

## Topography controls stream NO<sub>3</sub><sup>-</sup> concentration and sediment denitrification at headwater streams

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[Aim] The effects of topography and distribution of electron donors (DOC, and reduced sulfur) for denitrification on stream NO<sub>3</sub><sup>-</sup> concentration and sediment denitrification were examined at 34 headwater streams in the Lake Hachiro watershed. [Materials and Methods] Study catchments are 34 head streams (0.07-16.9 km<sup>2</sup>) in the lake Hachiro watershed at Akita, Japan. Stream water at each catchment was sampled four times a year from 2010 to 2011. Water discharge (L/s) was estimated from width, depth, and velocity measured at 8-10 points along a cross-sectional transect of the stream. At each catchment, sediment samples for denitrification assays were collected once in July, 2011 from the top 5 cm of the streambed across the width of the stream. Dissolved N<sub>2</sub>O in stream water was measured in December, 2011. Water and sediment samples were placed on ice, transported back to the laboratory, and refrigerated overnight until denitrification assays and water analysis were begun. The denitrification assays of the sediments were determined using the acetylene inhibition technique, which inhibits the final step in the conversion of N<sub>2</sub>O gas into N<sub>2</sub> gas. To determine the difference among catchments in the amount of organic C available to the denitrifying organisms, we defined denitrification potential (DP) as the denitrification rate that occurred under anaerobic conditions with abundant NO<sub>3</sub><sup>-</sup> at 25 deg C. Samples of fresh, homogenized soil (15 g) were placed into 150-mL glass bottles. A 50-mL aliquot of solution containing nitrate (5 mg-N L<sup>-1</sup> as KNO<sub>3</sub>) with chloramphenicol was added to the bottles. The bottles were purged with O<sub>2</sub>-free ultrapure N<sub>2</sub> for 3 min to ensure anaerobic conditions, and acetylene (C<sub>2</sub>H<sub>2</sub>) gas was added to a final concentration of 10% v/v (10 kPa) in the headspace. We extracted the headspace gas with a gas-tight syringe, and calculated denitrification rates from the linear portion of the curve for N<sub>2</sub>O production as a function of time. The stream water sample was filtered through 0.45-µm membrane filters and concentrations of dissolved components were determined. The fresh sediment samples were extracted in distilled water (soil:water, 1:5 w/v), and the concentrations of water-extractable soil organic carbon (WSOC) was determined. Easily oxidizable-S (EOS) content in the sediments, which can be an electron donor for denitrification as reduced sulfur, was measured by the difference between H<sub>2</sub>O<sub>2</sub>-S and H<sub>2</sub>O-S content. Topographic index (wetness index) in each catchment was calculated using 10 m grid digital elevation model (DEM) by GIS. [Result and Discussion] Stream NO<sub>3</sub><sup>-</sup> concentrations among catchments had a large spatial variability ranging from 0.06 to 0.52 mg N L<sup>-1</sup>. Stream NO<sub>3</sub><sup>-</sup> concentrations were negatively correlated with topographic index significantly ( $r=0.56$ ,  $p<0.01$ ), indicated NO<sub>3</sub><sup>-</sup> was reduced in a catchment which had a gentle slope area. Dissolved N<sub>2</sub>O in stream water was positively correlated with topographic index, which supported denitrification in such a catchment. A significant positive correlation was also observed between sediment DPs and topographic index. The WSOC content in the sediment, the primary predictor of denitrification rates, increased with topographic index and affected sediment DPs. NO<sub>3</sub><sup>-</sup> concentrations also had a negative correlation with EOS content in the sediments indicating sulfur denitrification in catchments, however, the distribution of EOS did not related topography. Multiple regression analysis showed topography and EOS content in the sediments affected concentration of stream NO<sub>3</sub><sup>-</sup> significantly.

Keywords: nitrate, headwater stream, denitrification, topography