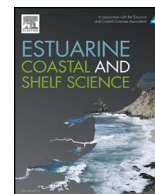




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Nitrogen fluxes between the ocean and a river basin using stable isotope analysis



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ABSTRACT

Total nitrogen (TN), consisting of total particulate nitrogen (TPN) and total dissolved nitrogen (TDN) pools, may be transported to the ocean not only via river channels but also from across the entire river basin via groundwater and migratory animals. Since marine derived nitrogen (MDN) is hypothesized to largely be transported in particulate form, it is necessary to investigate the contribution of particulate MDN in ground surface soils to the total MDN at the river ecosystem scale. In this study, we investigated TN export from an entire river basin to the ocean, and also estimated the contribution of pink (*Oncorhynchus gorbuscha*) and chum salmon (*O. keta*) to total oceanic nitrogen input across a river basin. The maximum potential contribution of MDN entering the river basin from the ocean as salmon was 23.8% relative to the total amount of TN exported from the river basin. The contribution of MDN from the ocean to particulate nitrogen in river basin soils was estimated to be 22.9% with standard deviation (SD) of 3.6% using stable isotope analysis (SIA) of nitrogen ($\delta^{15}\text{N}$).

1. Introduction

In river ecosystems, marine derived nitrogen (MDN) inputs have been shown to be important processes controlling the productivity of the ecosystem between the ocean and watersheds. For example, Merz and Moyle (2006) found that the contribution of MDN to the foliar nitrogen of wine grapes was about 18–25% from the ocean. Also, Hilderbrand et al. (1999) demonstrated that trees and shrubs near spawning streams received 24–26% of their foliar nitrogen from MDN, while Helfield and Naiman (2002) suggested that 15.5–17.8% of spruce foliage nitrogen may be provided by MDN. About the transport of MDN, Terrestrial consumers like mammals, birds, fishes and insects have been shown to play a large role in terms of providing MDN from the ocean to watersheds (Donaldson, 1966; Ben-David et al., 1997a; Hilderbrand et al., 1999; Gende et al., 2002; Naiman et al., 2002; Wilkinson et al., 2005; Bartz and Naiman, 2005; Koshino et al., 2013). In the other studies in natural rivers in cold regions, as a particularly important transfer mechanism, migrating fish such as salmon have been shown to be necessary for sustainable nutrient-cycles (nutrient fluxes) due to their important role as nutrient transporters (Ben-David et al., 1998; Wipfli et al., 1998; Yanai and Kochi, 2005; Gende et al., 2007; Hocking and Reimchen, 2009; Hocking and Reynolds, 2011). Additionally, the

transportation of nitrogen by migrating fish results in enhancement of biofilms and planktonic productivity in river systems (Juday et al., 1932; Cederholm and Peterson, 1985; Bilby et al., 1996; Gresh et al., 2000; Chaloner et al., 2002; Moore and Schindler, 2004; Yanai and Kochi, 2005; Levi and Tank, 2013; Marcarelli et al., 2014). For example, Cederholm et al. (1989) demonstrated that mammals and birds consume migrating fish, which may result in the secondary dispersion of MDN from the ocean across the river basin associated with the movement of these consumers. Other studies have revealed that mammals incorporate MDN from salmon, which may subsequently lead to re-export to the ocean through river flows (Bilby et al., 1996; Ben-David et al., 1997a, 1997b; Hilderbrand et al., 1999; Szepanski et al., 1999; Reimchen, 2000; Holtgrieve et al., 2009). Therefore, it is needed to clarify the nutrient fluxes from a river basin to the ocean and from the ocean to a river basin.

When we consider nitrogen flux in a river flowing from its upstream end into the ocean, the flux depends on nitrogen supplied from the entire river basin (Dutta and Nakayama, 2010; Alam and Dutta, 2012; Riggsbee et al., 2008). In mountainous regions, the total nitrogen flux is comprised mainly of particulate nitrogen, which is derived from surface soils. For example, in the mountainous regions of the Shiretoko Peninsula (site of this study), total particulate nitrogen comprised

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75%–95% of total nitrogen during a flood (Aynur et al., 2012). It is thus necessary to consider surface soils including particulate organic matter in order to analyze nitrogen transport from the river basin to the ocean. However, as mentioned in previous studies, surface soils including particulate organic matter consist of not only land derived materials but also marine derived materials, which means that surface soils may include more marine derived materials close to the ocean.

As one of the methods for analyzing the contribution of MDN, stable isotope analysis (SIA) is increasingly being used to examine connectivity in coastal aquatic-terrestrial ecosystems, such as the input of MDN from the open ocean to coastal and river ecosystems (Wyatt et al., 2010a, 2010b, 2012; Havik et al., 2014; Adame et al., 2015). Isotopic methods as an intrinsic geospatial tracer may thus provide a means to quantify cross-ecosystem transfer of nutrients from the ocean to watersheds. Therefore, this study aims to estimate the contribution of MDN to the particulate organic matter of surface soils by considering salmon as an oceanic source (end-member) in stable isotope analysis. In this study, we present total nitrogen (TN) transport across an entire river basin to the ocean, the potential contribution of MDN to the river basin by salmon, and the contribution of MDN to surface soils in the river basin. Integrated stable isotope analysis of geological, hydrological and biological compartments of the ecosystem allowed us to estimate the nitrogen budget between a natural river basin and the ocean, suggesting it may be important to conserve ocean-river connectivity in such systems.

2. Methods

2.1. Geophysical setting

Our target area, coastal land region and the ocean around the Shiretoko Peninsula, was registered as a World Natural Heritage area in July of 2005 (Fig. 1). Shiretoko is located at the southernmost extent of drift ice and its ecological systems exhibit high biodiversity and high rates of nitrogen circulation, particularly due to runs of pink (*Oncorhynchus gorbuscha*) and chum (*O. keta*) salmon from the Sea of Okhotsk (Aynur et al., 2012). Potential runs of salmon along the coast of Hokkaido in the Sea of Okhotsk have been estimated at about 29, 900, 000 individuals a year (Hokkaido National Fisheries Research Institute, Fisheries Research Agency, 2009). The average weight of chum and pink salmon in this region are 3.3 kg and 2.0 kg, respectively (Makiguchi et al., 2007), which include nitrogen of about 0.100 kg and 0.0608 kg, respectively (Larkin and Slaney, 1997). Therefore, the potential runs of salmon equivalents to 2590 tons of total nitrogen. The size of the Okhotsk coastal region of Hokkaido is about 24,000 km², which corresponds to that mean total nitrogen input from the ocean of about 108 kg km⁻² yr⁻¹ if we assume that all salmon run up rivers and the total nitrogen is completely distributed into river basins. Shiretoko is located on the northeast coast of Hokkaido, Japan (approximately 43°57′ N to 44°21′ N and 144°58′ E to 145° 23′ E), and has a width, length and maximum altitude of about 15 km, 50 km and 1660 m, respectively (Fig. 1). Therefore, the Shiretoko Peninsula is suitable for investigating nitrogen fluxes between coastal land region and the ocean. The Rausu River basin was selected as the main study area because its watershed is the largest in the region and it is considered a representative watershed in the Shiretoko Peninsula. The watershed area, river length, and the mean river slope are 32.5 km², 7 km, 1/7 (m/m), respectively. Because of the steep slope, nitrogen flux during a flood due to suspended particulate organic matters is larger than due to dissolved nitrogen (Aynur et al., 2012). Field experiments were carried out over 5 years from 2008 to 2012. For comparison with the Rausu River basin, stable isotope analyses were also carried out in 2014 in the Rusa River basin, which provided us equivalent plant and soil environments based on its old growth and conserved forests similar to those of Rausu River basin. Here, the watershed area, river length, and the mean river slope are 9.2 km², 5.5 km and 1/7, respectively (Fig. 1).

2.2. Nitrogen from a river basin to the ocean

MDN supplied from the ocean to surface soils in a river basin generally includes feces of mammals, droppings of birds, and the remains of salmon preyed upon by mammals, birds and insects. These MDN are recycled within the terrestrial ecosystems and mainly stored as soil organic matter (SOM). Thus, to focus on the influence of SOM on total particulate nitrogen (TPN) export, soil particles with diameter of less than 500 μm after rinsing in 1N-HCL solution were used in this analysis. The 500 μm cut-off was necessary because soil samples tended to include relatively large, low SOM particles because of the steep slope. The analysis does not allow evaluation of TN (TPN + TDN, TDN: total dissolved nitrogen) export from the river basin to the ocean. However, TPN export from an entire river basin has been revealed to be larger than TDN in the Rausu River basin due to its steep slope (Aynur et al., 2012).

We made an attempt to estimate the contribution of MDN to SOM resulting from the accumulation of particulate organic matter by sampling surface soils across the Rausu River basin (Fig. 2). It should be directly related to the potential riverine transport of MDN back to the ocean as suspended sediments. TDN was measured based on TN and TPN from glass microfiber filters 0.7 μm at St.0 around the river mouth from 2007 to 2009 in the Rausu River basin (TDN = TN – TPN; Fig. 2). The nitrogen concentration of filtered and non-filtered water samples were analyzed by the cadmium reduction-colorimetric method. Annual TN, TPN and TDN exports to the ocean were evaluated using the river discharge at St.0 with TDN-discharge and TPN-discharge curves. The TDN-discharge and TPN-discharge curves were produced using ten different peak discharge floods and base flow discharges. As river discharge was not measured during the winter season from January to March, river discharge was estimated using a storage function method from 2008 to 2012 (Michael, 1978; Michael et al., 1979). The validity of the storage function method was confirmed through comparison with the observed river discharge from April to December.

Surface soil samples were taken at 12, 20 and 21 stations in 2008, 2009 and 2012, respectively. Three soil samples (15 cm height × 15 cm width × 5 cm depth) were collected at each sampling station in order to account for small-scale variability in SOM (Fig. 2 and Table 1). In 2008, fewer samples were taken as we did not have permission to sample surface soils in special protection zones. Stable isotope analysis was conducted for fine particles whose diameter is less than 500 μm. After sieving through a 500 μm sieve with distilled water, the soil was dried for 3 days at 60–70 °C. The dried soil was ground using an alumina mortar. Surface soil sampling stations in 2012 are shown in Fig. 2. Since previous studies have revealed that surface soil transport is related to the spatial distribution of surface soil type, land-use type and vegetation (Ishida et al., 2010), the location of each sampling station was selected by dividing the river basin into 21 domains (sub-basin areas) that vary in soil type and vegetation (Fig. 1). The spatial distribution of surface soil types was divided into 6 categories. Although the spatial pattern in vegetation is complicated, the vegetation can generally be categorized in terms of altitude. Since Shiretoko is protected as a natural World Heritage area, all areas studied are classified as forest and have high vegetation cover by bamboo grass.

2.3. Salmon runs

To evaluate the contribution of salmon to SOM, salmon runs were investigated in the Rausu River. Salmon were caught at the river mouth for artificial incubation and release, providing an estimate of the number of salmon caught by the apparatus (Hokkaido National Fisheries Research Institute, Fisheries Research Agency, 2009). The apparatus for catching salmon consisted of a lattice fence, which does not obstruct flood flows or completely block the runs of salmon. Therefore, it was necessary to quantify the capture rate of the apparatus in order to estimate the actual volume of salmon runs. Field

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