

Reply to your comment (Referee #2).

Thank you very much for your valuable comments on our manuscript.  
We have responded to each of your comments and questions.

> First, the watersheds (KJ, IJ1 and IJ2) have extremely different size (3.84, 298, 108 ha respectively) and there are no information concerning their geology and soil depth/quality which are important parameters to interpret stream discharge and nutrient flow.

> Experimental section: KJ, IJ1 and IJ2 have very different size. Here (and probably in the discussion) you should somehow reassure the reader that it is not a source of bias in your interpretation.

In the same Niigata Prefecture in which KJ is located, Koshikawa et al. (2011) determined stream chemistry from streams (n=62) having various catchment areas, ranging from 0.007 km<sup>2</sup> (=0.7 ha) to 18 km<sup>2</sup> (=1800 ha). They performed primary component analysis (PCA) of the various factors possibly related to the stream chemistry including nitrate concentration, and, they could not find any significant relation between stream nitrate concentration and catchment area. Thus, the differences in the catchment area (from 0.7 to 1,800 ha) had little impact on the stream nitrate concentration. As a result, we targeted site KJ (3.94 ha) for analysis. We would like to present this information in section 2.1 along with this reference.

Koshikawa, M., M. Watanabe, T. Takamatsu, S. Hayashi, S. Nohara, and K. Satake (2011) Relationships between stream water chemistry and watershed geology and topography in the Miomote River System, Niigata, Japan. *Japanese Journal of Limnology*, 72, 71-80 (in Japanese with English abstract).

The geology and soil quality of the sites has been previously studied. In brief, the bedrock consists of granodiorite, and brown forest soils have developed at site KJ (Kamisako et al. 2008; Sase et al. 2008). The bedrock consists of the chert (90%) and mudstone (10%) of Middle Jurassic to Early Cretaceous age at sites IJ1 and IJ2 and the dominant soil type was brown forest soils (Nakahara et al., 2010). We will add information on geology and soil quality in the revised MS.

> Second, stream discharge is estimated with a different method for each stream (section 2.2).

> Section 2.2: Discharge is estimated with different methods for each stream. This is a source of variation for your results and you should acknowledge that somewhere in the discussion to convince the reader you took this issue into account in your interpretation.

Each method to estimate stream discharge was the same as those used in previous studies (site KJ: Kamisako et al. 2008; IJ1 and IJ2: Nakahara et al. 2010). Both methods are commonly used well established methods to measure stream discharges (U.S. Department of the Interior Bureau of Reclamation, 1997). Additionally, Kamisako et al. (2008) confirmed that the water balance was justified at site KJ, in which the evapotranspiration loss was

estimated based on Thornthwaite's (1948) method. We concluded that the discharge rate at site KJ was accurate (within the error range of 10%) and used in the discussions in this MS. In IJ1 and IJ2, there were no prior studies on water balance. The estimated annual discharge via the stream (2,057 mm on average during 2013–14) corresponds to 62% of the annual deposition in IJ1 (3,310 mm on average during 2013–14). Because the evapotranspiration loss from forested catchments in Japan was estimated to be 30 to 40% of deposition under the annual deposition rate of 3000 mm (Ogawa et al., 2003), we concluded that the estimated annual discharge via the stream was highly reliable in the sites as well, within the error range of 10%.

We will add this information concerning the water balance in the revised MS, together with references.

Ogawa, S. (2003) Chapter 3, Forests and water resources, In: Hydrological Cycle and Local Metabolic System of Water (eds. N. Tambo and T. Maruyama), Gihodo, pp. 45-71 (in Japanese).

U. S. Department of the Interior Bureau of Reclamation (1997) Water Measurement Manual. US Government Printing Office: Washington, DC.

> Third, atmospheric nitrate deposition rates were also obtained with different approaches in the three watersheds (section 2.4).

> There is the same risk of discrepancy with the way atmospheric nitrate deposition rates were obtained (section 2.4).

While the deposition rates of atmospheric nitrate at sites IJ1 and IJ2 were determined at a National Acid Rain Monitoring station based on the EANET protocol (EANET, 2014), those at site KJ were determined by a simple on site observation in the forested field, so that the deposition rates could be biased to the same extent, as presented in sections 2.4 and 3.1.

As a result, we compared the deposition rate at site KJ with those determined at the nearby national atmospheric observation site (Sado-seki National Acid Rain Monitoring station), where the deposition rates were determined based on the EANET protocol (the same protocol with IJ1/IJ2 sites), and found that the deposition rates at site KJ agreed (< 10% difference) with those determined at the Sado-seki National Acid Rain Monitoring station. Thus, we concluded that the methodological differences had little impact on the final results and we used the results obtained through the observation at site KJ, allowing a moderate error range (10%). (See section 3.1 for details)

> Finally one can wonder how much data variation is carried by each of these points and how this influences the results and their interpretation. My guess is that this should be at least acknowledged, analysed and discussed thoroughly to convince the reader.

For the isotopes ( $\delta^{15}\text{N}$ ,  $\delta^{18}\text{O}$ , and  $\Delta^{17}\text{O}$ ), we repeated the analyses at least three times and obtained the results presented in the manuscript as the average (P10/L15). While the errors associated with the  $\Delta^{17}\text{O}$  values directly influences the errors associated with the  $C_{\text{atm}}/C_{\text{total}}$  ratios and  $M_{\text{atm}}/M_{\text{total}}$  ratios, their influences on the values of  $M_{\text{atm}}$ , and  $M_{\text{atm}}/D_{\text{atm}}$  ratios were minor. Rather, the errors associated with the flow rates ( $V$  in Eqs. (3) and (4)) and the deposition rates ( $D_{\text{atm}}$ ) had much larger impact on the  $M_{\text{atm}}$  and  $M_{\text{atm}}/D_{\text{atm}}$  ratios.

We will make the suggested revision in section 5, where we have presented the major factors influencing the errors associated with the  $M_{\text{atm}}$  and  $M_{\text{atm}}/D_{\text{atm}}$  ratio.

> Abstract: L21: I suggest you give the annual fluxes of nitrate instead of concentrations.

We will make the revision suggested.

> Abstract: L23-25: It is misleading to compare, in the same sentence,  $^{17}\text{O}$ -excess in KJ soil water with those of KJ, IJ1 and IJ2 stream water. Add “in KJ” after “+0.1‰ to +5.7‰”.

We will make the revision suggested.

> Abstract: L25: ”was groundwater nitrate”.

We will make the revision suggested.

> Abstract: L25: I would also remove the end of the sentence “which buffered the seasonal. . .” as it is confusing.

We would like to revise here.

> Introduction: P2-L5 (and throughout the paper): Do not use the word “enrichment” when you simply mean “increase”, especially in a paper dealing with isotopes for which “enrichment” is usually used as in P4-L7.

We will make the revision suggested.

> P2-L25: There are other processes that should be mentioned: DNRA for instance.

We will change No. 4 to “(4) the removal of nitrate through dissimilatory reduction by microbes”.

> P3-L2-3: Remove the end of the sentence “, including [ . . . ] catchments.”, it does not bring new information.

We will revise here to bring new information.

> P3-L4: composition. . .has been (no plural)

We will make the revision suggested.

> P3-L8: “has survived”

We will make the revision suggested.

> P3-L13: remove “nitrate including”

We will make the revision suggested.

> P3-L21-22: this sentence is useless. I miss some clear hypotheses at the end of the introduction.

We will revise this sentence, and add a sentence here to present the hypothesis of this study more clearly.

> Section 1.2 is Materials and Method.

In section 1.2, we presented the background of this study (regarding the  $\Delta^{17}\text{O}$  tracer of nitrate), because this tracer is not very familiar for those studying Biogeosciences.

> P4-L6: atmospheric nitrate is not only produced by photochemical reactions.

We will add “most of” prior to “which is produced via photochemical reactions ...”.

> P4-L13: specify this is Equation 1

> P4-L24: Specify this is Equation 2

We are very sorry, but the numbering to the equations seems to have been removed while we were arranging formatting the paper for Biogeosciences Discuss. We will make this revision.

> Here you use  $C_{\text{atm}}$  and  $C_{\text{total}}$  for the first time. In the rest of the manuscript they represent either mean daily or mean annual concentrations which is confusing. Please use different codes for daily and annual.

We now use  $C_{\text{atm}}$  and  $C_{\text{total}}$  only for daily mean concentrations (i.e., concentrations in each sample) and use different characters ( $\bar{C}_{\text{atm}}$  and  $\bar{C}_{\text{total}}$ ) for annual mean concentrations in the revised MS.

> Experimental section: P5-L15: Remove “continuous” or replace by “weekly” as samples were collected once a week.

We will make the revision suggested.

> P5-L25: “2500 mm” seems a lot as the average precipitation in Shibata is 1263 mm.

1. The average precipitation in Shibata is estimated to be 1920 mm (<https://en.climate-data.org/region/2441/#example2>). Your precipitation data (1263 mm) may be inaccurate.

2. The annual precipitation determined at the AMeDAS (Automated Meteorological Data Acquisition System) observatory nearby (Nakajyo; 38°3.6'N; 139°24.5'E) is 2110 mm (2012), 2746 mm (2013), and 2506 mm (2014), so that the precipitation in Nakajyo supported the “2500 mm” value. You can find the data of AMeDAS on the web site of Japan Meteorological Agency (<http://www.jma.go.jp/jma/indexe.html>).

3. While Shibata is located on a flatland, Nakajyo is located at the eastern foot of the Kushigata mountain range so that the precipitation at Nakajyo is higher than that at Shibata. Because site KJ is located at the eastern edge of the Kushigata mountain range, we conclude that the precipitation at Nakajyo represents the precipitation in KJ.

> P6-L9: “3300 mm” seems a lot. Actually if I do the maths with your figures I get 1500 mm for 10 months/yr.

1. The annual precipitations determined at the AMeDAS observatory nearby (Tarumi; 35°38.3'N; 136°36.2'E) was 3414 mm (2012), 3021 mm (2013), and 3497 mm (2014), so that the precipitation data in Tarumi support the “3300 mm” value.

2. Figure 6 (a) shows the flow rate at IJ1. When we integrate the flow rates determined in this study (Figure 6 (a)), the annual discharge via the stream corresponds to 2057 mm for the catchment (298 ha) on average from 2013 to 2014, which almost corresponds to your rough estimate (1500 mm). To compare annual discharge with precipitation, however, we should consider the evapotranspiration loss from the catchment as well. Because the evapotranspiration loss from forested catchments in Japan was estimated to be 30 to 40% of deposition under the annual deposition rate of 3000 mm (Ogawa et al., 2003), we think the annual precipitation of 3300 mm is accurate for IJ1.

Ogawa, S. (2003) Chapter 3, Forests and water resources, In: Hydrological Cycle and Local Metabolic System of Water (eds. Norihito Tambo and T. Maruyama), Gihodo, pp. 45-71 (in Japanese).

> P6-L23: I am not sure *Clethra* sp. and *Ilex* sp. can be considered as trees.

While there is no precise botanical definition for tree, we confirmed that both *Clethra harbinervis* and *Ilex pedunculosa* were classified as trees in a Pictorial Book of Flora in Japan.

> P7-L1: You do not use water T, pH, alkalinity and EC in the paper. Please remove. In fact, maybe it could bring new insights in your data set and interpretation.

While these data were routinely obtained to check the reliability of sampling, we do not present the data as you pointed out, because we could not find anything new in the results than those reported in previous studies (Kamisako et al., 2008; Nakahara et al., 2010). We are presenting this data in a supplement.

> Section 2.3: what SLS and SMS stand for? You need to explain why you decided to sample next to the stream and 20m upland, and why at 20cm and 60cm depth.

The depths were chosen as the length of the porous cup soil solution sampler (20 cm and 60 cm). We will add this in the revised MS, in response to your request.

The three soil water sampling stations were chosen on the transect from the sampling station for atmospheric deposition toward the direction perpendicular to the slope (and thus almost perpendicular to the stream). In addition to the lower station (SLS) and middle station (SMS) on the transect, there was one station (SUS) at the upper end of the line. The SUS station, was hard to access and this limited the number of samples collected there (most of them were taken only in summer). Further, the volumes of solution for isotope analysis were also limited. As a result, we did not analyse isotopes for the samples and thus, we did not present the results.

Because we could not find any significant differences in the isotopes irrespective of the sampling depths and stations (Fig. 3), we concluded that the data on isotopes obtained at SLS and SMS represented that of soil solutions at the site. We will emphasize this in the revised MS.

> P7-L22: “between December and March”

We will make the revision suggested.

> Section 2.5: I am surprise samples were not acidified prior storage.

The oxygen isotope exchange reaction between  $\text{NO}_3^-$  and  $\text{H}_2\text{O}$  becomes much faster under low pH conditions (Böhlke et al., Rapid Commun. Mass Spectrom., 17, 1835-46, 2003; Kaneko and Poulson, Geochim. Cosmochim. Acta, 118, 148-156, 2013). We cannot store samples for oxygen isotope analyses of nitrate under acidified conditions.

> P9-L15: remove “, the procedure [. . .]. Approximately”

We will make the revision suggested.

> P10-L22: You mention NO<sub>2</sub> as a possible source of variation, what about NH<sub>4</sub>?

Nitrogen compounds that can be converted to N<sub>2</sub>O on addition of azide (N<sub>3</sub>H) can interfere with the results. To convert NH<sub>4</sub><sup>+</sup> to N<sub>2</sub>O, we must add an oxidizing agent (such as NO<sub>2</sub><sup>-</sup>), while azide is a reducing agent. Then, NH<sub>4</sub><sup>+</sup> cannot interfere with the results.

> Section 2.7: I suggest you give the units for each Equation.

Because these are general equations and no actual value was presented, there are no constraints on the units.

> P12-L1: replace “obtain” by “estimate”

We will make the suggested revision.

> Results: P12-L16-21: You can remove this paragraph which does not bring new information compared to the figures legends.

We will make the suggested revision.

> P12-L25: Could you add ranges or values to F<sub>total</sub> and F<sub>atm</sub>. Same thing for IJ1 and IJ2.

We will make the suggested revision.

> P14-L20-22: You can remove this sentence which does not bring new information compared to the figures legends.

We will make the suggested revision.

> Discussion: Specify to which category (catchment groundwater, through flow) belongs the water sampled in SLS and SMS.

We will make the suggested revision.

> P16-L25: Add to Fig 4 legend that the hypothetical mixing line is reported.

We will make the suggested revision.

> P17-L1: replace “buffered by” by “diluted in”

While we estimated that the total quantity of groundwater nitrate was much more than that of soil nitrate, the concentration of soil nitrate was not always higher than that of groundwater nitrate. As a result, we cannot agree with you on the use of the word “dilute” here for nitrate. We will revise here, in response to your request, but different way than what you suggest.

> P18-L22: “This delay time reflects the magnitude and flow of the nitrate. . .”

We will make the suggested revision.

> P19-L23-24: A figure showing the relationship between  $M_{atm}$  and stream  $NO_3$  conc. would be welcome.

We will add this as a new figure 8(c).

> P20-L3: replace “N” by “ $NO_3$ ”

> P20-L4: replace “nitrogen” by “nitrate”.

While we can use  $NO_3^-$  (nitrate) instead of N (nitrogen) in this text, what the original literature (Peterjohn et al., 1996) used was N (nitrogen). We would like to use the same here.

> P20-L8: Fig. 9 does not show that “ $M_{atm}/D_{atm}$  ratios are stable during the progress of nitrification in forested soils”

Please see the locations of the arrows (=flows) representing nitrification. All the arrows related to the  $M_{atm}/D_{atm}$  ratios are shown in red/pink, while those related to nitrification are shown in brown/yellow. The colour differences represented the  $M_{atm}/D_{atm}$  ratios were determined independent of nitrification.

We will add this explanation in the text and the caption to figure 9.

> P20-L17-22: this paragraph is not clear enough. In particular the last sentence compare “stream nitrate enrichment due to nitrogen saturation” with “stream nitrate enrichment due to artificial processes”, which I do not quite understand. Artificial processes (e.g. fertilizers, leguminous fields. . .) are responsible for N saturation. Please clarify.

The “artificial processes” we presented here correspond to the direct contamination processes of nitrate. Increases in stream nitrate concentrations due to secondary changes in the N cycles within forested catchments were not included. We will revise this paragraph to reconfirm this.

> P21-L24: “. . .by 6 and 20 times respectively in accordance. . .”

We will make the suggested revision.

> Fig.2: Underneath RW1 and RW3 there are distances (120m and 40m respectively), are they elevation a.s.l.?

Yes, they are. We will clarify this in the caption of revised MS.

> Fig1 and Fig.2: Please use the same kind of map for both figures.

The base altitude maps (colour altitude maps) are produced by using the same software. While the number of colours used in the altitude map of Fig. 1 was much lower than that for Fig. 3, this is because the variation range in altitudes was much smaller in the Fig. 1 area than that in the Fig. 3 area. In addition to the base altitude map, however, we added contours in Fig. 1 because it was difficult to understand the differences in altitudes in the Fig. 1 area without the contours. We will use the same figures (with contours only in Fig. 1) to clarify high and low areas in the catchments. Also, we will use similar formats for scale bars etc. in the figures, in response to your request.

> Fig.4: Please specify what the line stands for?

We will revise this figure.

We would like to thank you for the helpful comments and suggestions. We trust that our responses to your comments and questions are satisfactory.

Sincerely,  
Urumu Tsunogai

Cc: Fumiko Nakagawa, Yusuke Obata, Kenta Ando, Naoyuki Yamashita, Tatsuyoshi Saito, Shigeki Uchiyama, Masayuki Morohashi, Hiroyuki Sase