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Dr. Frank Hagedorn
Editor of Biogeosciences

Title: Errors associated with calculating the gross nitrification rates in forested catchments using the triple oxygen isotopic composition ($\Delta^{17}\text{O}$) of stream nitrate

Authors: Weitian Ding et al.

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Dear Dr. Frank Hagedorn:

Thank you very much for handling our manuscript. We would like to thank the referees as well for the constructive comments on our manuscript. We have carefully studied the comments and revised the manuscript accordingly. We include below point-by-point responses to the comments, and detailed descriptions of the modifications we made to the manuscript. Besides, we also uploaded the revised manuscript in MS Word, in which all the revisions from BGD version were recorded. We hope that with these changes you will find our revised manuscript appropriate for publication in your journal.

Sincerely yours,

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Cc: Drs. Urumu Tsunogai and Fumiko Nakagawa

Response to the handing associate editor:

Here, I ask the authors to add 1-2 sentences to the discussion that in the case of the water environments, the $\Delta^{17}\text{O}$ values and NO_3^- are mostly homogeneous in the water column due to the active vertical mixing, which is not the case in soils.

Thank you for the advising. We added the following sentences in the revised manuscript (P4/L61-62; P7/L114-116).

Contrary to water environments, where the $\Delta^{17}\text{O}$ values of NO_3^- in the water layers are homogeneous in the water column due to the active vertical mixing of water and can be measured easily, it is often difficult to determine the $\Delta^{17}\text{O}$ values of NO_3^- consumed in soil layers.

Differ from water environments, vertical mixing of water/soil is difficult in forested soil, so the $\Delta^{17}\text{O}$ values of soil NO_3^- are often heterogeneous.

The manuscript requires an improved wording

Thank you for the advising. We improved wording in the revised manuscript. Besides, the revised manuscript reviewed by an experienced editor whose first language is English and who specializes in editing papers written by scientists whose native language is not English.

Response to the referee #1:

Their simulation is based on a case where nitrate fluxes decrease with soil depth, which is the case if net nitrification is negative (nitrate consumption larger than gross nitrification). It would be very interesting to see what happens in the case of a positive net nitrification. This could be done with just one more simulation.

Thank you for your comment. Our simulation was done for the forested catchment reported by Hattori et al. (2019). While the deposition flux of NO_3^- was $7.0 \text{ kg of N ha}^{-1} \text{ y}^{-1}$, the leaching flux of NO_3^- was $2.6 \text{ kg of N ha}^{-1} \text{ y}^{-1}$ in the forested catchment, so that the influx of NO_3^- was higher than that of outflux. Thus, the NO_3^- fluxes always decreased with soil depth in our original simulations, shown by Figures 1c and 2c in the manuscript.

In response to your request, we made a new simulated calculation, in which NO_3^- fluxes increased with soil depth in the soil layers from 1 to 5 with an increasing rate of $0.44 \text{ kg of N ha}^{-1} \text{ y}^{-1}$ for each layer, while NO_3^- fluxes decreased with soil depth in the soil layers from 6 to 10 with a decreasing rate of $1.32 \text{ kg of N ha}^{-1} \text{ y}^{-1}$ for each layer (Table R1). While the newly estimated GNR ($19.1 \text{ kg of N ha}^{-1} \text{ y}^{-1}$) was comparable with that estimated for the forested catchment with the profile shown by Figure 1 ($13.0 \text{ kg of N ha}^{-1} \text{ y}^{-1}$), it was still significantly smaller than the GNR calculated by using Eq.6 ($83.6 \text{ kg of N ha}^{-1} \text{ y}^{-1}$). Such additional simulated calculation by changing the nitrate fluxes with soil depths further supports our conclusion that the GNR estimated from the $\Delta^{17}\text{O}$ value of stream nitrate in forested catchments was, to an extent, an overestimate of the actual GNR.

In addition, the present results imply that the most important parameter to determine total GNR (and thus total GDR + uptake) is the $\Delta^{17}\text{O}$ value of NO_3^- consumed in soil layers. That is, the depth profile of NO_3^- fluxes has little impact on GNR.

Table R1. $\Delta^{17}\text{O}$ values of NO_3^- , leaching flux of NO_3^- , total consumption rate of NO_3^- (GDR + uptake), and GNR in the simulated forested soil where the distribution of $\Delta^{17}\text{O}$ values of NO_3^- is heterogeneous. While the net nitrification from soil layer 1 to 5 showed positive values, the soil layer 6 to 10 showed negative values.

Depth layer	$\Delta^{17}\text{O}$ ‰	NO_3^- flux kg of N ha ⁻¹ y ⁻¹	GDR +uptake	GNR
0	28.0	7.0	0.0	0.0
1	25.4	7.4	0.3	0.7
2	22.8	7.9	0.4	0.8
3	20.2	8.3	0.6	1.0
4	17.7	8.8	0.8	1.2
5	15.1	9.2	1.1	1.5
6	12.5	7.9	3.2	1.9
7	9.9	6.6	3.4	2.1
8	7.3	5.2	3.6	2.3
9	4.7	3.9	4.2	2.9
10	2.2	2.6	6.0	4.7
11	2.2	2.6	0.0	0.0
Total			23.5	19.1

In many soils, preferential water flow can be observed. In such cases, there is not a single, homogeneous nitrate pool per soil layer but nitrate that is more or less mobile along the flow paths and nitrate that is more bound within the soil matrix. The first is more prone to leaching and perhaps uptake, the second to denitrification. Simulating this would be a difficult task, probably out of the scope of the present article. Nevertheless, it would be useful if the authors would discuss this point

Thank you for your comment. As you point out, the leaching flux of soil NO_3^- in each layer is complex. Thus, we have added the following sentences in the manuscript (P11, L191-198 in revised manuscript).

The linear variation in the leaching flux and $\Delta^{17}\text{O}$ values of soil NO_3^- used in the simulated calculations (Fig. 1) is just one of many possible variations in forested catchments. It is impossible to determine whether the linear variation was realistic or not until the downward water flux, along with the concentration and $\Delta^{17}\text{O}$ value of NO_3^- , was determined for each soil layer. However, the simultaneous observations of the oxygen isotopes of soil NO_3^- and stream NO_3^- (Hattori et al., 2019; Osaka et al., 2010; Nakagawa et al., 2018; Rose, 2014) implied that the approximation of the $\Delta^{17}\text{O}$ values of soil NO_3^- to that of the stream NO_3^- (Fig. 2b) was unrealistic.

and especially if they could make recommendations on how to sample nitrate from the soil for $\Delta^{17}\text{O}$ determination: zero-tension lysimetry, tension lysimetry, centrifugation, extraction? I'm not sure if clear answers can be given with the present knowledge of soil nitrate transformations, but at least the question would deserve to be raised.

Thank you for your comment. We have recommended the sampling method of soil nitrate in the manuscript (P11, L199-202 in revised manuscript).

If we estimate the downward water flux at each soil layer, with the NO_3^- concentration and $\Delta^{17}\text{O}$ value of NO_3^- in each soil layer using, e.g., a tension-free lysimeter (Inoue et al., 2021), we could estimate the vertical change in the leaching flux of NO_3^- for each soil layer along with the $\Delta^{17}\text{O}$ of soil NO_3^- .

L. 3: the word "eluted" is rather used for the what is done on purpose in the lab. In this case, for the process observed in the nature, a better choice would probably be "leached".

Thank you for your suggestion. We changed the "eluted" to "leached" in the revised manuscript (P2/L4).

Line 6: instead "nitrate metabolized", it would be better to write "nitrate that is consumed", first because as soon as it is consumed, it is no longer nitrate, and second because "metabolized" is rather used to indicate that it is incorporated into organic matter, which is not the case for the denitrification.

Thank you for your suggestion. We changed the “nitrate metabolized” to “nitrate consumed” in the revised manuscript (P2/L6).

L. 24: on the same idea: "consumption" instead of "metabolic".

Thank you for your suggestion. We changed the “metabolic” to “consumption” in the revised manuscript (P3/L23).

L. 27: "is negligible" is too general, better add "often".

Thank you for your suggestion. We revised this in the revised manuscript (P3/L26).

L. 28: "by order of magnitude": do you mean "one" order?

We revised the sentence to “the GNR often exceeds the net nitrification rate by several orders of magnitude.” (P3/L28-29)

L. 21-29: very long sentence.

We revised the sentence in the revised manuscript (P2-3/L21-29).

The net nitrification rate can be estimated from an increase in NO_3^- concentration during a certain period. However, the gross nitrification rate (GNR) (net nitrification rate + consumption rate of NO_3^- (e.g., that assimilated by plants or decomposed through denitrification)), reflects the internal N cycling better than the net nitrification rate (Bengtsson et al., 2003), especially in forested ecosystems. Although the net nitrification rate is often negligible (Stark and Hart, 1997), the consumption rate is significant in forested ecosystems, such that the GNR often exceeds the net nitrification rate by several orders of magnitude (Verchot et al., 2001).

L. 31: it would be useful to explain shortly that the Δ anomaly is based on the δ of both ^{17}O and ^{18}O and that it is purposely defined so as to make it independent of mass-dependent fractionation.

We added the information in the revised manuscript (P3-4/L41-44).

This is because possible variations in the $\delta^{17}\text{O}$ and $\delta^{18}\text{O}$ values during the processes of

biogeochemical isotope fractionation follow the relation of $\delta^{17}\text{O} \approx 0.5 \delta^{18}\text{O}$, which cancels out the variations in the $\Delta^{17}\text{O}$ value.

L. 31: in my opinion, "conservative" would be better than "conserved" (because it tends to be conserved but it is not always perfectly conserved).

Thank you for your suggestion. We changed the “conserved” to “conservative” in the revised manuscript (P3/L31).

L. 33: it seems strange to write "REmineralized" when it may be mineralized for the first time after centuries of N staying in the soil in the organic matter.

We revised “remineralized nitrate ($\text{NO}_3^-_{\text{re}}$)” to “biologically produced nitrate ($\text{NO}_3^-_{\text{bio}}$)” in the revised manuscript (P3/L32-33).

L. 34-38, 53-57: long sentences.

We revised the sentences in the revised manuscript (P3/L34-39; P4/L57-60).

The $\text{NO}_3^-_{\text{bio}}$ always shows the $\Delta^{17}\text{O}$ value close to 0 ‰ because its oxygen atoms are derived from either terrestrial O_2 or H_2O through nitrification. Contrarily, the $\text{NO}_3^-_{\text{atm}}$ always displays an anomalous enrichment in ^{17}O with $\Delta^{17}\text{O}$ value being approximately $+26 \pm 3$ ‰ in Japan (Tsunogai et al., 2010, 2016; Ding et al., 2022, 2023) because of oxygen transfers from atmospheric ozone (Michalski et al., 2003; Nelson et al., 2018).

Using the deposition flux of $\text{NO}_3^-_{\text{atm}}$ into the catchment and the leaching flux of unprocessed $\text{NO}_3^-_{\text{atm}}$ and $\text{NO}_3^-_{\text{bio}}$ from streams, the GNR in a forested catchment was estimated similarly to the estimation for water environments (Fang et al., 2015).

L. 76: in this equation, some processes are denoted as subscript of NO_3^- (like deposition) while others are denoted for themselves (like GNR). GNR and GDR are usually expressed as a nitrogen rather than as a nitrate flux. As it is written, the equation lets it open. It would be better to explicitly express all rates either as N or as NO_3^- .

Thank you for your suggestion. Our simulation was done for the forested catchment reported by Hattori et al. (2019). Thus, the symbols used in the manuscript were in accordance with Hattori et al. (2019) as well. We mentioned this in the revised manuscript (P8/L144-145).

All the symbols (e.g., GNR) used here were consistent with those of Hattori et al. (2019).

L. 74-80, 85-90: it is not clear why the word "each" is always used for the catchments (not only here, in general in the text).

We removed the "each" in the revised manuscript.

L. 112-113: repeated usage of the word "limited".

We revised this in the revised manuscript (P7/L113).

L. 116: which one of the $\Delta^{17}\text{O}$ is this? Or is it the difference?

The $\Delta^{17}\text{O}$ denotes the $\Delta^{17}\text{O}$ of stream nitrate. We added this in the revised manuscript (P7/L118).

L. 127-128: fine roots would be much more relevant than the total root biomass (with coarse roots obviously overrepresented close to the stem and thus close to the surface).

Thank you for your suggestion. We added the information in the revised manuscript (P8/L128-131).

As most fine roots and root biomass are concentrated in the top 10 cm of the soil in forested catchments (Jackson et al., 1996; Li et al., 2020), most uptake reactions should occur in that top 10 cm of soil.

L. 146-148: it may be useful to explain this as a gradual uptake (consumption) of nitrate as water moves down the profile.

Thank you for your suggestion. We added the information in the revised manuscript (P9/L151-152).

This simulated the gradual net consumption of NO_3^- in accordance with water flow in forested soils.

L. 152-160: these assumptions are obviously simplifications compared to real measurements, but they make sense for the demonstration. It would be interesting to test also the assumption of nitrate fluxes increasing with depth because of a positive net nitrification.

Thank you for your suggestion. We have simulated the positive net nitrification in the soil layers above.

L. 175-179: long sentence.

We revised the sentence in the revised manuscript (P11/L195-198).

However, the simultaneous observations of the oxygen isotopes of soil NO_3^- and stream NO_3^- (Hattori et al., 2019; Osaka et al., 2010; Nakagawa et al., 2018; Rose, 2014) implied that the approximation of the $\Delta^{17}\text{O}$ values of soil NO_3^- to that of the stream NO_3^- (Fig. 2b) was unrealistic.

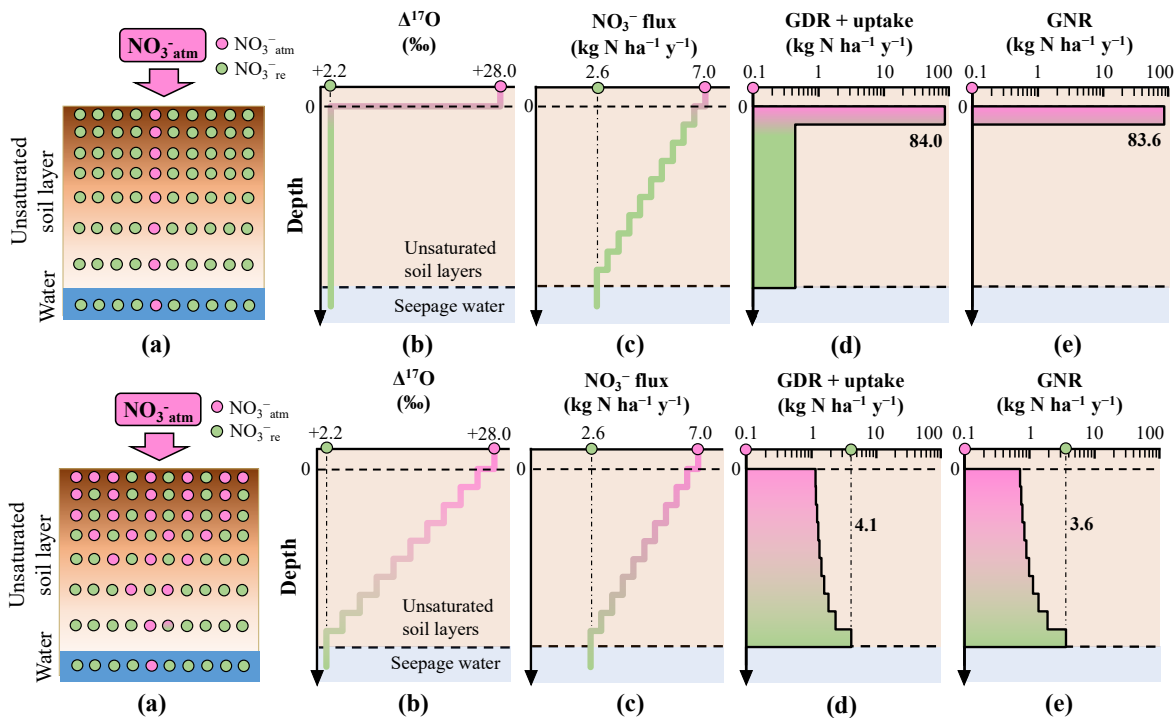
L. 220: as written, it is like anonymous reviewers would be named, which does not make sense.

We revised the sentence in the revised manuscript (P13/L228-229).

We thank Dr. Joel Bostic, Dr. Lucy Rose and other two anonymous referees, for their valuable remarks on an earlier version of this paper.

Fig. 1, fig. 2: the soil does not float above water and therefore "soil layers" and "water layer" should rather be marked "unsaturated soil layers" and either "water-saturated soil layer" or "seepage water" (as these two are considered to exhibit the same flux).

We revised the "soil layers" and "water layer" to "unsaturated soil layers" and "seepage water" in the revised manuscript, respectively.



Reference

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- Osaka, K., Ohte, N., Koba, K., Yoshimizu, C., Katsuyama, M., Tani, M., Tayasu, I., and Nagata, T.: Hydrological influences on spatiotemporal variations of $\delta^{15}\text{N}$ and $\delta^{18}\text{O}$ of nitrate in a forested headwater catchment in central Japan: Denitrification plays a critical role in groundwater, *Journal of Geophysical Research: Biogeosciences*, 115, n/a-n/a, <https://doi.org/10.1029/2009jg000977>, 2010.
- Rose, L. A.: Assessing the nitrogen saturation status of appalachian forests using stable isotopes of nitrate [PhD thesis, University of Pittsburgh]. Retrieved from http://d-scholarship.pitt.edu/22783/1/LRose_ETD_081914_revised1.pdf.

Response to the referee #2:

The authors assume in Equation (4),

$$\Delta 17\text{O}(\text{NO}_3)\text{uptake} = \Delta 17\text{O}(\text{NO}_3)\text{denitrification} = \Delta 17\text{O}(\text{NO}_3)\text{stream}.$$

However, this assumption is not necessarily correct. It requires the assumption that nitrates deposited from the atmosphere are first diluted by nitrification (increasing nitrate amount with decreasing D17O) and then (i.e., “afterward”), reduced in nitrate amount without changing D17O by uptake and/or denitrification.

Who assumed Eq. (4) were the authors of the papers in which Eq. (6) had been used to estimate GNR, such as Fang et al. (2015), Hattori et al. (2019), and Huang et al. (2020). While none of the authors clarified that they had assumed Eq. (4) in their papers, Eq. (4) should be needed to derive Eq. (6) from Eqs. (2) and (3). In addition, we also presented that this assumption (Eq. (4)) is not necessarily correct. In short, you have the same opinion with us at least on this point.

Another reverse possibility could be that atmospheric nitrates are reduced in quantity through uptake and/or denitrification without changing D17O, and then nitrates are added through nitrification (by decreasing D17O). In this assumption, one could hypothesize:

$$\text{D17O_uptake} = \text{D17O_denitrification} = \text{D17O_atm} \quad (\text{A1}),$$

and calculate GNR as follows:

$$\text{GNR} = \text{NO}_3\text{_st} \times (\text{D17O_atm} - \text{D17O_st}) / \text{D17O_st} \quad (\text{A2}).$$

To compare using Equation (4) versus Equations A1 and A2, let's assume a system where 100 nitrates (assuming D17O is 24‰) are initially deposited. In this case, when suppose the stream water nitrate is also 100 but with D17O decreased to 3‰. Using the same assumption as the authors (using Eq. 4 and 6), GNR is calculated as 700 using Equation (6) in the manuscript ($\text{GNR} = 100 \times (24-3)/3 = 700$). However, assuming A1 and A2, GNR can be calculated as 87.5 ($\text{GNR} = 100 \times (24-3)/24 = 87.5$), which is an extremely lower result compared to another case. Yet, in both outcomes, the final stream water remains the same at 100 in nitrate amount and 3‰ in D17O of nitrate from the same starting point (100 of nitrate with D17O = 24‰). It is necessary to find a converging point by differentiation, and it can be understood that this is the “heterogeneous” method assumed by the authors in the manuscript with 10 soil layers. In the above-mentioned case, a GNR of ~208 will be the case when considering production and consumption occur simultaneously, as far as I calculated briefly (dividing layers > 1000).

The equation A2 you wrote may be a typo. Under the assumption of A1, A2 should be:
$$\text{GNR} = \text{NO}_3\text{_st} \times (\text{D17O_atm} - \text{D17O_st}) / \text{D17O_atm} \quad (\text{RA2})$$

The equation A1 can be possible for forested catchments in which possible variations

in both the leaching flux and $\Delta^{17}\text{O}$ values of soil NO_3^- were not determined for each soil layer. When we apply the equation RA2 to the forested catchment we used for the simulation (i.e. the forested catchment studied by Hattori et al., 2019), we obtain much smaller GNR of 2.4 kg of N $\text{ha}^{-1} \text{y}^{-1}$ ($\text{GNR} = 2.6 \times (28-2.2)/28 = 2.4$) compared to the GNR calculated by using Eq. (6) (83.6 kg of N $\text{ha}^{-1} \text{y}^{-1}$; $\text{GNR} = 7.0 \times (28-2.2)/2.2 = 83.6$) that had been used in the literatures (Fang et al., 2015; Hattori et al., 2019; Huang et al., 2020). Thus, you reached the same conclusion with us that the GNR estimated from Eq. (6) using the $\Delta^{17}\text{O}$ values of stream nitrate was, to some extent, an overestimate of the actual GNR.

In reality, production and consumption occur simultaneously. Therefore, both cases may overestimate or underestimate GNR to an extreme.

Both nitrification and consumption (uptake + denitrification) of NO_3^- usually occur simultaneously in forested soil, as you pointed out. This is the reason we done a simulated calculation for the case shown in Figure 1 in the manuscript, in which both nitrification and consumption of NO_3^- occur simultaneously in the soil.

Thus, authors should consider this case considering equations A1 and A2, in addition to the case considered in this study.

Thank you for your advice. The aim of this paper is to clarify that the GNR estimated by using Eq. (6) was not the only GNR that can be expected in each forested catchment. Rather, the GNR estimated by using Eq. (6) often overestimate actual GNR to some extent. We trust that the case shown in Figure 1 is sufficient to accomplish our aim shown above.

Additionally, the authors have limited their verification of GNR calculation overestimation in their manuscript (underestimation in the case of A1 and A2 in this review report) to the soil profile. However, if pointing out such overestimation in GNR calculation methods, it would be better to also consider similar considerations for N cycling rates (e.g., GNR) calculated for lake systems, as advanced by the authors' group. Hasn't there been an overestimation for similar reasons in studies using nitrogen cycling rates in Lake systems, as shown in Tsunogai et al. (2011 and 2018) and other previous research? In lake and/or river studies, might they have calculated rates assuming that nitrates are added by nitrification (increasing the amount and decreasing D17O), and then the amount reduces by uptake and denitrification without changing D17O “only once” within each observation period unit (monthly or quarterly)? Wouldn't both assumptions based on Equation 4 and those similar to A1 and A2 be equally valid? Assuming simultaneous production and consumption as in lake mass balance calculations, converging to a single value might provide a more reliable N cycle rate. It should

also be pointed out that the authors' group's previous N cycling research may have been overestimated.

Your claim on our studies applying the $\Delta^{17}\text{O}$ tracer to water environments is wrong. In case of the water environments, differ from forested catchments, the $\Delta^{17}\text{O}$ values of NO_3^- were mostly homogeneous in the water column due to the active vertical mixing in the water column during cold seasons and storm events. Additionally, the homogeneity of the $\Delta^{17}\text{O}$ values had been verified through actual observation prior to calculating GNR (Tsunogai et al., 2011, 2018). Furthermore, the extent of heterogeneities of the $\Delta^{17}\text{O}$ values in the water column had been evaluated in calculating GNR etc., so that the calculated values of GNR were reported with the ranges of errors (Tsunogai et al., 2011, 2018). These are the essential differences between the past studies on the water environments and those on the forested catchments using Eq. (6) to estimate GNR.

Especially, since Tsunogai et al. (2018) concluded that the nitrogen cycle rate was faster compared to ^{15}N tracer experiments, which makes their study significant, it is important to consider the possibility of overestimation. Overall, this manuscript should consider and comment also on the case of their application for other systems like lake/river.

Your understanding on Tsunogai et al. (2018) is wrong. Please note that the difference in the fluxes between the $\Delta^{17}\text{O}$ method and the ^{15}N tracer method estimated in a water environment by Tsunogai et al. (2018) was only 20 % on the annual base, while the difference in the forested catchment between the calculation methods was more than 500 %. In addition, Tsunogai et al. (2018) had discussed the reason for the difference (20 %) between the $\Delta^{17}\text{O}$ method and the ^{15}N tracer method in detail in the paper and concluded that the differences in the period of observation (instantaneous for the ^{15}N tracer method vs. long-range average for the $\Delta^{17}\text{O}$ method) were primarily responsible for the discrepancy so that the reason was essentially different from the discrepancy in the forested catchment. We don't see any merit in discussing the water environments again in this manuscript.

Based on the above two major comments, here are some suggestions for the cases considered in this study:

- 1. Consider that the case of Equation (4) may not always be correct.**

As we already explained, those who assumed Eq. (4) were the authors of the papers in which Eq. (6) had been used to estimate GNR, such as Fang et al. (2015), Hattori et al. (2019), and Huang et al. (2020). We presented that this assumption (Eq. (4)) is not necessarily correct, in lines from 66 to 68 and from 131 to 134 in the revised manuscript, so that you have the same opinion with us on this point.

Consider also the case assuming Equations A1 and A2 provided in this review report.

As we already explained, the aim of this paper is to clarify that the GNR estimated by using Eq. (6) was not the only GNR that can be expected in each forested catchment. Rather, the GNR estimated by using Eq. (6) often overestimate actual GNR to some extent. We trust that showing the case Figure 1 is sufficient to accomplish our aim.

2. Instead of comments using other group's case as an example, verify the calculation process and resulting GNR in the more general system.

As presented in the manuscript (L199-209 in the revised manuscript), our conclusion is that it is impossible to estimate reliable GNR in each ecosystem (e.g., forested catchments, lakes, glaciers) in general using $\Delta^{17}\text{O}$ as a tracer without measurement on the $\Delta^{17}\text{O}$ values of NO_3^- actually consumed in each ecosystem. It is impossible to present the calculation process in the more general system without actual observation.

3. Not only soil profile cases, but also consider possible changes for the other systems led by their research group (e.g., Tsunogai et al. 2011 Biogeos; Tsunogai et al. 2018 L&O).

In case of the water environments, differ from forested catchments, the $\Delta^{17}\text{O}$ values of NO_3^- were mostly homogeneous in the water column due to the active vertical mixing in the water column during cold seasons and storm events. Additionally, the homogeneity of the $\Delta^{17}\text{O}$ values had been verified through actual observation prior to calculating GNR (Tsunogai et al., 2011, 2018). Furthermore, the extent of heterogeneities of the $\Delta^{17}\text{O}$ values in the water column had been evaluated in calculating GNR etc., so that the calculated values of GNR were reported with the ranges of errors (Tsunogai et al., 2011, 2018). These are the essential differences between the past studies on the water environments and those on the forested catchments using Eq. (6) to estimate GNR. We added the new information to emphasized this in the revised manuscript (P7/L114-116)

Differ from water environments, vertical mixing of water/soil is difficult in forested soil, so the $\Delta^{17}\text{O}$ values of soil NO_3^- are often heterogeneous.

I also note that the current manuscript seems to criticize other groups' research, which may be due to language issues, so I want to avoid pointing out each by each in this review report. However, it might be worthwhile for the authors to reflect similar self-criticism on their group's previous nitrogen cycle research.

To be honest, the current manuscript feels like an incomplete consideration that

criticizes others' research one-sidedly. I also note that a similar modification of the calculation way for GNR based on D17O, considering both the production and consumption of nitrate simultaneously, has been already considered/published in another paper (Hattori et al. 2023).

Because we found a problem in applying the $\Delta^{17}\text{O}$ method to forested catchments by using Eq. (6) to estimate GNR as Fang et al. (2015) did, we just ignored and did not estimate GNR in our subsequent manuscripts studying forested catchments using $\Delta^{17}\text{O}$ of NO_3^- as a tracer, such as Nakagawa et al. (2018) and Ding et al. (2022, 2023). Because no one pointed out the problem to use Eq. (6) in calculating GNR in forested catchments, however, apparently overestimated GNR by using Eq. (6) became “normal” in the papers subsequent to Fang et al. (2015) (Hattori et al., 2019, Huang et al., 2020), which seems to have reduced reliability of the $\Delta^{17}\text{O}$ method. We trust this paper is worthy of publication in Biogeosciences to clarify the problem inherited in this method. Concerning to Hattori et al. (2023), please note that the first preprint of our paper was published on 12 Jan 2023 (<https://bg.copernicus.org/preprints/bg-2022-236/>). Because this was 5 months earlier than the submission of Hattori et al. (2023), who should “consider” must be the authors of Hattori et al. (2023).

Title: It is better to replace “error” with “bias”?

Thank you for your advice. We revised the “error” to “bias” in the revised manuscript.

L149: Why 10 layers? If you consider fewer or more layers, do you expect any changes?

Thank you for your questions. Dividing the forested soils into more layers can enhance the precision of the simulated GNR. By dividing the forested soils into 10, 20, 30, 50, 100, and 1000 layers, the simulated GNR was 13.0, 11.4, 11.0, 10.5, 10.3, and 10.1 kg of N ha⁻¹ y⁻¹, respectively. We added this information to the revised manuscript (P10/L171-174).

Even if the number of layers in the forested soils was increased to 20, 30, 50, 100, and 1000 to enhance the precision of the GNR simulated for the catchment with the heterogeneous soil, the GNR was 11.4, 11.0, 10.5, 10.3, and 10.1 kg of N ha⁻¹ y⁻¹, respectively.

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