Nitrification, Denitrification and Nitrate Reduction Rates in the Sediment of Hiroshima Bay, Japan

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Nitrification, denitrification and nitrate reduction rates in the sediments at two stations in Hiroshima Bay were studied seasonally from August 1994 to May 1995. The acetylene (C2H2) inhibition technique was used to evaluate the rate of these processes. The seasonal average of nitrification and denitrification rates ranged from 0.00 to 299 µg atN m–2 h–1 and 0.00 to 69.0 µg atN m–2 h–1, respectively. These two processes showed seasonal variations which mainly depended on the temperature of the sediment and the concentration of dissolved oxygen in the overlying water. In the present study, 11% of NO3– produced by nitrification was estimated to be denitrified as an average for a whole year. The seasonal average of NO3– reduction rate ranged from 0.00 to 794 µg atN m–2 h–1. The denitrification rate accounted for only 3% of the total NO3– reduction rate.

Keywords:
- Denitrification
- nitrification
- nitrate reduction
- Hiroshima Bay.

1. Introduction

The significance of nitrification and denitrification in coastal marine sediments has received considerable attention in recent years, because of their geochemical and ecological importance. In the presence of dissolved oxygen, a portion of the ammonium (NH4+) regenerated from benthic decomposition of organic matter is oxidized to nitrate (NO3–) before it is released from the sediments. This may be used as a terminal acceptor by denitrifying bacteria producing gaseous forms of nitrogen (N2, N2O) which are essentially unavailable to most coastal phytoplankton (Jenkins and Kemp, 1984; Howarth et al., 1988).

Nitrification, the microbial transformation of NH4+ to NO2– and NO3– is an important process in the nitrogen cycle, particularly because it links nitrogen mineralization to potential nitrogen loss from the benthic system through denitrification (Seitzinger, 1990; Sloth et al., 1992). Other ecological implication of nitrification are detoxification of high NH4+ concentrations and consumption of dissolved oxygen which may lead to the formation of oxygen depleted bottom water (Hall, 1986; Sloth et al., 1992).

Denitrification mitigates coastal nitrogen loading from terrestrial sources (Seitzinger and Nixon, 1985) and may in some cases limit coastal primary production (Boydton et al., 1980). Thus, the coupled processes of nitrification-denitrification are quantitatively important in the N budgets of coastal sediments where nitrogen loss by denitrification takes place (Kemp et al., 1990; Seitzinger, 1993).

Several researchers have recently reported on the rate of nitrification, denitrification and NO3– reduction in aquatic sediment or the coupling of nitrification and denitrification, and the part played by denitrification on the total NO3– reduction in coastal and estuarine sediments. In addition, there has been some arguments regarding the regulatory factors controlling the nitrification and denitrification in aquatic sediments (Nishio, 1983; Henriksen and Kemp, 1988; Jørgensen and Sørensen, 1988; Kemp et al., 1990; Sloth et al., 1992). However, the detailed process of overall inorganic nitrogen transformation including seasonal variation in the marine sediments, has not yet been well investigated.

In this study, we concurrently measured the seasonal variation of nitrification, denitrification and NO3– reduction rates in the sediments of Hiroshima Bay, estimated the coupling of nitrification and denitrification, and identified the environmental factors which control these processes.

2. Materials and Methods

2.1 Study sites and sampling

The field investigation was carried out four times, August and October in 1994 and January and May in 1995, at two stations, HO-1 (34°19.5′N, 132°23.8′E) and HO-4 (34°08.1′N, 132°22.0′E) in Hiroshima Bay. The depth was 15 m at Stn. HO-1 and 31 m at Stn. HO-4. The bay is located in the Seto Inland Sea and receives considerable input of nutrients and organic substances from several rivers. Advective motion of water by the river discharge generally causes the transportation of suspended matter from Hiroshima Bay through the Seto Inland Sea towards the Pacific Ocean. However, the narrow mouth of the bay restricts the water exchange, and deposition of organic matter sometimes causes oxygen depletion of bottom water in part of the bay (Yuasa...
et al., 1985). As a result of these physical features, silt sediment prevails at almost all locations except the mouth of the bay where the sediment is sandy.

For the present investigation, we collected 28 intact sediment samples with a core sampler (gravity type KK, Hashimoto Kagaku Co., Ltd.) provided with an acrylic tube (5.2 cm diameter and 67 cm long) that allows visual inspection of the sediment. Cores with any signs of artificial disruption of surface or layered structure were discarded. First, the four sediment cores were used for the measurements of dissolved oxygen (DO) and dissolved inorganic nitrogen (DIN) in overlying water sampled with a siphon from 20 cm above the sediment surface. For the analysis of DIN, overlying water was filtered through a membrane filter of pore size 0.45 µm (Millipore HA). The remaining sediments were used for the measurements of temperature, oxidation reduction potential (ORP; Eh), ignition loss (IL), acid volatile sulfide (AVS-S) and NH₄⁺ and NO₂⁻ + NO₃⁻ in the sediment pore water. The other four cores were used for the sampling of nitrous oxide (N₂O) as the background concentration.

C₂H₂-saturated sea water was injected into the other 10 cores to inhibit the oxidation of NH₄⁺ and the reduction of N₂O to N₂. Six cores served as a control for nitrification in overlying water and sediment, and the remaining four cores served as a control for denitrification and nitrification in overlying water. Incubation was carried out in a water bath by circulating surface sea water.

2.2 Water and sediment chemistry

The temperature of surface sediment was measured immediately using a thermometer. Winkler titration was performed to determine dissolved oxygen in the overlying water (Strickland and Parsons, 1972). Sea water samples for the analyses of NH₄⁺ and NO₂⁻ + NO₃⁻ were filtered through a membrane filter of pore size 0.45 µm (Millipore HA) and stored frozen (−20°C) until the chemical analysis. NH₄⁺ was measured by the modified indophenol method (Sasaki and Sawada, 1980), NO₂⁻ + NO₃⁻ was measured by the cadmium reduction method (Bendschneider and Robinson, 1952; APHA-AWWA-WPCF, 1980).

Duplicate determinations of IL on dry samples after 3 h at 600°C were performed as an approximate indicator of organic content of the sediment. The concentrations of NH₄⁺ and NO₂⁻ + NO₃⁻ in the sediment pore waters were measured. The upper 5 cm (ca. 20 g) of the intact sediment was transferred into a 100 ml glass centrifuge tube. Pore waters was extracted by adding 20 ml of 2 N KCl solution and centrifuged for 10 min at 2000 rpm, followed by filtration and freezing until the analysis of NH₄⁺ and NO₂⁻ + NO₃⁻.

2.3 Denitrification

Recently, several studies on the measurement of in situ denitrification in coastal marine sediments have been performed and a variety of methods including the C₂H₂ inhibition technique (Sørensen, 1978; Kaspar, 1982), ¹⁵N isotope techniques (Nishio et al., 1982, 1983) and direct assays of N₂ production (Seitzinger et al., 1980; Devol, 1991) have been introduced. The most direct method is the measurement of N₂ production. However, this N₂ production approach may suffer from an obvious contamination risk during the long preincubation periods (1 to 2 weeks) in addition to the exhaustion of both dissolved oxygen and nitrate in the overlying water (Binnerup et al., 1992).

The C₂H₂ inhibition method has been extensively used out to measure both denitrification and nitrification in aquatic sediments. C₂H₂ concentration >10% (v/v) in pure culture was reported to inhibit not only NH₄⁺ oxidation but also N₂O reduction (Balderston et al., 1976; Berg et al., 1982; Jørgensen and Sørensen, 1988; Bedard and knowles, 1989; Sloth et al., 1992).

In the present study we employed the C₂H₂ inhibition method to estimate denitrification, because the C₂H₂ inhibition method has advantages of high reproducibility, even with a brief incubation time (Sloth et al., 1992), and high sensitivity (Binnerup et al., 1992). We essentially employed the experimental design and incubation procedure of Jensen et al. (1988) and Kemp et al. (1990). The water phase above the sediment was adjusted to a height of 52 cm by the addition of the bottom water. The sediment phase was adjusted to a length of 10 cm by discarding the lower part of the sediment. The tube was filled with the same bottom water and closed with a gas-tight rubber stopper. To obtain a final concentration of 10% C₂H₂ (v/v), 110 ml of the overlying water over the sediment core was replaced by the same volume of C₂H₂-saturated bottom sea water. C₂H₂-saturated seawater 200 µl was injected into the sediment from four different directions through a vertical series of silicon rubber-filled septa placed at 1 cm intervals along the side of the tube. Andersen et al. (1984) reported that stirring could stimulate denitrification by 50%, probably as a result of a facilitated uptake and transportation of NO₃⁻ to the denitrifying sites in the deeper layer of the sediment. In the present study, considering the in situ conditions, a small magnetic stirring bar was hung under the stopper in order to mix the overlying water without resuspension of sediment particles. The C₂H₂ treated cores and prepared control cores were then incubated on board in a water bath at surface sea water temperature for 3 h.

After the incubation, 120 ml of the water phase was sampled and 60 ml of the water subsample was transferred into a closed serum bottle (120 ml volume) which was pre-evacuated to obtain normal atmospheric pressure after the water sample was injected. The remaining 60 ml water sample was used for the later analysis of NH₄⁺ and NO₂⁻ + NO₃⁻. The serum bottle in which the water sample was contained was shaken vigorously for 2 min to equilibrate the dissolved N₂O with the gas phase. After that, 8 ml of gas sample was transferred to a pre-evacuated 8 ml glass vial.
After water samples were removed, the tube was capped and shaken vigorously for 2 min to equilibrate the remaining N₂O with the 120 ml gas phase in the sediment cores. Again, duplicate 8 ml gas samples were transferred to pre-evacuated 8 ml glass vials.

The gas sample was analysed for N₂O on a gas chromatograph (Shimazu, model GC-RIA) equipped with a thermal conductivity detector (at 140°C) and a 80/100 mesh Porapak Q column (2 m long and 3.2 mm diameter) operated at 55°C. Pure helium gas was used as a carrier at a flow rate of 50 ml min⁻¹, the detection limit of N₂O was 0.01 µM.

Total N₂O accumulation in the cores was the sum of N₂O in the water phase and sediment pore waters. The volumetric solubility coefficients of N₂O at various salinities and temperatures were used to correct for dissolved N₂O in the equilibrations (Weiss and Price, 1980). The N₂O recovery in the C₃H₂ inhibition method was checked by measuring the amount of N₂O gas at 3 h after the injection of standard N₂O to the sediment cores. The recovery rate of the standard N₂O gas was over 70%. Denitrification rate was calculated as the mean of N₂O accumulation rates measured in six different cores. After the extraction of N₂O from the water sample, the remaining water sample was filtered through a membrane filter of pore size 0.45 µm (Millipore HA) and stored frozen for later analysis of NH₄⁺ and NO₂⁻ + NO₃⁻ in the overlying water.

2.4 Nitrification

The method used in this study is based on the fact that NH₄⁺ oxidation is inhibited by C₃H₂ (Walter et al., 1979). The C₃H₂ inhibition technique is applicable to the measurement of nitrification rate with simultaneous measurement of denitrification rate. Both the sampling and incubation procedures have been described in Subsection 2.3. After the incubation, 60 ml water samples were collected from the overlying water and treated as described in Subsection 2.2 for the analysis of NH₄⁺; the detection limit of NH₄⁺ was 0.5 µM.

Sediment nitrification rate was calculated as the difference in buildup in overlying water column of the C₃H₂ treated core and the control core after a 3-h incubation. Four cores were used for the treatment and control, respectively. Although incubation time causes many problems, including a considerable decrease in dissolved oxygen both in the water column as well as sediment (Sloth et al., 1992). Some time lag would be expected between the accumulation of ammonium in the surface sediment and increase in ammonium in the overlying water within 3 h, and the calculated nitrification rate represents a rather conservative evaluation of total sedimentary nitrification. Binnerup et al. (1992) reported that the C₃H₂ inhibition method used in this study was useful for measuring nitrification and has no influence on the measuring of NO₃⁻ reduction in sediment.

2.5 Nitrate reduction to ammonium in the sediment

Since C₃H₂ also inhibits NH₄⁺ oxidation by nitrifying bacteria (Walter et al., 1979; Hynes and Knowles, 1982), consumption of NO₂⁻ + NO₃⁻ should represent the total NO₃⁻ reducing activity (Jørgensen and Sørensen, 1985, 1988; Sørensen, 1987). The total NO₃⁻ reduction rate of sediment was measured as the difference between NO₂⁻ + NO₃⁻ concentration in both overlying water and sediment pore water at 0 h and in C₃H₂-treated cores after 3 h incubation. The detection limit of NO₂⁻ + NO₃⁻ was 0.1 µM. The NO₃⁻ reduction rate of sediment was calculated by subtracting the measured denitrification rate from the total NO₃⁻ reduction rate.

Four cores were used for the determination of NO₂⁻ + NO₃⁻ concentration at 0 h and 3 h incubation, respectively. Details of the sampling and incubation procedures are given in Subsection 2.3. The analytical method of NO₂⁻ + NO₃⁻ was also described in Subsection 2.2.

3. Results and Discussion

3.1 Water and sediment qualities

Seasonal variations of DO in the overlying water and IL, AVS-S, ORP (Eh) and temperature of the sediments collected from two stations are shown in Fig. 1. The sediment temperature varied from 13.7 to 25.2°C. Dissolved oxygen in the overlying water ranged from 2.60 to 8.93 ml l⁻¹, without hypoxic (O₂ < 2 ml l⁻¹) or anoxic conditions in the bottom water during the study period. The range of IL in the sediment was 7.04–12.0%, with the highest value in October and the lowest in January. The range of AVS-S in the sediment was 0.12–0.65 mg (g dw)⁻¹, with high values in August and October, and the lowest in May. The range of Eh in the sediment was −185 to 302 mV, indicated that the most oxidized condition occurred in August and the most oxidized condition in January.

Concentrations of NH₄⁺ and NO₂⁻ + NO₃⁻ in the overlying water and the sediment pore waters of these two stations are shown in Fig. 2. The ranges of average NH₄⁺ concentrations in the overlying water and the sediment pore waters were 1.06–5.56 µg atN l⁻¹ and 209–508 µg atN l⁻¹, respectively, the highest being in August, due the elevated NH₄⁺ regeneration rate at high temperature, and the stratification of the water column. The ranges of average NO₂⁻ + NO₃⁻ concentration in the overlying water and the sediment pore waters were 0.60–5.29 µg atN l⁻¹ and 6.45–224 µg atN l⁻¹, respectively, the highest being in the overlying water and the lowest in the sediment pore waters in October. NH₄⁺ and NO₂⁻ + NO₃⁻ concentrations were always higher in the sediment pore waters than in the overlying water.

3.2 Nitrification rate

Generally, sediment nitrification rate is regulated by the availabilities of dissolved oxygen and NH₄⁺ (Henriksen...
and Kemp, 1988), and it also depends on the temperature. \( \text{NH}_4^+ \) regeneration rate also tends to increase with increasing temperature (Nixon, 1981).

The average nitrification rate in August, 1994 was 0.00 \( \mu \text{g \text{atN} m^{-2} h^{-1}} \) at both stations (Table 1). In October, it was 299 \( \mu \text{g \text{atN} m^{-2} h^{-1}} \) at Stn. HO-1 and 164 \( \mu \text{g \text{atN} m^{-2} h^{-1}} \) at Stn. HO-4, respectively. In January 1995, the rates were 0.00 and 283 \( \mu \text{g \text{atN} m^{-2} h^{-1}} \), respectively. In May they were 198 and 121 \( \mu \text{g \text{atN} m^{-2} h^{-1}} \), respectively.

The depression of nitrification rate in August may be explained by the decrease of dissolved oxygen concentration in the overlying water accompanied by the stratification of the water column, resulting in the being under reduced conditions sediment (Eh: –51 to –185 mV). Kemp et al. (1990) reported that nitrification rate was zero at redox levels of –200 mV and significant rates were observed in sediments of Chesapeake Bay when redox values were between –100 and 0.00 mV. By contrast, the high nitrification rate in October may be due to the acceleration by higher temperature (23 to 24°C) compared to those of the other seasons (ca. 14°C), since \( \text{NH}_4^+ \) concentration (227 to 340 \( \mu \text{g \text{atN} l^{-1}} \)) and dissolved oxygen (4.2 to 4.9 ml l^{-1}) concentrations are supposed to be sufficiently high for the nitrification process to occur (164 to 299 \( \mu \text{g \text{atN} m^{-2} h^{-1}} \)).

From the results described above, dissolved oxygen level and temperature are considered to play important roles on nitrification process in the study area, since the \( \text{NH}_4^+ \) concentration in the sediment pore water seems to be sufficient for the processes throughout the year. Henriksen and Kemp (1988) summarized the nitrification rate in various coastal areas, which varied from 29 to 75 \( \mu \text{g \text{atN} m^{-2} h^{-1}} \). Kemp et al. (1990) also reported that the nitrification rate of sediment in Chesapeake Bay ranged from 0.00 to 70.4 \( \mu \text{g \text{atN} m^{-2} h^{-1}} \) from Spring to Autumn.

The nitrification rates observed in this study were variable and the maximum rate was higher than previously reported values. This shows that the sediment of Hiroshima Bay has high nitrification potential, due to the high concentration of \( \text{NH}_4^+ \) in the sediment and \( \text{O}_2 \) in the overlying water, except for August in this study, which leads to a higher nitrification rate.

### Denitrification and nitrate reduction rates

The average denitrification rate at Stn. HO-1 and Stn. HO-4 in August was 68.9 and 0.00 \( \mu \text{g \text{atN} m^{-2} h^{-1}} \), respectively. In October it was 0.00 and 33.0 \( \mu \text{g \text{atN} m^{-2} h^{-1}} \), respectively. In January, denitrification rate was zero at both stations. In May, the rates were 0.00 and 9.73 \( \mu \text{g \text{atN} m^{-2} h^{-1}} \), respectively. In January, high dissolved oxygen in the overlying water (5.8 ml l^{-1}) due to vertical mixing of water column is supposed to have caused the highly oxidized condition of the sediment (Eh: 153 and 302 mV) and suppressed the denitrification processes. The highest denitrification rate observed in August, may be explained by high temperature as described for the highest nitrification rate in October.

Recently, several studies of the coupling of nitrification and denitrification have been published. A strong coupling of denitrification and nitrification has been demonstrated by Jenkins and Kemp (1984) for the sediment of Patuxent River estuary, Chesapeake Bay, who found that more than 99% of the produced \( \text{NO}_3^- \) was subsequently denitrified in the sediment during Spring and Summer. However, a weak coupling of two processes in estuarine sediments was also reported by Nishio et al. (1983). In Tama estuary, about 6 to 9% of \( \text{NO}_3^- \) produced by nitrification was denitrified in May and December, and in Odawa Bay, about 30% was denitrified in May. Lohse et al. (1993) also reported that in the southeastern North Sea, 3 and 25% of the \( \text{NO}_3^- \) produced was denitrified in August and February, respectively. In the present study, at Stn. HO-1 and HO-4, 0 and 89% of \( \text{NO}_3^- \) produced by nitrification was denitrified in May.
Table 1. Nitrification, denitrification and NO$_3^-$ reduction rates (µg atN m$^{-2}$h$^{-1}$) in the sediments of Hiroshima Bay.

<table>
<thead>
<tr>
<th>Month</th>
<th>Station</th>
<th>Nitrification ($n = 6$) NH$_4^+$ Rate (µg atN m$^{-2}$h$^{-1}$)</th>
<th>Denitrification rate (mean)</th>
<th>NO$_3^-$ reduction rate (mean ±SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aug. 94</td>
<td>HO-1</td>
<td>0.00</td>
<td>3.38 ± 1.06</td>
<td>3.90 ± 1.60</td>
</tr>
<tr>
<td></td>
<td>HO-4</td>
<td>0.00</td>
<td>4.59 ± 1.30</td>
<td>5.40 ± 1.93</td>
</tr>
<tr>
<td>Oct. 94</td>
<td>HO-1</td>
<td>299</td>
<td>5.96 ± 1.40</td>
<td>4.05 ± 1.11</td>
</tr>
<tr>
<td></td>
<td>HO-4</td>
<td>164</td>
<td>2.89 ± 0.91</td>
<td>1.85 ± 0.74</td>
</tr>
<tr>
<td>Jan. 95</td>
<td>HO-1</td>
<td>0.00</td>
<td>6.36 ± 0.85</td>
<td>7.50 ± 0.65</td>
</tr>
<tr>
<td></td>
<td>HO-4</td>
<td>283</td>
<td>2.97 ± 1.59</td>
<td>1.71 ± 0.65</td>
</tr>
<tr>
<td>May 95</td>
<td>HO-1</td>
<td>198</td>
<td>2.95 ± 0.89</td>
<td>1.68 ± 0.52</td>
</tr>
<tr>
<td></td>
<td>HO-4</td>
<td>121</td>
<td>5.83 ± 0.35</td>
<td>5.06 ± 1.37</td>
</tr>
</tbody>
</table>

*HO-1: Inner area of Hiroshima Bay, HO-4: Central area of Hiroshima Bay.

Fig. 2. Concentrations of ammonium and nitrite+nitrate (a) in the overlying water, and (b) in the sediment pore water. Open bars and shaded bars indicate the inner and the central area of Hiroshima Bay, Japan.
there was no denitrification found at both stations. In October, 0 and 20% of NO$_3^-$ produced by nitrification was denitriﬁed at each station. The level of coupling of nitrification and denitriﬁcation in Hiroshima Bay sediment varied over a wide range depending on the seasonal and regional conditions. The annual mean (11%), however, was very low compared with the other areas mentioned above. Since the condition of Hiroshima Bay sediment is usually oxidized in many cases except for Summer (Fig. 1), it has a high potential for nitrification. But denitriﬁcation is generally considered to be an anaerobic process and requires organic carbon as substrate (Seitzinger, 1993), in addition to nitrite or nitrate. For this reason, it was supposed that the aerobic–anaerobic interface where the coupling of nitrification and denitriﬁcation occurred did not extend into deeper sediments.

The average NO$_3^-$ reduction rates at Stn. HO-1 and Stn. HO-4 in August were 776 µg atN m$^{-2}$h$^{-1}$ and not detected, respectively (Table 1 and Fig. 3). In October, they were 974 and 216 µg atN m$^{-2}$h$^{-1}$, respectively. In January they were 518 and 0.00 µg atN m$^{-2}$h$^{-1}$, respectively. In May they were 17.1 and 215 µg atN m$^{-2}$h$^{-1}$, respectively. Seasonal variation was also found in the NO$_3^-$ reduction rate, which was high in August and October when the temperature was high, and low in May and January when temperature was low. The trend appeared to be highly related with the seasonal temperature variation and the sediment redox condition (Fig. 4). We conclude from the facts that the NO$_3^-$ reduction rate is mainly controlled by the temperature and sediment redox condition.

Denitriﬁcation rate and the correlation between total NO$_3^-$ reduction rate and denitriﬁcation rate in estuarine and coastal marine sediments have been reported over several decades. Kemp et al. (1990) reported that the denitriﬁcation rate of sediment in Chesapeake Bay in Spring and Summer ranged from 0.00 to 26.2 µg atN m$^{-2}$h$^{-1}$. Jørgensen and Sørensen (1988) reported that the denitriﬁcation rate in a small estuary Norsminde Fjord on the east coast of Jutland, Denmark were 83.3–417 µg atN m$^{-2}$h$^{-1}$ and NO$_3^-$ reduction rates were 417–1667 µg atN m$^{-2}$h$^{-1}$, and the denitriﬁcation accounted for a most variable fraction, from 15 to 70% of the total NO$_3^-$ reduction for a whole year. Nishio et al. (1982) also reported that denitriﬁcation rate in Summer accounted for about 27 to 57% of the nitrate consumption in the sediments of Odawa Bay and Tokyo Bay.

In this study, denitriﬁcation accounted for 0 and 4.3% of the total NO$_3^-$ reduction rate at Stn. HO-1 and Stn. HO-4 in May, respectively. In August it accounted for 47% and not detected, respectively. In October, denitriﬁcation accounted for 0 and 13% of the total NO$_3^-$ reduction rate, respectively. In January, denitriﬁcation accounted for 0% of
the total NO$_3^-$ reduction rates at both stations. The range of NO$_3^-$ + NO$_2^-$ concentration in the upper 5 cm sediment of Hiroshima Bay was seasonally variable, with observed values from 6.53 to 224 µM (Fig. 2). The seasonal cycle of total NO$_3^-$ reduction rate was higher in both Summer and Autumn than Winter and Spring, as found for denitrification. This indicates that the two processes might be coupled. However, the contribution of denitrification to the total NO$_3^-$ reduction in Hiroshima Bay sediment was small over a whole year. It can be inferred that the other reducing processes, such as NO$_3^-$ ammonification, occurred more than denitrification.

4. Conclusions

We have summarized the nitrification, denitrification and NO$_3^-$ reduction rate in the sediments of two stations in Hiroshima Bay over a whole year (Fig. 5). The main conclusions drawn from this study are as follows:

1. The coupling of denitrification and nitrification was very variable, from 0 to 89%, which depended on regional and seasonal conditions. As an average, 11% of NO$_3^-$ produced by nitrification was denitrified.
2. The proportion of denitrification rate to the total NO$_3^-$ reduction rate ranged from 0 to 47% while the annual average proportion was only 3%.
3. From (1) and (2), the major pathway of benthic inorganic nitrogen transformation appears to be between nitrification and dissimilative NO$_3^-$ reduction to NH$_4^+$.
4. The main factors regulating these processes turn out to be temperature and redox condition of sediments and dissolved oxygen in the overlying water.

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