

In situ estimation of new and regenerated production in lakes using triple oxygen isotopes as tracers

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The gross primary production rate is an essential parameter to study biogeochemical processes in hydrosphere, having strong relations with environmental changes in lakes and oceans, such as eutrophication and global warming. Supplying rates of fixed nitrogen, especially dissolved nitrate (NO_3^-) and ammonium (NH_4^+), to each hydrospheric environment often control each gross primary production rate. As a result, the primary production is often divided into the following two categories: “new production” that uses NO_3^- supplied either from atmosphere or from aphotic layer, and “regenerated production” that uses a recycled nitrogen in the form of NH_4^+ or dissolved organic nitrogen excreted or produced during biogeochemical processes within photic layer.

All the above-mentioned parameters had been traditionally estimated based on incubations of sampled water in bottles by adding isotope-labeled compounds such as $^{13}\text{CO}_2$ or $^{14}\text{CO}_2$ for the primary production rates and/or $^{15}\text{NO}_3^-$ or $^{15}\text{NH}_4^+$ for nitrogen uptake rates. In these approaches, however, sampled water in bottles is incubated under artificial conditions that must be somewhat different from actual in-situ conditions and the results could represent different rates from the original in aquatic environments. Moreover, the estimated values based on the incubation corresponds to instantaneous uptake rates when sampling was done so that large errors could be expected for the hydrospheric environments with significant temporal variations, otherwise we must increase a number of observations using time and costs.

In this study, we determined the parameters using natural isotopes in lake-dissolved materials instead of using incubations. Most of the oxygen-containing molecules on earth show mass-dependent relative variation between $^{17}\text{O}/^{16}\text{O}$ ratios and $^{18}\text{O}/^{16}\text{O}$ ratios. On the other hand, atmospheric O_3 photochemically produced from O_2 shows an anomalous enrichment in ^{17}O , so that residual atmospheric O_2 is slightly depleted in ^{17}O in comparison with the mass-dependent relative relation. Besides, at least one of the O atoms in atmospheric NO_3^- is derived from atmospheric O_3 owing to the contribution of O atoms from O_3 during the photochemical oxidation processes of NO_x in atmosphere, so that the triple oxygen isotope ratios ($\Delta^{17}\text{O}$ values) of NO_3^- also deviate from the mass-dependent relative relation. Since $\Delta^{17}\text{O}$ value does not vary during the general mass-dependent reactions such as decompositions, we can estimate the mixing ratio between atmospheric O_2 and photosynthetic O_2 from $\Delta^{17}\text{O}$ value of O_2 and that between atmospheric NO_3^- and remineralized NO_3^- from $\Delta^{17}\text{O}$ value of NO_3^- . If we determine the $\Delta^{17}\text{O}$ values of both dissolved O_2 and NO_3^- in a hydrospheric environment as well as supplying rates of atmospheric O_2 and NO_3^- , we can determine both the primary production rate and NO_3^- uptake rate simultaneously. One of the priorities of this $\Delta^{17}\text{O}$ method is that the estimated rate corresponds to the average value of each rate, so that the values can be a more reliable and accurate than the values estimated from the incubation methods.

In this study, we determined both gross primary production rate and new primary production (NO_3^- uptake) rate simultaneously based on the $\Delta^{17}\text{O}$ value of dissolved O_2 and NO_3^- in two oligotrophic lakes (Lake Shikotsu and Lake Kuttara) and one mesotrophic lake (Lake Biwa) in Japan. The regenerated production rate was then calculated by deducing the later from the former. Water samples were collected twice (spring and summer) in a year for each lake. Both primary production rates and NO_3^- uptake rates were determined from the vertical distribution of $\Delta^{17}\text{O}$ values of O_2 and NO_3^- and their difference between the seasons. We found that the f-ratios (relative use of NO_3^- among the total use of nitrogen) were lower in oligotrophic lakes than in the mesotrophic lake.

Keywords: new production, regenerated production, gross primary production, lakes, triple oxygen isotopes, hypolimnion

Nitrogen isotopic measurement of NO_x gas with the filter-pack method

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Estimation of the nitrogen loss as N₂ and NO_x from ecosystem is quite important to close the nitrogen budget. However, due to the high spatio-temporal variations of the nitrogen dynamics in soils does not allow us to investigate the production/consumption processes of these gaseous forms of nitrogen. Although natural abundance of stable isotope is considered to be able to use for the investigation of the N dynamics with respect to gaseous nitrogen losses, nitrogen isotopic measurement of NO_x is quite difficult due to its high reactivity. Here we present our preliminary work on the nitrogen isotopic measurement of NO_x gas with the filter-pack method (Watanabe et al. 2006) together with the denitrification method (Sigman et al. 2001). NO gas produced from NaNO₂ with known nitrogen isotopic ratio via several chemical treatments, then the trapped NO as NO₂⁻ and NO₃⁻ ions were converted to N₂O with denitrifier, then nitrogen isotopic signature was measured by GC-IRMS. We found that the filter-pack method can be applied for the nitrogen isotopic measurement. We applied this method to measure nitrogen isotopic signature of atmospheric NO_x and present these data in the poster.

Seasonal and spatial variation of dissolved iron transformation kinetics in the Shizugawa bay and its adjacent rivers

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Importance of Fe as an essential nutrient for microorganisms is well recognized such as cellular respiration, photosynthesis and nitrogen fixation. However, the extremely low solubility of thermodynamically stable Fe(III) in oxygenated and circumneutral pH natural waters resulted in low concentration of dissolved Fe in some coastal and oceanic seawaters. The bioavailability of Fe in natural waters can be affected by concentration of dissolved Fe and its redox kinetics, given that Fe(II) has much higher solubility and thus higher bioavailability than Fe(III) does. In the river-coastal dynamic system, changes in land cover, salinity gradients, types of riverine input or organic matter, and seasonal changes may affect to the Fe oxidation kinetics as the Fe(II) oxidation is affected by water qualities such as pH and organic matter. In this study, we mainly focus on the Fe(II) oxidation kinetics in the river-coastal system in order to grasp the seasonal and spatial scales of Fe bioavailability between two different aquatic environments.

The study area was located in Shizugawa bay, Miyagi prefecture (N38° E141°) and totally 6 stations from near to offshore (SB-1 to SB-6) were selected as sampling stations. Also, this study included upstream and downstream of three rivers: Hachiman upstream (HR Up) and downstream (HR Down), Oritate upstream (OR Up) and downstream (OR Down), and Mizushiri upstream (MR Up) and downstream (MR Down). All the surface water samples were collected manually with acid-wash 1 L Nalgene bottles for three consecutive seasons starting from summer year 2014. All water samples were filtered through 0.45 micrometer Milipore membrane and stored in the dark at 4 °C for further analysis.

Fe(II) oxidation rates were determined by measuring time course of Fe(II) concentration using a flow injection analysis (FIA) system with a luminal chemiluminescence detection. Water sample and luminol reagent were both simultaneously pumped into the system using a peristaltic pump with flow rate at 2.4 mL per min. Water sample and luminol mixing was conducted in the flow cell situated at the front of a photomultiplier tube (PMT, Hamamatsu) and the PMT signal was recorded by WA control v91 software. Calibration was conducted for each water sample from three standards addition of Fe(II) (varied between 0.5 and 40 nM depending on signal response of each water sample). The initial signal (i.e., signal when Fe(II) was added) was obtained by extrapolation of signal data collected after 50 seconds back to time zero using a regression line which was obtained from time versus log-transformed signal data. The linear response of initial signal to the nominal initial Fe(II) concentration was found. Thus, the signal value was converted to the Fe(II) concentration by using the linear regression equation. The Fe(II) oxidation rate constant ($M^{-1} s^{-1}$) for each standard Fe addition was derived by assuming a pseudo-first order reaction.

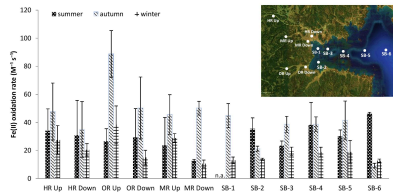
Fe(II) oxidation rates in Shizugawa for three consecutive seasons from summer to winter were shown in Figure 1. The Fe(II) oxidation rates shown here are the average of three standards addition of Fe(II). A distinct seasonal trend of Fe(II) oxidation was observed particularly in autumn season with higher Fe(II) oxidation rates. Generally, Fe(II) oxidation is a pH-dependent reaction. Supposedly, the oxidation rate at seawater pH (>7.9) should be higher than that for typical freshwater pH (e.g. pH ranges between 6.8 and 7.9 in the river investigated). In our study, however, the oxidation rates in coastal seawater tended to be slower compared to those in freshwater. The results of Fe(II) oxidation in freshwater indicated that water quality variables other than pH affect the oxidation process. These factors may include dissolved organic matter concentration and its chemical properties (binding strength) and/or interaction between Fe and other trace metals which will be investigated further.

Keywords: dissolved iron, oxidation, river, coastal

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Aerobic methane production by planktonic microbes under nitrogen and phosphorus starved conditions in a lake

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Lake ecosystems are now recognized as an important source of atmospheric methane (CH₄), which account for about 6-16% of global methane emission from natural sources. In lake ecosystems, it has long been believed that CH₄ is produced only in anoxic environments (e.g., lake sediments and anoxic hypolimnion) by anaerobic methanogens. However, recent empirical and experimental works have revealed that planktonic microbes can produce methane in aerobic water columns of oligotrophic lakes through the use of methylphosphonic acid (MPn) by C-P lyase enzyme under P-limited conditions. But, there is no study examining the effects of cell nutritional conditions (N starved or P starved) on aerobic methane production by bacterioplankton.

We performed the batch-culture experiments to identify the effects of cell starvation on the rate of aerobic methane production under nitrogen and phosphorus limited conditions. Planktonic microbes collected from well-oxygenated water of Lake Saiko (Yamanashi Prefecture) were incubated with a growth medium (BG-11) for several months and used for the starvation experiment to make their cells N-starved or P-starved conditions by removing either element from the BG-11 medium. Then, we added MPn and/or inorganic nitrogen (N_i) and inorganic phosphorus (P_i) to confirm the response of N-starved or P-starved microbes to such experimental additions.

The results showed that although the cell nutritional conditions did not affect the production of CH₄, nutrient balance of lake water (N excess or P excess) greatly influenced the aerobic methane production. First, we confirmed aerobic CH₄ production in the MPn addition treatment, suggesting the active C-P lyase catalysis that converts MPn to methane and inorganic phosphate. Moreover, we found that MPn + N_i addition accelerated the aerobic CH₄ production. This is due probably to the fact that N_i addition promoted the biosynthesis of C-P lyase and/or made lake water more P-limited condition (increase of N/P ratio); both may contribute to increasing the MPn utilization by microbes. However, MPn + P_i addition did not increase the CH₄ production, indicating the opportunistic utilization of MPn alternative to P_i under phosphorus limited conditions. The present results suggest that the input of excess N into lake ecosystems promotes the metabolism of MPn by planktonic microorganisms, which leads to increase of aerobic methane production in phosphorus-limited oligotrophic lakes.

Keywords: Aerobic methane production, C-P lyase, methylphosphonic acid, phosphorus and nitrogen starvation, planktonic microbes

Evaluation for load of bioavailable particulate phosphorus during rain events from Yasu river, at Lake Biwa catchment

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It is well known that primary production in Lake Biwa is limited by phosphorus, and that means phosphorus load into Lake Biwa influence on its environment. In general, it is considered that algae in lake use $\text{PO}_4\text{-P}$ as a phosphorus nutrient, however, it has been revealed that a part of particulate phosphorus (PP) also might be used as nutrient in recent study. However, there are a few studies that quantify the load of bioavailable PP discharged through river in Japan. The purpose of this study is to quantify the load of bioavailable PP discharged through Yasu river into Lake Biwa during rainfall events. Water samples were collected at one to six hours interval in two rainfall event (May and July) in Yasu River using automatic river water collector. We measured several forms of PP by sequential extraction methods (ammonium chloride, bicarbonate dithionite, NaOH, HCl extraction) in river water sample. In rainfall event in May and July, about 70 - 90 % of PP was bioavailable and that part were larger than $\text{PO}_4\text{-P}$ load from Yasu river, indicate that PP discharge from river have large impact on primary production in downward lake.

Spatio-temporal variation of phosphate concentration at a high P concentration groundwater in the Hachirogata polder

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[Aim] The present study was conducted to elucidate spatio-temporal variation of phosphate (PO_4) concentration and the PO_4 release mechanism at a high PO_4 concentration groundwater in the Hachirogata polder, Akita, Japan.

[Materials and Methods] The study site was at a pristine wilderness area after the land reclamation in southwest part of the Hachirogata polder. Groundwater samples were collected once a month from December 2013 to December 2014 from seven groundwater wells at a depth of 3.1, 4.3, 5.6, 6.6, 12.4, 19.5, and 45.0 m, respectively. The 30 m (ϕ 5 cm) boring core sample was collected from near the wells in Feb. 2013. We measured water temperature, pH, and electrical conductivity (EC) using a pH/EC meter (D-54, Horiba, Kyoto, Japan) and measured oxidation-reduction potential (ORP) using a ORP electrode (D-55, Horiba) on site. The water samples were also filtered through a 0.45 μm membrane filter on site. The concentrations of PO_4 in the groundwater were determined using an autoanalyzer (QuAAtro2-HR, BLTEC, Osaka, Japan). The fresh boring sediment was extracted twice with distilled water (soil:water, 1:2.5 for pH and 1:5 for EC), and pH and EC of in the extract were determined using pH/EC meter (LAQUA F-74BW, Horiba). Water soluble P were determined using an autoanalyzer (QuAAtro2-HR, BLTEC, Osaka, Japan). Sediment samples were digested with a combination of $\text{HF-HNO}_3\text{-HClO}_4$ acids in Teflon beakers at 180 $^\circ\text{C}$ and element concentration in solutions were determined by ICP-OES (iCAP 6000, Thermo Fisher Scientific).

[Results and Discussion]

TP content in sediments increased in clay and silt layers while PO_4 content increased in sandy layers. This indicated that clay and silt layers were a P sink/source and released PO_4 was moving in sand layers. Sediment EC increased in clay layer and drastically increased from 21 m deeper layer likely influenced by sea water. Groundwater ORP showed a moderately reducing (-113 ± 42 mV) environment in all the wells. PO_4 concentration in groundwater was ranged from 5.7 to 18.2 mg L^{-1} , and the highest concentration was observed at the well of 6.6 m depth (18.2 ± 0.7 mg L^{-1}) at sandy layer, the second was at the well of 12.4 m depth (10.2 ± 0.9 mg L^{-1}). Positive correlation was observed between PO_4 and F^- concentrations in groundwater ($r = 0.96$, $P < 0.01$, $n = 7$), indicated fluorapatite would be a PO_4 source. At the well of 6.6 m depth, Cl^- concentration was the lowest (29.8 ± 2.9 mg L^{-1}) value and was similar to the lake (Lake Hachiro) water (20.6 mg L^{-1}), suggesting that freshwater was preferentially flowing into the land through groundwater around the depth. At the well of 6.6 m depth that had little fluctuation of water qualities all year round, Na^+ , bicarbonate and carbonate ion were higher but Ca^{2+} concentration was lower compare to other wells. At the well of 12.4 m depth that had a large fluctuation of water qualities, PO_4 , pH, Na^+ , bicarbonate and carbonate ion, and Fe increased while Ca^{2+} and ORP decreased when Cl^- concentration decreased from 72 to 33 mg L^{-1} during May to Oct. 2014. These phenomena indicated PO_4 release induced likely by Na-saturated sediment might be due to release of P associated with oxide surfaces or to dissolution Ca-P at clay layer by increasing pH and dissolution of CaCO_3 (from shell) by inflowing freshwater to the groundwater.

Keywords: phosphate, groundwater, polder, Na saturation, freshwater

Spatial and temporal heterogeneity of the sources of streamwater sulfate in tropical dry forest catchment in Thailand

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In Southeast Asia an increase in emissions of sulfur (S) into the atmosphere may introduce new risks for the plant, soil and inland-water through acidification. However, the effect of the atmospheric S deposition on acidification by an increase in sulfate is poorly understood in tropical forests with possible S sources and processes in the internal cycle. S isotopic ratio ($\delta^{34}\text{S}$) could be a good indicator to identify the source of sulfate in soil and inland-water because only dissimilatory S reduction results in a large fractionation of S isotope. Our objectives are to clarify the spatial and temporal variability of $\delta^{34}\text{S}$ in rainfall, throughfall, soil and stream water within the catchment and discuss the influence of the atmospheric S input on the stream in tropical forest.

Study catchment has been established at dry evergreen forest in Sakaerat silvicultural research station, northeastern Thailand. Anion-exchange-resin columns were installed for rainfall, throughfall, soil-water and stream-water through a year to collect and concentrate sulfate in the field. The sulfate retained in the resin was extracted by NaCl and precipitated as BaSO_4 . We determined $^{34}\text{S} / ^{32}\text{S}$ of the BaSO_4 by mass spectrometer (IR-MS) and calculated $\delta^{34}\text{S}$ (‰) using the reference material (Canyon Diablo Troilite). Annual weighted-mean $\delta^{34}\text{S}$ was calculated from sulfate flux ($\text{kg ha}^{-1} \text{ year}^{-1}$) and $\delta^{34}\text{S}$ in each period. We also determined $\delta^{34}\text{S}$ by the concentration method for the water samples of rainfall and streamwater in some cases.

Annual weighted-mean $\delta^{34}\text{S}$ and S deposition in rainfall were 4.1 ‰ and $6.4 \text{ kg ha}^{-1} \text{ year}^{-1}$, respectively. $\delta^{34}\text{S}$ in streamwater was 4-5 ‰ higher than rainfall during late-wet and dry season, whereas $\delta^{34}\text{S}$ in rainfall and streamwater was mostly comparable during early and middle wet season. In late-wet and dry season, $\delta^{34}\text{S}$ in sub-soil water was particularly higher in the riparian zone near the outlet of the study catchment than in the area near the headwater and on the slope. Sulfate enriched ^{34}S might be increased due to bacterial dissimilatory S reduction in late wet season and retained in the sub-soil during dry season, which could be a main source for the streamwater sulfate during base-flow periods. Meanwhile, in early and middle wet season, streamwater sulfate could be directly affected by atmospheric S input. These heterogeneity of internal S dynamics should be considered to examine the effect of atmospheric deposition on soil and inland-water ecosystems in tropical dry forest. The project is supported by the grant from APN (ARCP2012-18NMY-Sase: ARCP 2013 -13 CMY -Sase).

Keywords: tropical dry forest, stream water, sulfur dynamics, stable sulfur isotope ratio, atmospheric deposition, soil water

Long-term changes in nitrogen discharge from watershed of restored artificial forest

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Forest restoration practice has a greatest impact on nitrogen (N) dynamics in forest ecosystems. While there are a number of studies surveying forest cutting and successive regrowth of vegetation, the extent of increase and its duration of N leaching differs among these studies (Oda et al. 2013). There are mainly three processes that affect N leaching from a forested watershed after cutting; (1) N input via precipitation, (2) N uptake by vegetation and (3) N mineralization and nitrification in soil. To understand the impact of forest cutting both quantitatively and periodically, it is necessary to investigate the long-term changes in these three processes before and after the practice.

In Japan, the area of old-aged forest is now increasing due to the decreases in forest activities and restoration practices (Forest Agency 2013). While old-aged sugi (*Cryptomeria japonica*) lowers nutrient uptake (Ohata 1996), soil N mineralization and nitrification retain considerably high rates (Oyanagi et al. 2004). Therefore, increase in N leaching from old-aged artificial forest is expected.

There are "nitrogen saturated" forested watersheds in the suburban region (Ohruai and Mitchell 1997). Because the amount of cross-border pollutants from continental region is increasing, the nitrogen saturation might become widespread. Therefore, it is necessary to maintain the streamwater quality by enhancing nutrient uptake of forest stands by efficient restoration of artificial forest.

The objective of this study is to clarify the changes in N dynamics caused by cutting and restoration of the artificial forest. We investigated the changes in three processes before and after the partial cutting of old-aged forest which had been in a state of nitrogen saturation.

The study site locates in Field Museum Oyasan in Gunma prefecture, Japan. The watershed area is 1.8 ha, and sugi was planted on the lower to middle slopes, while hinoki (*Chamaecyparis obtusa*) was planted on the upper slope. The sugi plantation on the lowest slope (0.3 ha, 18% of the watershed area) was felled in 2000, and sugi was replanted the following year. The ages of replanted and un-cut old-aged stands are now 15 and 107, respectively.

The long-term hydro-chemical monitoring has been conducted in this site. The amount of N input via precipitation and N leaching from streamwater was estimated from Urakawa et al. (2012). N uptake by sugi before cutting was referred from Oyanagi et al. (2004) and that after cutting was estimated from the aboveground tree biomass surveyed in 2014. Soil net N mineralization and nitrification was measured by in situ incubation conducted intermittently before and after the cutting.

The amount of N leaching from streamwater, which was 10-15 kgN/ha/y before cutting, increased to 15-20 kgN/ha/y for 11 years after cutting, and declined to 10 kgN/ha/y in recent 3 years. By contrast, N input via precipitation maintained stable amount of 9-13 kgN/ha/y. Increases in annual amounts of N mineralization and nitrification were limited for 4 years after cutting, but from the fifth year of the cutting, these rates settled back to the level of the pre-cutting. Rapid growth of replanted sugi began from the 10th year after the cutting, suggesting that the significant decline in streamwater nitrate concentration in recent years attributed to increase in N uptake.

Forest restoration even in 20% of the area was suggested to recover the state of nitrogen saturation caused by aging stands and increase in N input.

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Keywords: long-term monitoring, forested watershed, forest restoration, nitrogen leaching, nutrient uptake by vegetation

Measurement of inorganic nitrogen leaching and its origin in forest soil by resin core method and tension free method

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Nitrogen loading from forest watershed can affect downstream ecosystem, therefore, to clarify the interaction between nitrogen cycle and nitrogen loading from forested ecosystem is important. In this study, we measured nitrogen leaching in forest soil by both resin core method and tension free lysimeter method and estimate nitrate origin by measuring oxygen isotope in nitrate in order to clarify the interaction between nitrogen cycle and nitrogen leaching process. Resin core method are very useful method, however, there are a few studies that compared nitrogen leaching by method and other method. Moreover, there are a few studies that test of measurement of oxygen isotope in nitrate in resin core methods. This study was conducted at two forested watersheds (Aburahi-S and Surumi-A) in Shiga Prefecture. In Aburahi-S, we measured nitrogen leaching by resin core method and tension free lysimeter method, and nitrogen leaching were measured by only resin core method in Surumi-A.

The amount of nitrogen leaching were averaged $0.61 \pm 0.79 \text{ kgN/ha}$ in resin core method and $0.21 \pm 0.26 \text{ kgN/ha}$ in Aburahi-S from May 2014 to February. Little ammonium was not leaching in both methods. In tension free lysimeter method, there is the possibility that unsaturated flow was difficult to collected, and that may lead underestimated of nitrogen leaching at tension free lysimeter method. The results of nitrogen leaching at Surumi-A watershed and oxygen isotope of nitrated will be explain at presentation on this day.

Temporal changes in the soil microbial biomass and N dynamics in eastern Hokkaido

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1. Introduction

In arctic and alpine regions having seasonal snow cover, it has reported that the microbial activity in winter had impact on the annual nitrogen (N) cycling and the soil N availability in the growing season. However, the study focused on the soil microbial biomass and N dynamics from winter to spring in temperate region with seasonal snow cover is limited. In eastern Hokkaido in northern Japan, the soil often experiences soil freezing and freeze-thaw cycle due to the less snowpack in winter. Previous study in this region has reported that the ammonium production rate in soil increased in the late-winter compared to the much snowpack region. However, the pattern of soil microbial biomass and N dynamics from winter to spring is not clear. The objective of this study was to clear the pattern of soil microbial biomass and N dynamics and the relationship between their pattern and the environmental factor.

2. Methods

This study was conducted on the Shibechea Experimental Forest, Kyoto University located in eastern Hokkaido, northern Japan. The main vegetation are Mongolian oak (*Quercus crispula*) and Sasa (*Sasa niponica*) that is understory vegetation. The study plots (5m×15m) were established in the slope of east and west. The plot number in each slope was six. The study period was from October 2013 to September 2014. In each plot, the soil moisture and temperature were measured using moisture sensor and temperature probe at 5 cm depth and 0, 5, 25 cm depth, respectively. The soil sampling from 0 to 10 cm was conducted in almost monthly. In same period, the exchange (collecting and setting) of resin was also conducted. The soil incubation from 0 to 10 cm and 10 to 20 cm was conducted by cylinder method. Collected interval of the incubated soil was from 1 to 2 month. We also measured snowpack and soil freezing depth in winter.

3. Results and discussion

The soil temperature at 0 and 5cm depth showed constantly 0 degree as snowpack increased. Microbial biomass C and N and inorganic N amount in the soil from 0 to 10 cm depth peaked in mid-winter. The inorganic N amount decreased from mid-winter to late-winter. On the other hand, both net N mineralization and nitrification rates showed higher trend in the growing-season than in the winter-season. These results indicated that the soil N utilization by living matter was significantly difference between winter- and growing-season. Although the microbial activity was inhibited by the low soil temperature in winter, the microbes would function as N sink because there is not competition for soil N resource between microbe and plant. The NH₄⁺-N and NO₃⁻-N peaked in October and December, respectively. Then both inorganic N amounts, especially in NO₃⁻-N decreased rapidly, although the NO₃⁻-N leaching at 20 and 30 cm depth was not found in same period. Furthermore, the values of ratio of fungi to bacteria and net nitrification rate decreased from October to March. These results suggested that the change of microbial flora might be important for the N sink in winter. However, the microbial biomass C significantly decreased at end of the April that the critical disappearance of snowpack was measured. These results suggested that the microbe could not tolerate to the freeze-thaw cycle and dramatic rise of soil temperature in the winter-spring transition.

Keywords: nitrogen cycling, soil freezing, freeze-thaw cycle, nitrogen mineralization, nitrogen leaching

Bamboo lodging associated with nitrogen saturation: its morphological and mechanical reasons

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[Introduction]

Nitrogen saturation, a situation of available nitrogen in excess of plant and microbial nutritional demand due to nitrogen deposition, has been suggested to affect plant growth and the root-shoot biomass allocation; an increase in foliar biomass and a decrease in fine root biomass under high nitrogen availability (Aber et al. 1989). In recent years, some reports have suggested that tree windthrow damages may be associated with increased nitrogen status (Braun et al., 2003; Meyer et al., 2008). Here, we report a phenomenon of lodging (falling down) of moso bamboo (*Phyllostachys pubescens*) in a forest site of nitrogen saturation. We suggest that the bamboo lodging is associated with the following morphological and mechanical anomalies; 1) an increase in branches-and-leaves biomass, 2) a decrease in root system, and 3) a decrease in bending strength.

[Materials and methods]

We studied the biometry of bamboo in a nitrogen-saturated site (Tama hill, Tokyo) and control sites (Fukushima and Izu), such as diameter at breast height (DBH), culm height, and mass of branches-and-leaves. Root density and soil nitrate concentration were measured for soil cores 25, 50, 75, 100cm away from culm. The total carbon and total nitrogen content of leaves and culms were measured with dry combustion method. As an index of mechanistic strength, Young's modulus (E) and flexural rigidity (EI) of culm were measured with a bending test of test piece.

[Results and discussion]

In a N-saturated site, leaf and culm nitrogen concentration were significantly higher than control sites. Any elongation growth, which was initially hypothesized, was not observed in a N-saturated site. However, some bamboos in a N-saturated site had significantly larger mass of branches-and-leaves. Very low root density associated with elevated nitrate concentration was also demonstrated, in contrast with a root mat in the soil surface observed in control sites. Culm density and culm thickness showed a negative correlation with bamboo nitrogen concentration. Consequently, culm flexural rigidity (EI) also declined with the increase in nitrogen concentration. A combination of these observed changes, a higher load of canopy, a lower culm strength and a lower uprooting resistance by root system, may be responsible for bamboo lodging observed in the N-saturated site. The results suggest that nitrogen saturation significantly affect morphologies and mechanical properties in bamboo to cause the lodging.

Keywords: nitrogen saturation, lodging, bamboo, fine root biomass, mechanistic strength, morphological change

The impact of nitrogen saturation on tree roots, which lead to uprooting

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[Introduction]

Anthropogenic nitrogen deposition has been increasing for these decades (Galloway, 2004). Some adverse effects on plants have been reported, such as root / shoot ratio, severity of disease, plant-fungal relationships (Veresoglou et al., 2012; Gojon et al., 1994; van Diepen et al., 2010 etc.). However, most of these experiments have used herbs or seedlings; there have been few studies that targeted adult trees in forest ecosystems (Meyer et al., 2008 etc.). We investigated a possibility whether adult tree root biomass and physical properties are affected by elevated nitrogen concentration, which enhances uprooting risks.

[Materials and Methods]

Field research was conducted at a nitrogen-saturation site (Tama-hill, Tokyo), a middle N status site (Karibasaka, Saitama), and a nitrogen-limited control site (Ogawa, Ibaraki). Nitrate concentration in a stream for these watersheds was 280, 86, 16 $\mu\text{mol} / \text{L}$, respectively. The sites are deciduous broad-leaf forests with altitudes between 150 ? 650 m and slopes between 22 ? 35 degrees.

Roots and soils of 0 ? 40 cm depth, for konara oak (*Quercus serrata*) and cherry (*Cerasus jamasakura*), were taken 1 m apart from a stand with a core sampler (7.5 cm diameters). Live roots were sorted into two diameter class of >2 mm and <2 mm and measured for dry weight. Soil nitrate concentrations were measured for water extraction. Wood cores collected from root of these stands with borer auger (5 mm diameters and about 15 cm length) were measured for dry density and Young's modulus (mechanical strength). Angles of stem inclination were measured graphically as an indicator of uprooting risk.

[Results and Discusses]

Dry root weight (both >2 mm and <2 mm) decreased by 60% with the increase of soil nitrate concentration. Cherry's physical properties didn't show any significant changes with different soil nitrate concentrations. However, core's dry density from konara decreased significantly under higher soil nitrate concentrations ($p < 0.01$). Young's modulus was also smaller under higher soil nitrate (more 50 $\mu\text{mol} / \text{kg}$ soil) than lower soil nitrate concentration (less 50 $\mu\text{mol} / \text{kg}$ soil) ($p < 0.05$). Moreover, a part of Konara trees in Tama-hill (N-saturation site) showed distinct stem inclinations as contrast to Ogawa (N-limited site).

The results suggest that nitrogen saturation may have resulted in decreasing root biomass and physical properties to lead to higher uprooting risk.

Keywords: nitrogen saturation, tree root, uprooting

Comparison of SRP (soluble reactive phosphorous) with orthophosphate in riverwater

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Introduction

Orthophosphate is essential nutrient for primary production in waters and it is thought to be the main chemical form of phosphorous directly available to organisms. In oligotrophic and mesotrophic waterbodies, phosphorous often plays as controlling factor of primary production. For the determination of phosphorous, spectrophotometric method based on the formation reduced form of phosphomolybdate (molybdenum blue) is usually applied. Determined value is called as SRP: soluble reactive phosphorous, because various kinds of phosphorous compounds in water also reacts with molybdate by hydrolysis in acidic solution. Molybdenum blue method is very useful but this method does not show practical value of orthophosphate. The authors applied suppressed ion chromatography to determine solely orthophosphate. Obtained phosphate concentrations were compared with the values of SRP concentration for inflowing rivers of Lake Biwa, Japan (phosphorous limiting and mesotrophic freshwater lake).

Materials and Methods

River water samples were collected 4 times from April to November in 2014 at 7 inflow rivers (Yasu, Amano, Ta, Ane, Yogo, Nishino Creek of Yogo, Ado) of Lake Biwa, Shiga Prefecture, Japan. Water samples of Seta River, the only outflow river were also collected. Samples were filtered with a Nuclepore membrane filter (0.2 μ m pore size) and stored in a cool dark container below 10 degree in celcius. Orthophosphate concentration was measured by suppressed ion chromatography. Dionex AS-23A analytical column (250 was with electrochemical suppressor in electric suppression mode. Injection of high volume sample enhanced detection limit of orthophosphate to 10 nmol/L or less. SRP was measured according to the method JIS K0102 using ascorbic acid as reducing reagent. Micro glass cells of 50 mm path length (approximate volume: 3 mL), was used.

Results and discussion

Determined value of orthophosphate varied from 0.04 to 0.58 micro mol/L, while SRP showed values from 0.34 to 2.31 micro mol/L. There was so much difference between orthophosphate concentration and SRP in all river water samples collected. Ratios of orthophosphate to SRP in water differed between rivers sampled regardless of sampling season. In case of Yasu River, the ratio showed values from 0.06 to 0.14, while Ane River they were between 0.26 and 0.52. These differences might be caused by land use of watershed. In consideration of effects of river water quality to trophic status and primary production in Lake Biwa, these results might show the needs to consider direct impact of orthophosphate and indirect impact of other phosphorous compounds included in SRP separately.

Keywords: Lake Biwa, Inflow rivers, orthophosphate, SRP, Ion chromatography

MIS26-P14

Room:Convention Hall

Time:May 27 18:15-19:30

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Keywords: soil aggregate, macropore, soil moisture, infiltration

Effects of group cutting on litterfall and organic horizon in the secondary forest dominated by hinoki cypress

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Nitrogen cycling in forest ecosystem is influenced by cutting and vegetation recovery after cutting. The growth of under vegetation after cutting of a coniferous forest may contribute to nutrient input by litterfall and the decomposition process of organic horizon (O-horizon). These changes may have a significant impact on nitrogen cycling in the forest ecosystem.

Litterfall and O-horizon were investigated to determine the cutting effects on nitrogen cycling at three different slope positions in the secondary forest dominated by hinoki cypress 10 years after group cutting to promote regeneration of broad-leaved tree species.

Carbon inputs by litterfall in the cutting plots were lower than those in the control plots. Nitrogen inputs by litterfall in the cutting plots were similar those in the control plots. In the cutting plots at the middle and lower positions where abundant tall-trees of broad-leaved species regenerated, carbon and nitrogen inputs by broad-leaved tree species leaf-litter were higher and litterfall C/N ratio was lower than those in the control plots, whereas those at the upper position where abundant woody shrubs and red pine regenerated did not differ between the control and cutting plots. Litterfall C/N ratio decreased with increasing nitrogen inputs by broad-leaved species leaf-litter.

Stocks of carbon and nitrogen and the mean residence time (MRT) of carbon and nitrogen in the O-horizon in the cutting plots were lower than those in the control plots at the same position. The MRT of carbon and nitrogen in the O-horizon was shorter with decreasing litterfall C/N ratio. The decrease of MRT of O-horizon in the cutting plots at the middle and lower positions was much more notable, whereas at the upper position that was smaller than in the other two positions.

The results suggest that the change in MRT of O-horizon between the cutting and control plots at each position 10 years after group cutting is strongly influenced by regenerated tree species in the cutting plots, but not by the difference of soil nutrition along a slope.

Keywords: group cutting, litterfall C/N ratio, organic horizon, regenerated tree species, nitrogen input, slope position

Evaluating nutrient removal in a large river by in situ spiraling metric measurements

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Fluvial transports of excess nutrients can induce explosive growth of primary producers in aquatic ecosystems, thereby degrading the structure and function of downstream areas. River networks have traditionally been regarded as a conduit of such land-derived nutrient loads to coastal ecosystems. However, recent studies have increasingly identified the importance of in-stream processes that retain, transform and remove nitrogen and phosphorus from water column to benthic environments. Therefore, elucidating the pattern and mechanisms of nutrient removal processes in river networks has now become an important requirement to prevent the eutrophication of coastal waters.

Small headwater streams have been recognized as a vital element of riverine ecosystems as they are believed to be far more efficient at processing and transforming inorganic nutrients than large rivers. However, no study has hitherto directly quantified the nutrient removal rate in large rivers that exceed $18\text{m}^3/\text{s}$ in discharge. Therefore, the role of large rivers in controlling nutrient flux to downstream ecosystems has rarely been evaluated. In this study, we performed the in situ longitudinal measurements of dissolved inorganic nitrogen and phosphorus, as well as physico-chemical environmental gradients, to estimate the spiraling metrics (areal uptake rate, uptake velocity and uptake length) in the 6th-order mainstems of the Fuji River ($Q > 40\text{m}^3/\text{s}$), central Japan.

The present result showed that the areal uptake rates of NH_4 and PO_4 in the Fuji River are relatively fast compared with those estimated in the 1st-to-5th order rivers by previous studies, although the net uptake rates of NO_3 were usually negative due probably to the stoichiometric imbalance of river waters. In contrast, the metrics of nutrient removal efficiency of NH_4 and PO_4 (uptake velocity and uptake length) did not differ from or even low relative to the previous findings, as a result of the high nutrient concentration and high water velocity in this steep terrain watershed. In the presentation, we will also introduce the analyses on the effects of spatial heterogeneity in river environments on the nutrient spiraling metrics in order to identify the hotspots of nutrient removal in this large river.

Keywords: spiraling metric, large river, nutrient, uptake rate, hot spot