates via denitrification were increasing with precipitation increasing in the semiarid soils'.

Line 244- There are several potential mechanisms for chemodenitrification (see again Homyak 2016)

Reply: Thank you. We will add more discussion on chemodenitrification based on the references provided. Please see our above response to the 'NO losses' (Specific comments 2) discussion).

Line 255- Soper et al (2016, Global Biogeochemical Cycles) did find increased NH_3 flux with wetting in an arid system.

Reply: Soper et al. (2016) reported increased NH_3 flux 24 hours after a 15 mm artificial rain in soils with pH 7.1, likely due to the stimulation of NH_3 production (ammonification) followed by NH_3 volatilization. However, the dominant post-rain N loss was still NO loss in that study, likely due to the enhanced nitrification, as this reviewer had emphasized. In our transect within the semi-arid zone spanning a precipitation gradient from 140 mm to 436 mm, and pH decrease from 8.6 to 6.7, plus associated vegetation change and plant $\text{NH}_4^+/\text{NO}_3^-$ uptake, we believe the dominant drivers on soil NH_4^+

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consumption are 1) plant uptake, and 2) nitrification. Nevertheless, the study from Soper et al. (2016) is interesting for our understanding of dryland N loss and will be cited in our revision. Thank you!

Line 256- “First, plant uptake will be enhanced when it is coupled with the microbe-regulating N cycling”- I’m not sure what this means

Reply: Sorry for the confusing. The stimulation of pulse rainfall events to microbes and plant N uptake is different, with a lower stimulation threshold for microbes in extremely dry areas (Dijkstra et al. 2012). Below the MAP threshold, soil microbes may be activated by small rainfall events compared with plants, producing a pulse of high N availability to plants. But if there is an asynchrony in N-cycling via water limitation on plant N uptake, the mineralized N is subject to nitrification and denitrification losses. Above the MAP threshold, these two processes are probably coupled (i.e., microbial mineralized N immediately used by plant), resulting in higher N retention efficiency (Wang et al., 2014). We will rewrite this sentence as well as the entire paragraph to discuss both plant N uptake and nitrification on the consumption of soil NH_4^+ and its isotopic signal.

Line 268- This doesn’t make sense. I don’t know of any evidence showing preference for enriched substrates- I would expect it to be exactly the reverse in fact.

Reply: Thank you. To avoid misleading, we will delete this sentence.

Line 324- This paragraph should be rewritten for clarity- right now it’s just listing off a bunch of processes and it’s confusing. What are the processes that would explain more NH_4^+ , with higher enrichment, at low precipitation? Increasing volatilization with precip explains the concentration gradient, but would induce the opposite isotopic pattern (though it depends really on how much volatilization occurs as a fraction of the standing pool). A greater proportion of atmospheric deposition versus mineralization at low precip might explain the higher ^{15}N . If you invoke fixation by BSCs at low precip, this would also tend to decrease, rather than increase, the ^{15}N at those sites. This also

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needs to be clarified- are there BSCs on the transect? However it looks from the gene abundance data like N fix genes increase along the transect. Rethink this paragraph.

Reply: We have rethought the whole paragraph and will rewrite it to address the respective contribution of aerosol deposition and BSC to NH_4^+ accumulation in the arid zone soils. For example, this paragraph will be modified as, 'There was also a slight NH_4^+ accumulation in the arid zone soils, with higher $\delta^{15}\text{N}$ enrichment in some sites (Fig. 2b, e). This results might be driven by the mixing of many input and output processes. Input processes mainly include NH_4^+ deposition (accompanying with nitrate deposition) and ammonification, and output processes include NH_3 volatilization and nitrification mentioned above. Ammonium has been shown to be the dominant species in bulk N deposition in China (Liu et al., 2013), thus it could be one of those processes contributing to NH_4^+ accumulation in the arid zone soils. Dry deposition is the dominant form of deposition in arid climate. Furthermore, higher $\delta^{15}\text{N}$ values in dry deposition than wet deposition have been reported (Elliott et al., 2009; Garten, 1996; Heaton et al., 1997). In the drylands, biological N fixation is considered to be an important source of N input (Evans and Ehleringer, 1993). With the exception of the biological N fixation by legume plant (*Caragana* spp.) showed in the same transect (Wang et al., 2014), in this study, we speculated that biological N fixation by BSCs (Wu et al., 2009; Zhuang et al., 2015) also contributed to soil NH_4^+ pool in the arid zone. We did observe BSCs during sampling in the arid zone soils (personal observation). Besides, the previous research has reported the potential N-fixing activity and potential N input of BSCs in the grasslands of Inner Mongolia (Liu et al., 2009). Biological N fixation by BSCs provided NH_4^+ with $\delta^{15}\text{N}$ value around zero, contributing to the pool size of soil NH_4^+ but not its ^{15}N enrichment.'

Line 336- Does fractionation during mineralization actually increase with mineralization rate though? I don't recall any evidence for this. Also I think invoking heterotrophic nitrification, when as far as I know there isn't a lot of evidence this is an important process in the field, is a stretch. Maybe remove.

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Reply: Though the isotope effect of N mineralization is most often said to be low or negligible, it might be higher than we expected. Our lab recently reported that $\delta^{15}\text{N}$ values of soil NH_4^+ were lower than that of bulk soil N by 6-8 permil in two forest soils collected in northern China (Zhang et al. 2015). As had suggested by review 2, they can be as high as 20 permil if one looks at enzymes and their isotope effects that are most likely involved in deamination of organic N forms in cells (Werner and Schmidt 2002). We are not sure about the changes of fractionation effect during mineralization along the precipitation gradient, but fractionation effect could exist. We are not stating that fractionation itself increased with increasing mineralization rate. Clearly nitrification occurs in our study area. Ammonium was ^{15}N depleted relative to bulk soil, indicating that there might be only a small fraction of soil ammonium was oxidized by autotrophic nitrifiers. Heterotrophic nitrification therefore could be one of the reasons for the source of soil nitrate, and contributes to the size increase of soil nitrate pool, but not to the ^{15}N signal of soil ammonium. Both the fractionation effect over mineralization and the occurrence of heterotrophic nitrification were the alternative reason for ^{15}N depleted soil ammonium in the semiarid zone soils. Based on those reason, we would like to keep this assumption although we did not test it in this paper, and there were few reports on the fractionation effect of heterotrophic nitrification till now.

Line 359- "Increasing ammonification with increasing MAP both reduced NH_3 volatilization" Why would more mineralization reduce volatilization? Unless you mean that volatilization decreases with precip? Again, I'm not sure that this is necessarily true. pH probably the main driver.

Reply: Sorry for the confusing. This sentence will be modified as 'With the increasing of precipitation, both the stimulated ammonification and reduced NH_3 volatilization (with low pH) may contribute to ^{15}N depleted soil ammonium pool'.

Line 360- why does more mineralization mean more plant uptake? Plant uptake is likely more a function of water availability. These things likely co-occur, but it's not causal. Perhaps misinterpretation of wording- re-write.

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Reply: Sorry for the confusing. In the semiarid zone, with increasing precipitation, plant N uptake was going to couple with N mineralization (Please see response to line 256 above). So N mineralization, nitrification, and plant N uptake all increase with increasing precipitation. We will rewrite this sentence as 'Higher ammonification (N mineralization) would couple with plant N uptake and also favor soil nitrification'.

Figure 8- I think it needs to be clear that the size of arrows indicates qualitative interpretation of these fluxes rather than actual measurement- the presence of pool sizes on the boxes makes this especially necessary. And again, NO loss is likely much more important than anammox and should appear here.

Reply: Thank you for your suggestion. We will add 'qualitative interpretation' in the figure legend. This figure was going to illustrate relative importance of various N processes. The specific N loss was not shown in the figure. We recognized that NO loss was very important in the N losses in these drylands, however, it is part of the nitrification process (as well as part of the denitrification process according to the 'Leaking Pipe Hypothesis'). Similarly, we did not show N₂O/N₂ loss separately from denitrification. Rather, NO loss would be discussed in specific section (e.g. soil nitrate losses) in the discussion.

Technical corrections: The manuscript contains many examples of awkward or technically incorrect English that can obscure meaning and requires editing by a native English speaker before publication. E.g. in the abstract –'our understanding of' might replace 'understanding about' and 'nitrogen cycling in drylands' rather than 'nitrogen cycling of drylands'. Also 'the patterns and mechanisms of water availability on soil N cycling' doesn't make technical sense. 'Driving' rather than 'driven'. 'Above and below' rather than 'on the two sides of'. 'Preference for' rather than 'preference of', etc. Reply: Thank you very much for your comments and correction. In the revised version of the manuscript, we will make further efforts to improve writing. The reviewer 2 had also give a lot of suggestions on how to improve the writing.

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Respectively,

Liu D, Zhu W, and Fang Y, on behalf of all co-authors.

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