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ORIGINAL ARTICLE

Influence of water discharged from a reservoir on reclaimed land into Isahaya Bay (Kyushu, Japan) on the regeneration of NH_4^+ in the water column

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Abstract
In 1997, the inner part of Isahaya Bay on the west coast of Ariake Bay, western Japan, was separated from the sea by dikes, leading to the formation of a freshwater reservoir. Since the Isahaya Reclamation Project began in the 1990s, red tides and hypoxia have become more common in the bay. Eutrophic high-turbidity water from the reservoir is frequently discharged into the bay, so NH_4^+ regenerated from organic matter in the discharge may be a source of nutrients for the red tides. In this work, we mixed reservoir water and seawater from the bay in experiments aimed at quantifying the NH_4^+ regeneration caused by the direct decomposition of labile organic matter in the high-turbidity drainage water. By comparing the results of field monitoring and mixing experiments, we estimated that the NH_4^+ regeneration rate under the influence of the drainage water was 192–501 µmol m^{-2} h^{-1}, which was 31.5–46.8% higher than the natural NH_4^+ regeneration rate. This suggests that the additional NH_4^+ regeneration induced by the drainage water is non-negligible and that the organic matter in the drainage water may be an important source of NH_4^+ for red tides in the bay. This is the first study to provide data on the non-negligible amount of NH_4^+ regenerated upon the direct decomposition of labile organic matter in high-turbidity water drained from a reservoir on reclaimed land into the bay.

Keywords NH_4^+ regeneration · Organic matter decomposition · Reservoir of reclaimed land · Drainage · Isahaya Bay

1 Introduction

Land reclamation is a coastal development process that involves the construction of dikes across tidal flats to create new land. Reclamation replaces the seawater zone with a freshwater reservoir that is used as a water supply. However, such reservoirs often suffer from eutrophication and toxic cyanobacterial blooms (Takahashi et al. 2014), and the eutrophic reservoir water is discharged into the sea (Migita et al. 2006; Sin et al. 2013). This discharging of reservoir water is considered problematic, given its impact on organisms such as phytoplankton and benthic animals in the seawater (Sin et al. 2013; Ishimatsu et al. 2018). Introducing allochthonous organic matter into the seawater via drainage can affect biogeochemical processes such as nutrient and oxygen dynamics through the microbial decomposition of organic matter in the seawater. Information on these processes is required in order to effectively manage coastal environments. However, few studies have examined the effects of reservoir drainage on biogeochemical processes.

Isahaya Bay is located on the west coast of Ariake Bay in western Japan. In 1997, the inner part of Isahaya Bay was separated from the sea by dikes. The separated area consisted of reclaimed land and a freshwater reservoir (26 km^2). Since the Isahaya Reclamation Project began in the early 1990s, the bivalves in and around the bay have declined in number and red tides and hypoxia have become more common in the bay (Kim et al. 2018). Because the recent environmental changes have also caused some social problems, a number of studies have been conducted to clarify the impact of the drainage on the ecosystem of Isahaya Bay, particularly

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the dynamics of toxic cyanobacteria (Takahashi et al. 2014; Umehara et al. 2015), the effects of hypoxic water on the macrobenthic community (Ishimatsu et al. 2018), and the diffusion of particulate organic matter from the reservoir (Yoshino et al. 2018). However, no study has clarified the impact of introducing organic matter from drainage water into the seawater on biogeochemical processes, which may be linked to the recent environmental changes (i.e., the increased frequency of red tides and hypoxia) in and around Isahaya Bay and the Isahaya Reclamation Project.

The total nitrogen and phosphorus concentrations increased three- to fourfold in the reservoir after the construction of the dike (Sasaki 2005) due to the lengthened freshwater residence time and the loss of vast tidal flats (Sasaki et al. 2003). Eutrophic high-turbidity water in the reservoir is often discharged into the bay through north and south drainage gates in the dike during or just after precipitation in order to maintain the water level in the reservoir at between 1.5 and 1.8 m. It is speculated that this discharged water is one of the most important nutrient sources for red tides of *Chattonella* spp. (Raphidophyte) in Isahaya Bay (Tada et al. 2010), because there are no major rivers other than the Honmyo River behind the dike in this bay (Fig. 1). Our preliminary study indicated that it is NH_4^+ regeneration within the high-turbidity water, not the direct input of NH_4^+, that facilitates the aforementioned red tides (Komorita et al. 2015). Thus, we hypothesized that the reservoir discharge is one of the most important labile organic matter sources for red tides in Isahaya Bay. Although the discharged water can affect multiple NH_4^+ regeneration pathways such as the direct decomposition of labile organic matter in the high-turbidity water and the decomposition of resuspended organic matter from sediment near the discharge gates, only the total regenerated NH_4^+ affected by the discharge has been reported (Komorita et al. 2015); the main NH_4^+ regeneration pathway induced by the discharge remains unknown. To prevent red tides and preserve the bay ecosystem, it is important to examine the causal relationship between the direct decomposition of labile organic matter in the high-turbidity water and NH_4^+ regeneration.

In this study, we mixed reservoir water and seawater in experiments to quantify the NH_4^+ regeneration induced by the direct decomposition of labile organic matter in the high-turbidity drainage. We also estimated the in situ oxygen consumption and NH_4^+ regeneration rates near the south and north discharge gates in Isahaya Bay. Given the results of the field monitoring and mixing experiments, we discuss how NH_4^+ regeneration is affected by the direct decomposition of labile organic matter in the drainage from the reservoir into the bay.

2 Methods

2.1 Sampling

We conducted experiments to examine NH_4^+ regeneration in the water column in Isahaya Bay due to the direct decomposition of labile organic matter in the high-turbidity drainage. On 16 August and 2, 6, 9, and 15 September 2017, water samples were collected from both the reservoir and Isahaya Bay at the dike near the north discharge gate. We also established two sampling stations (B1-1 and B1-3) in Isahaya Bay (Fig. 1) and monitored the in situ NH_4^+ regeneration rate at these two stations on 16 August and 13 September 2017. These stations were located near the dike drainage gates (Fig. 1).

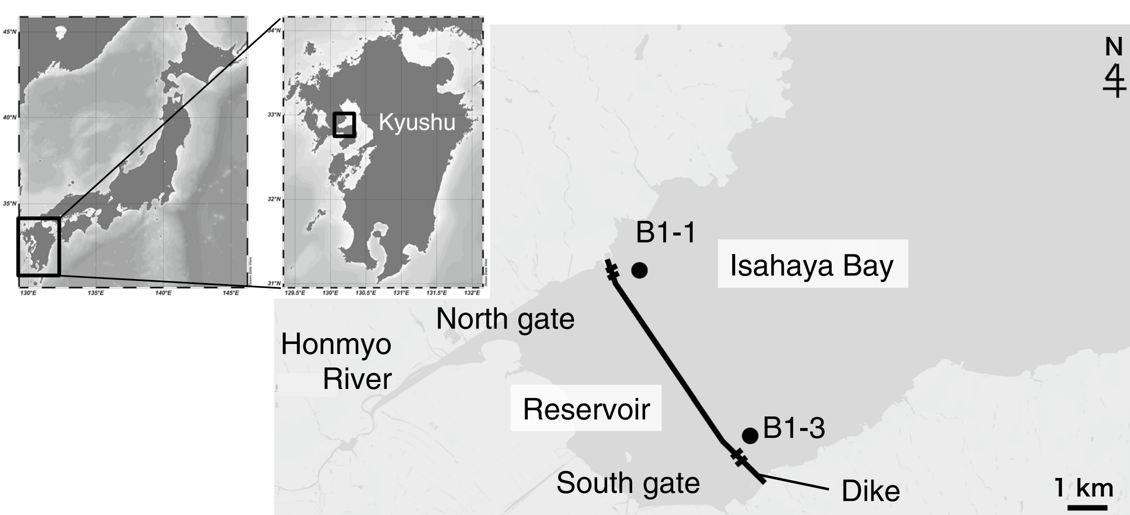

Fig. 1 The study area and sampling locations

2.2 Monitoring oxygen consumption and NH_4^+ regeneration rates in the field

A Van Dorn water sampler was used to collect seawater samples at predetermined depths (0.5-m or 1-m intervals). The seawater samples were prefiltered using a 330-μm pore mesh and stored in acid-washed 2-L polyethylene bottles. To estimate the NH_4^+ regeneration rates in situ, water samples in 250-mL amber polycarbonate bottles were incubated for 24 h in a water tank at the temperature of the bottom water.

2.3 Reservoir water addition experiment

Surface seawater and reservoir water were collected using a clean plastic bucket, transferred to 10-L polycarbonate tanks, and taken to the laboratory. Direct field observations of the reservoir drainage made by Komorita et al. (2014) suggest that the water in front of the gate is approximately 20% reservoir water. Therefore, the addition treatment consisted of adding reservoir water to seawater samples such that the reservoir water comprised 20% of the total volume. The control treatment consisted of seawater only. To estimate the oxygen consumption and NH_4^+ regeneration rates, the treatments were poured into 100-mL and 300-mL BOD bottles (3–5 bottles per treatment), respectively, and incubated for 5–12 h at the temperature in situ (25–29 °C) in the dark.

2.4 Determination of the oxygen consumption and NH_4^+ regeneration rates

Dissolved oxygen was measured using an optical oxygen probe (YSI ProOBOD, Xylem, USA) at the beginning (0 h) and end of the incubation. The oxygen consumption was calculated as the hourly change in oxygen in the sample over the course of incubation. Subsamples for measuring the NO_3^-, NO_2^-, and NH_4^+ concentrations were collected at the beginning (0 h) and end of incubation and filtered through 0.7-μm-pore glass fiber filters (GF/F, GE, UK). NO_3^-, NO_2^-, and NH_4^+ were determined calorimetrically using an autoanalyzer (AACS II, Bran+Luebbe, Germany) according to Armstrong et al. (1967). NO_3^-, NO_2^-, and NH_4^+ regeneration rates were calculated as the hourly change in concentration in the sample from the beginning to the end of incubation. The NH_4^+ regeneration rate in situ was evaluated as the depth-integrated value calculated according to the trapezoid rule, and was used for comparison with the results of the addition experiment.

2.5 Statistical analysis

The null hypothesis that dissolved nutrient mineralization rates were the same in the control water and the reservoir water mixed with the bay water was tested using a *t* test. Pearson's correlation between variables was computed. A *p* value of less than 0.05 was considered statistically significant. All statistical analyses were performed using the R software package, version 3.3.1 (R Core Team 2017).

3 Results and discussion

Figure 2 shows the temporal variation in the water discharge from the reservoir. By selecting samples taken at appropriate times, we ensured that measurements of the natural NH_4^+ regeneration rate in the bay would not be affected by the periodic reservoir drainage, as Komorita et al. (2014) suggested that the residence time of the drainage water near the dike was 6 h at most. During the field mornitoring, the NH_4^+ regeneration rate was higher near the north gate than near the south gate (Fig. 3, Table 1); it ranged between 0.06 and 0.11 μmol L^{-1} h^{-1} at St B1-1 and between 0.02 and 0.06 μmol L^{-1} h^{-1} at St. B1-3 (Fig. 3). The NH_4^+ regeneration rate was also higher in August than in September (Table 1), suggesting that NH_4^+ regeneration differed spatiotemporally within the bay. In Isahaya Bay, heterogeneous distributions have been observed for salinity (Saita et al. 2010), suspended solids (Komorita et al. 2014), and microcystins, which are by-products of toxic cyanobacteria (Umehara et al. 2015), without no obvious discharge. These heterogeneous distributions of freshwater and particulate matter should lead to heterogeneous distributions of labile organic matter and/or bacteria within the bay.

The NH_4^+ concentration in the reservoir water used in the addition experiments was lower than that in the seawater except on 2 September (Fig. 4). This supports the notion that the reservoir water discharge is not a direct source of NH_4^+ in Isahaya Bay (Komorita et al. 2015) and other sea dike systems (Sin et al. 2013). The low NH_4^+ concentration in the reservoir water may be due to (1) rapid consumption by

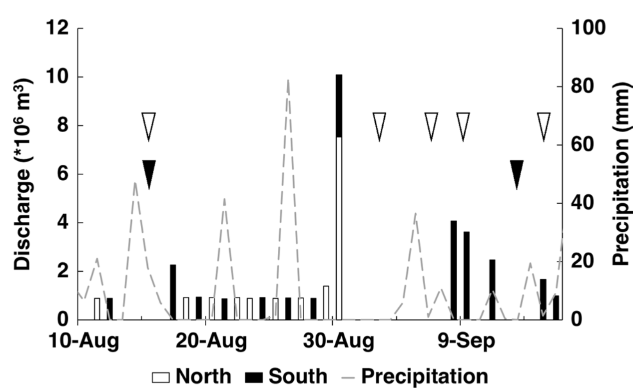

Fig. 2 Temporal fluctuations in the water discharge from the reservoir and the precipitation at Isahaya. *Open* and *solid triangles* indicate the dates of the addition experiments and routine monitoring, respectively

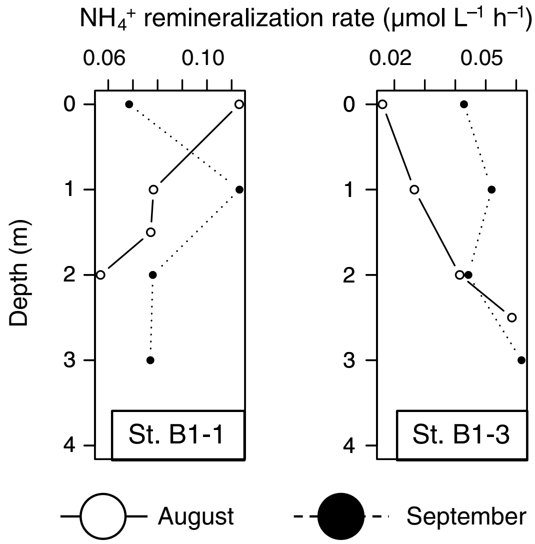

Fig. 3 Variation in the regeneration rate of NH_4–N as a function of depth in August (*open circles*) and September (*filled circles*)

freshwater phytoplankton and (2) nitrification in the water column and/or sediment surface in the reservoir. During the experiment, extremely high concentrations of chlorophyll *a* (108.7 ± 537.5, mean ± SD), indicative of the presence of phytoplankton, were found in the reservoir water. Alternatively, nitrification—which is inhibited by sunlight (Guerrero and Jones 1996)—may also have been active because the reservoir water presented extremely low transparencies, ranging from 0.1 to 0.5 m (Umehara et al. 2012). Further studies are required to examine the nitrogen cycling within the reservoir.

Statistically significant differences in $NO_3^- + NO_2^-$ concentration between the addition and non-addition treatments were observed in most of the experiments (Fig. 5). However, there was no consistent trend in $NO_3^- + NO_2^-$ release with the addition of reservoir water (Fig. 5). The addition of reservoir water enhanced the oxygen consumption (data not shown) and NH_4^+ regeneration rates, albeit with some exceptions (Fig. 5). There was a significant positive correlation between the oxygen consumption rate and the NH_4^+ regeneration rate in the addition experiment ($r = 0.88$, $p < 0.05$, Fig. 6). The NH_4^+ regeneration rate roughly corresponded to the theoretical NH_4^+ regeneration rate calculated from the oxygen consumption rate, assuming that the molar ratio of oxygen consumed to nitrogen released during organic matter decomposition is 17.3 (based on $O_2/N = 8.63$ from Redfield et al. 1963). These results strongly suggest

Table 1 Standing stock and flux of NH_4^+ in two field observations

Month	Station	Bottom depth (m)	Areal depth-integrated standing stock of NH_4–N (mmol m^{-2})	Areal depth-integrated flux (μmol m^{-2} h^{-1})	
				NH_4–N	NH_4–N addition*
Aug	B1-1	3.00	1.96	225	321
	B1-3	3.40	2.28	133	192
Sep	B1-1	3.00	0.93	341	501
	B1-3	3.40	2.30	216	285

*Indicates the simulated NH_4^+ regeneration rate in the presence of discharged water, based on the relationship shown in Fig. 7

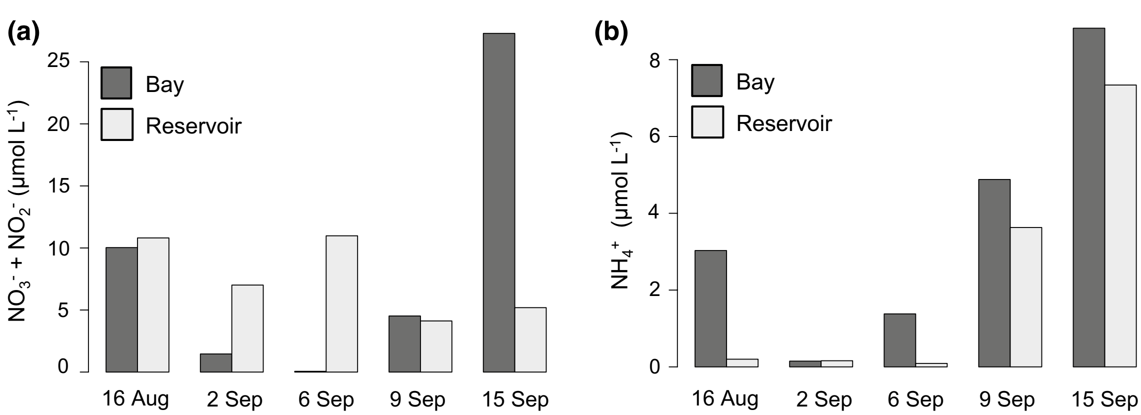

Fig. 4a–b Background concentrations of $NO_3 + NO_2$ (**a**) and NH_4^+ (**b**) in Isahaya Bay and the reservoir

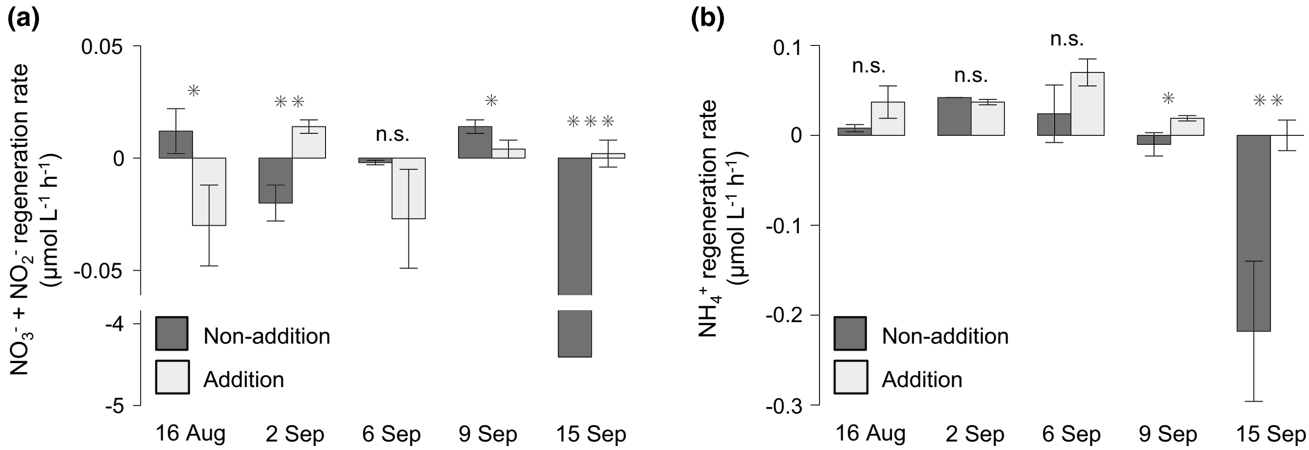

Fig. 5a–b $NO_3 + NO_2$ (**a**) and NH_4^+ (**b**) regeneration rates during the addition experiments. *n.s.* not significant; **p* < 0.05; ***p* < 0.01; ****p* < 0.001

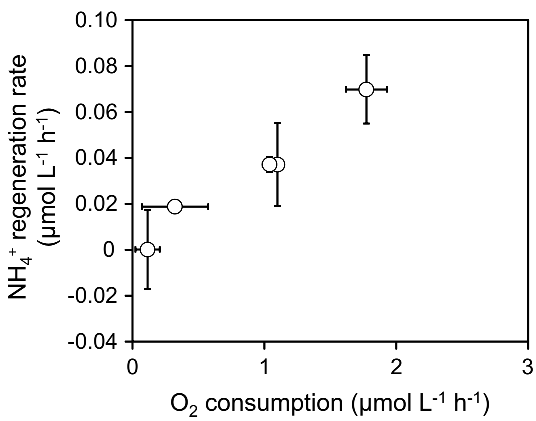

Fig. 6 Relationship between the O_2 consumption rate and the NH_4^+ regeneration rate in the addition experiments

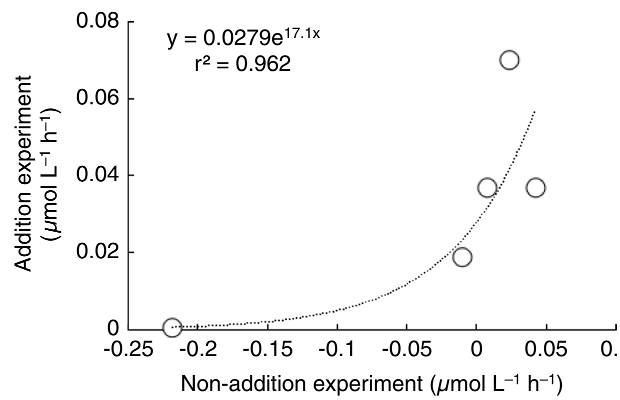

Fig. 7 Relationship of the NH_4^+ regeneration rate obseved in the non-addition experiment (control) to that observed in the addition experiment (in which reservoir water was added to seawater): $y = 0.0279e^{17.1x}$ ($r^2 = 0.962$)

that the decomposition of organic matter in the discharged reservoir water is a source of the regenerated NH_4^+ in the seawater in Isahaya Bay. In the laboratory experiment performed in this study, there was no significant correlation between chlorophyll *a* concentration and either the NH_4^+ regeneration rate or the oxygen consumption rate (data not shown), but organic matter in freshwater containing a high density of phytoplankton (108.7 ± 537.5 µg chlorophyll *a* L^{-1}, mean ± SD) should be affected strong osmotic pressure during the experiment. Phytoplankton cells that are heavily distorted by strong osmotic pressure can release labile organic matter into the seawater, which should increase the NH_4^+ regeneration rate, albeit only for a short period (only 5–12 h).

We found a significant positive correlation between the NH_4^+ regeneration rates in the addition and non-addition experiments (Fig. 7), so the NH_4^+ regeneration rate under the influence of drainage was calculated using the following formula:

$$NH_4^+ \text{ regeneration } (\mu\text{mol m}^{-2}\text{ h}^{-1}) = 0.0279e^{17.1a},$$

where *a* is the NH_4^+ regeneration rate in situ (µmol m^{-2} h^{-1}). The estimated NH_4^+ regeneration rate under the influence of the drainage water was 192–501 µmol m^{-2} h^{-1}, whereas the depth-integrated NH_4^+ regeneration in situ was 133–341 µmol m^{-2} h^{-1}, as calculated from the values in Fig. 3 (Table 1). Therefore, under the influence of the drainage water, the NH_4^+ regeneration rate increased by 31.5–46.8%. These results strongly suggest that the additional NH_4^+ regeneration induced by drainage is non-negligible and that the organic matter from drainage water is an important source of NH_4^+ for red tides in the bay. However, Komorita et al. (2015) estimated the regeneration rate of NH_4^+ under the influence of water discharged from the reservoir in Isahaya Bay to be 1.8 mmol m^{-2} h^{-1}, based on the results of field monitoring. Their estimated NH_4^+ regeneration rates were 3.6–9.4 times higher than those observed in our study. Therefore, the decomposition of resuspended organic matter from sediment near the discharge gate may be the most important drainage-induced NH_4^+ regeneration pathway.

This study is the first to provide data on the non-negligible amount of NH_4^+ regenerated following the direct decomposition of labile organic matter in the high-turbidity water drained from a reservoir on reclaimed land into a bay.

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