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出版情報：International Journal of Marine Science. 4 (1), pp.1-15, 2014-01-04. Sophia Publishing Group, Inc.
バージョン：

International Journal of Marine Science 2014, Vol.4, No.1, 1-15
http://ijms.sophiapublisher.com

Research Article Open Access

The Nutrient and Carbon Dynamics that Mutually Benefit Coral and Seagrass in Mixed Habitats under the Influence of Groundwater at Bise Coral Reef, Okinawa, Japan

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International Journal of Marine Science, 2014, Vol.4, No.1 doi: 10.5376/ijms.2014.04.0001
Received: 27 Oct., 2013
Accepted: 29 Nov., 2013
Published: 04 Jan., 2014

Preferred citation for this article:
Higuchi et al, 2014, The Nutrient and Carbon Dynamics that Mutually Benefit Coral and Seagrass in Mixed Habitats under the Influence of Groundwater at Bise Coral Reef, Okinawa, Japan, International Journal of Marine Science, Vol.4, No.1 1−15 (doi: 10.5376/ijms.2014.04.0001)

Abstract The coral species, *Montipora digitata* and seagrass, *Thalassia hemprichii*, co-inhabit the southern portion of the reef moat in Bise, Okinawa, Japan. To elucidate the biogeochemical relationship between coral and seagrass in mixed communities of the coral reef ecosystem, the carbon metabolisms and the inorganic nitrogen flux rates were estimated in various reef habitats. We used benthic chambers to investigate sandy, seagrass, coral-seagrass mixed communities, coral, and acorn worm habitats. Relatively high concentrations of nitrate and nitrite ions (NO_x) were observed in all habitats due to coastal groundwater inflow. The uptake rate constant of NO_x was the highest in the coral-seagrass habitat and was significantly different from the rate constant in the seagrass habitat, indicating that seagrass benefits from co-inhabitation with coral. Dissolution of $CaCO_3$ was observed in the seagrass and coral-seagrass communities. This decline in basal coral carbonate substrate may contribute to increased fragmentation and dispersal of the coral habitat. On a biogeochemical scale, the coral-seagrass relationship benefits the seagrass in terms of NO_x availability and benefits the coral in terms of carbonate dissolution, increasing fragmentation, and furthering habitat development.

Keywords Carbon metabolism; Inorganic nitrogen flux; Coral-seagrass mixed community; Coral reef; Benthic chamber

1 Introduction

The biogeochemical dynamics between coral and seagrass ecosystems are important components of ecosystem management. Understanding these dynamics is especially important in coastal areas where increasing populations threaten the economic services these ecosystems provide. In order to understand broad-scale ecosystem function, recent management concerns have addressed the importance of functional group dynamics (Bellwood et al., 2004). Naeem (1998) stated that in order for ecosystems to resist “failure” after a disturbance event, species richness within functional groups is critical in maintaining ecosystem stability and reliability. For example, coral reefs serve as physical buffers for oceanic currents and waves, creating, over geologic time, a suitable environment for seagrass beds. In addition to these physical interactions, there are several biological and biogeochemical interactions between these interconnected ecosystems (Moberg and Folke, 1999). Seagrass beds interrupt freshwater discharge, are sinks for organic and inorganic materials as well as pollutants, and can generate an environment with the appropriate nutrient levels that promotes the growth of coral reefs (Miyajima et al., 2001; Umezawa et al., 2002).

If seagrass and coral in mixed habitats establish mutually symbiotic functional groups, it is possible that the presence of both can create an ecosystem that is more resilient to disturbance. However until recently, research has generally focused on the nutrient and carbon dynamics within these two ecosystems separately (Badgely et al. 2006, Grover et al., 2003; Marbà et al., 2006; Ohde and van Woesik, 1999; Tenore, 1988), and while few, if any, have addressed

cases in which coral and seagrass co-inhabit the same area.

Ninomiya et al. (2006) suggested that there is a physically mutualistic relationship between coral and seagrass in mixed habitats. The study asserts that the vertical and horizontal entwining of seagrass stems and coral branches both on and under the surface of the seafloor gives stability to seagrass beds. Jompa and McCook (2003) also showed that in the case of canopy forming macrophytes, understory corals are often protected from bleaching damage by shading. This implies that a dense seagrass community formed around fragmented coral can encourage coral survival by providing protection from bleaching.

Manzellos et al. (2012) suggested that coral calcification rates are higher near seagrass beds, because seagrasses draw down CO_2 in primary production and can store the carbon as biomass in their root systems. Thus, there is likely a biogeochemical relationship between seagrass and coral. Through the analysis of carbon and inorganic nitrogen dynamics, this study aims to elucidate this biogeochemical relationship. In Bise, Okinawa, Japan, there is a high population density of co-inhabiting seagrass and corals. Here, the southern portion of this reef moat is co-inhabited by the coral species, *Montipora digitata* and the seagrass species, *Thalassia hemprichii*. By comparing the biogeochemical interactions in the sand, seagrass, coral, and acorn worm habitats, with those in the coral-seagrass mixed habitat, we could identify the roles that each component has in the carbon and inorganic nitrogen dynamics both separately and interactively within the coral reef ecosystem.

2 Materials and Methods

2.1 Study site

This study was conducted in the subtropical reef moat off of Bise Coast, Okinawa, Japan, from Aug. 1 to 6, 2008 (Figure 1). It is characterized by coral, seagrass, and coral-seagrass mixed communities. The dominant coral species is *Montipora digitata,* and the dominant seagrass species is *Thalassia hemprichii*. The most conspicuous benthic organisms inhabiting this area are the acorn worms; *Schizocardium* sp. and *Ptychodera flava*. These invertebrates are bioturbators which produce fecal casts on the sediment surface while burrowing into the coral reef sands (up to 24 indivduals m^{-2}, pers. obsv.). The underlying substrate of these habitats is primarily dominated by limestone.

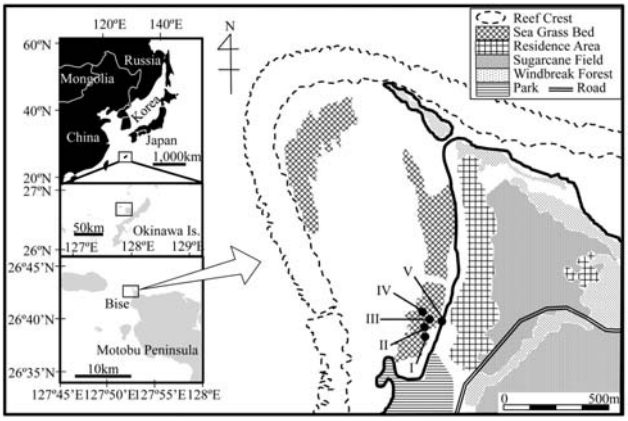

Figure 1 Map of the study site which shows the sand (I), seagrass (II), coral-seagrass (III), coral (IV) and sand-acorn worm (V) habitat

2.2 Chamber experiment

Benthic chambers (Ishikawa et al., 2007) were used to conduct 2 h incubations in each habitat (Figure 2). Using gardening stands fitted with plastic bags as incubation chambers, the nutrient and carbon dynamics of five benthic habitats were assessed: sand only (SD, 100% sand), seagrass only (SG, 100% *T. hemprichii*), coral-seagrass (CS, *T. hemprichii* : 50%, *M. digitata* : 45%, sand : 5%), coral only (CR, *M. digitata* : 95%, sand : 5%), and acorn worm habitats (AC, 100% sand with 2-4 acorn worms). Three incubation chambers were placed in each habitat and water samples were collected from each chamber. For the SG, CS, and CR habitats, 44 L benthic chambers (1479 cm^2 base area, 40 cm ht.) were deployed. Smaller chambers were used for the SD and AC habitats, 15 L benthic chambers (984 cm^2 base areas,

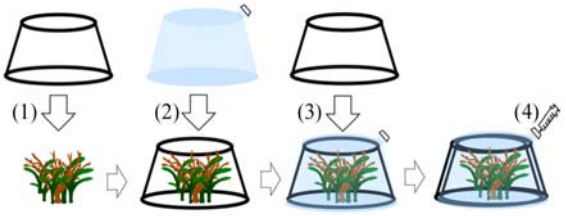

Figure 2 Schematic diagram of the benthic chamber. First, hard flames are put on each habitat (1). Then, clear plastic bags were covered on the flames (2). And then, hard flames was put again on the plastic bags (3). Water samples are taken by using syringes before and after the 2-h chamber incubations (4)

20 cm ht.) to prevent the chamber from emerging out of the water during low tide. Before beginning the incubations, each habitat was cleared of benthic macroalgae, snails and other visible macrofauna to minimize other variables that could affect the inorganic nitrogen and carbon dynamics. In the SD habitat, the contributions of phytoplankton and/or other microorganisms in benthos and water column were estimated as a control.

2.3 Chemical and environmental parameters

Seawater samples were collected by syringe at the beginning and end of each of the incubations. The pH and total alkalinity (TA) were determined using a pH electrode (pH meter Orion 4-star, Thermo-Fisher Scientific) on the NBS scale and the Gran plot method using a total alkalinity titrator (ATT-05, Kimoto), respectively. Accuracy of these measurement were ±0.05 for pH and ±10 μmol kg^{-1} for TA. Salinity (S) was measured by a salinometer (Portsal 8410A, Guildline) based on comparison with IAPSO standard water (S = 34.993). Ammonium, nitrate, and nitrite ions were measured by an automatic water analyzer (QuAAtro, BRAN+LUEBBE).

Data loggers (Water Temp Pro v2 Data Logger, HOBO) and photon sensors (MDS-MkV/L, Alec Electronics Co.) were deployed beside the chamber to record temperature and photon flux. Tidal level data were provided by the Okinawa Meteorological Observatory.

2.4 Groundwater inflow

Groundwater comes out of sediment through the limestone. To examine the effects of groundwater outflow into the reef moat, the initial total alkalinity (TA), nitrate + nitrite (NO_x), and ammonium (NH_4^+) concentrations during the lowest spring tides (1-Aug, 12:00, 1-Aug, 16:00, and 4-Aug, 16:00) of the sampling period were plotted against salinity (S). The slope of each regression line (R_{slope}) was used to calculate the change in concentration (ΔC_{inflow}) due to groundwater inflow during the chamber incubation.

$$\Delta C_{inflow} = R_{slope} \bullet \Delta S_{final\text{-}initial} \quad \text{(Eq.1)}$$

where ΔC_{inflow} is the inflow concentration of total alkalinity, NO_x or NH_4^+, R_{slope} is the slope of each regression line in Figure 3, $\Delta S_{final\text{-}initial}$ is the difference in salinity between final and initial samples in each incubation chamber.

2.5 Data analysis

The difference between chemical constituents in each chamber over a given period was attributed to biological activity. However, as we detected a decrease in salinity during the incubations, most of the chambers in this study were influenced by groundwater inflow. Therefore, we accounted for the effect of inflowing water to estimate the biological activity as follows:

$$\Delta C_{biological} = C_{final} - C_{initial} - \Delta C_{inflow} \quad \text{(Eq.2)}$$

Where, $\Delta C_{biological}$ is the change in TA, DIC, NO_x, or NH_4^+ during an incubation due to the biological activity within a given chamber.

Using the $\Delta C_{biological}$ of total alkalinity (TA) and total dissolved inorganic carbon (DIC) data, the rates of organic carbon production (OP, mmol m^{-2} h^{-1}) and inorganic carbon production (IP, mmol m^{-2} h^{-1}) were calculated using the alkalinity anomaly technique (Smith 1973, Smith and Kinsey 1978). IP was expressed by using: the changes in TA, volume of the chamber (V), base area of the chamber (A) and incubation time (t), as follows:

$$IP = \frac{-\Delta TA_{biological} \cdot V}{2 \cdot A \cdot t} \quad \text{(Eq. 3)}$$

OP was calculated by using the changes in total dissolved inorganic carbon (DIC), volume of chamber (V), the base area of chamber (A), incubation time (t) and IP, as follows:

$$OP = \frac{-\Delta DIC_{biological} \cdot V}{A \cdot t} - IP \quad \text{(Eq. 4)}$$

Total dissolved inorganic carbon (DIC) was also calculated using pH, total alkalinity (TA), and equilibrium constants for the carbonate system, as follows:

$$DIC = \frac{a_{H^+}{}^2 + K'_1 a_{H^+} + K'_1 K'_2}{K'_1 (a_{H^+} + 2K'_2)} \cdot \left(TA - \frac{K'_B \cdot 1.212 \times 10^{-5} \cdot S}{a_{H^+} + K'_B} \right) \quad \text{(Eq. 5)}$$

where DIC is the total dissolved inorganic carbon (μmol kg^{-1}), TA is the total alkalinity (μmol kg^{-1}), a_H^+ is the activity of hydrogen ions (i.e. = 10^{-pH}), K'_1 is the apparent first dissociation constant of carbonate ions

(9.91×10^{-7} at 25°C), K'_2 is the apparent second dissociation constant of carbonate ions (7.72×10^{-10} at 25°C), K'_B is the apparent dissociation constant of boric acid (2.02×10^{-9} at 25°C), and S is the salinity.

Photosynthetic and respiratory carbon metabolisms were estimated using the average value of OP (mmol m^{-2} h^{-1}) during the light (n=24) and dark (n=9) conditions in each habitat. The photosynthesis of gross production (P_{gross}) rate and the 24-h respiration (R_{24h}) rate were estimated as:

$$P_{gross} = (OP_{light}^{avg.} - OP_{dark}^{avg.}) \times T_{light} \quad \text{(Eq. 6)}$$

$$R_{24h} = 24 \times (-OP_{dark}^{avg.}) \quad \text{(Eq. 7)}$$

where T_{light} is the time of the light period (13 hours in this study) and the units of P_{gross} and R_{24h} are mmol m^{-2} d^{-1}. The duration ratio of daylight and night was 13-h (6:00-19:00) and 11-h (19:00-6:00) respectively, during the chamber experiment.

Note that in the present study, we assume that the daytime respiration is equal to the night time respiration as reported in previous studies (e.g. Tribollet et al. 2006, Bensoussan and Gattuso 2007). This neglects the possibility that light-respiration may be higher than dark-respiration, as shown in a large scale coral reef mesocosm (Langdon et al. 2003).

The calcification rates were calculated for each light and dark period (G_{light} and G_{dark}), respectively. G_{light} and G_{dark} were estimated by the average value of IP (mmol m^{-2} h^{-1}) in the light (n=24) and dark (n=9) conditions in each habitat.

$$G_{light} = IP_{light}^{avg.} \times T_{light} \quad \text{(Eq. 8)}$$

$$G_{dark} = IP_{dark}^{avg.} \times T_{dark} \quad \text{(Eq. 9)}$$

Net calcification (G_{net}) can be expressed as the sum of G_{light} and G_{dark}

$$G_{net} = G_{light} + G_{dark} \quad \text{(Eq. 10)}$$

where T_{dark} is the time of night period (11 hours) and the units of G_{light}, G_{dark} and G_{net} are mmol m^{-2} d^{-1}. A negative calcification value indicates $CaCO_3$ dissolution.

The rate of inorganic nitrogen flux was also calculated using the $\Delta C_{biological}$ during the chamber incubation periods.

$$\textit{Inorganic nitrogen flux (uptake /release)} = \frac{-\Delta C_{biological} \cdot V}{A \cdot t} \quad \text{(Eq.11)}$$

where $\Delta C_{biological}$ is the change in either NO_x or NH_4^+ by biological activity of the benthic organisms within a given chamber. A positive flux value indicated nutrient uptake whereas a negative value indicated release.

To calculate uptake/release rate constants of inorganic nitrogen, the total abundance of either NO_X or NH_4^+ which potentially affects a habitat was estimated as:

$$C_{total} = C_{initial} + \Delta C_{inflow} \quad \text{(Eq.12)}$$

Using the slope of the linear regressions, we calculated the NO_x and NH_4^+ uptake/release rate constants, to compare the uptake efficiencies within each habitat.

To compare the differences in habitat nutrient metabolism in relation to the tidal cycle, we classified the experimental times into either low (<100 cm) or high (>100 cm) tide. Statistical analysis was conducted using one-way analysis of variance (ANOVA) and Student's t-tests (JMP 8, SAS). Analysis of covariance (ANCOVA) with a Bonferroni adjustment was conducted to compare the uptake rate constants of the inorganic nitrogen. To compare the differences in habitat organic and inorganic carbon production, Tukey–Kramer honestly significant difference (HSD) tests were conducted.

3 Results

3.1 Groundwater inflow

The negative relationship between total alkalinity, nitrate + nitrite (NO_x) and ammonium concentrations with salinity (R^2=0.905, 0.921 and 0.723, respectively) are characteristic of mixing with groundwater (Figure 3). The high values of total alkalinity in the groundwater are attributed to the $CaCO_3$ that is dissolved by groundwater passing through the limestone dominated underlying geology. Also, as Okinawa uses nitrate based fertilizers for agriculture (Kawahata et al., 2000), and there are sugarcane fields near the coastal area (Figure 1), the dissolution of these compounds in freshwater and hence, in the groundwater, causes high NO_x concentrations. However, despite the residential area next to the study area (Figure 1), higher ammonium concentrations which are generally caused by human activities were

not observed. As groundwater inflow is apparent, ΔC_{inflow} must be accounted for to derive the effect of benthic activity on carbon and inorganic nitrogen flux.

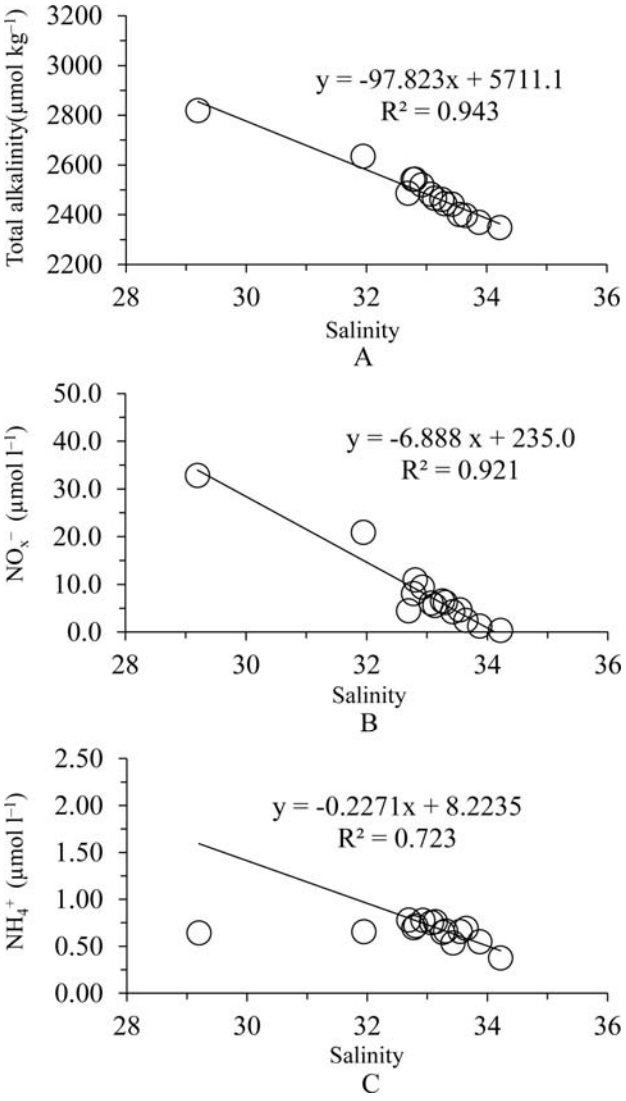

Figure 3 Correlations between total alkalinity (A), nitrite + nitrate (NO_x) (B), and ammonium (NH_4^+) (C) with salinity.

3.2 Organic carbon production (photosynthesis-respiration process)

Figure 4 shows organic carbon production rate vs photon flux fitted to the function of $y=a(1-e^{-x/b})+c$ which is a typical equation for photosynthesis-respiration vs irradiance curves (P-I curve). In the sand habitat (SD) and acorn worm habitat (AC), organic carbon production (OP) ranged from –4.49 to 5.53 and –5.18 to 4.47 mmol m^{-2} h^{-1} respectively indicating photosynthesis and respiration were low in comparison to the other habitats (Figure 4). The seagrass habitat (SG), coral-seagrass habitat (CS) and coral habitat (CR) showed higher organic production rates than the SD and AC habitats, and ranged from –12.11 to 27.26, –12.46 to 21.70 and –10.36 to 17.53 mmol m^{-2} h^{-1} along with the P-I curve, respectively. The results of the metabolic parameters of photosynthesis and respiration which shows gross production (P_{gross}), 24h respiration (R_{24h}), and P_{gross}/R_{24h} ratio are summarized in Table 2. P_{gross} and R_{24h} in SG were both significantly different from the other habitats (Tukey-Kramer HSD; $p<0.05$) except for the coral-seagrass habitat. P_{gross}/R_{24h} ratios were approximately 1 in all habitats except for the AC habitat where the P_{gross}/R_{24h} = 0.64 and was significantly lower than the CS, SG and CR habitats (Tukey-Kramer HSD; $p<0.05$). This indicates the AC habitat has a higher respiration rate relative to the photosynthesis.

3.3 Inorganic carbon production (calcification-dissolution process)

The inorganic carbon production (IP) rate was linearly correlated with photon flux in the CS and CR habitat while the other habitats were not (Figure 5). IP in the coral habitat was about 2-fold higher than that in the CS habitat, ranging from 0.23 to 14.42 and –0.39 to 7.14 mmol $CaCO_3$ m^{-2} h^{-1}, respectively. On the other hand IP rates in the SD, SG and AC habitat which did not include the coral component showed lower values; from –0.85 to 1.26, –2.14 to 1.17 and –1.68 to 0.89 mmol $CaCO_3$ m^{-2} h^{-1}, respectively. The negative values of IP indicate $CaCO_3$ dissolution. Most of the IP rates in the AC habitat indicated dissolution and were independent of photon flux. Dissolution was also notably high in the SG habitat at night. Metabolic parameters of calcification during the light and dark periods and net rate during the day are also summarized in Table 1. Most of the calcification in the dark (G_{dark}) showed negative values except for the CR habitat. The value in the SG habitat showed a significantly higher dissolution rate than that in the SD and CS habitats (Tukey-Kramer HSD; $p<0.05$). At night time, a slight dissolution of $CaCO_3$ was found in the CS habitat. Both calcification rate during the light (G_{light}) and net calcification rate (G_{net}) in the CR habitat were the highest followed by those in the CS habitat and were significantly different from the other habitats (Tukey-Kramer HSD; $p<0.05$). From these results, it appears that inorganic carbon production in Bise moat is primarily controlled by coral calcification.

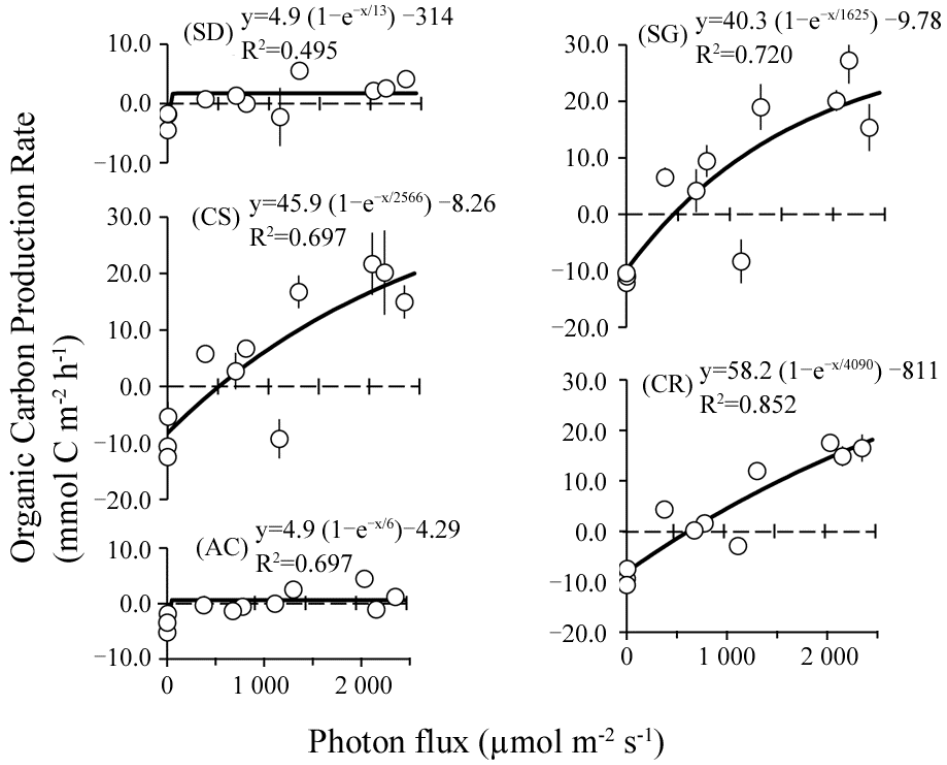

Figure 4 Comparisons of the organic carbon production rate with photon flux in the sand (SD), seagrass (SG), coral-seagrass (CS), coral (CR), and sand-acorn worm (AC) habitats. Organic carbon production values are ± SE (n=3, each). Exponential functions of $y = a\,(1 - e^{-x/b}) + c$ are fitted to the experimental data and shown with correlation of R^2

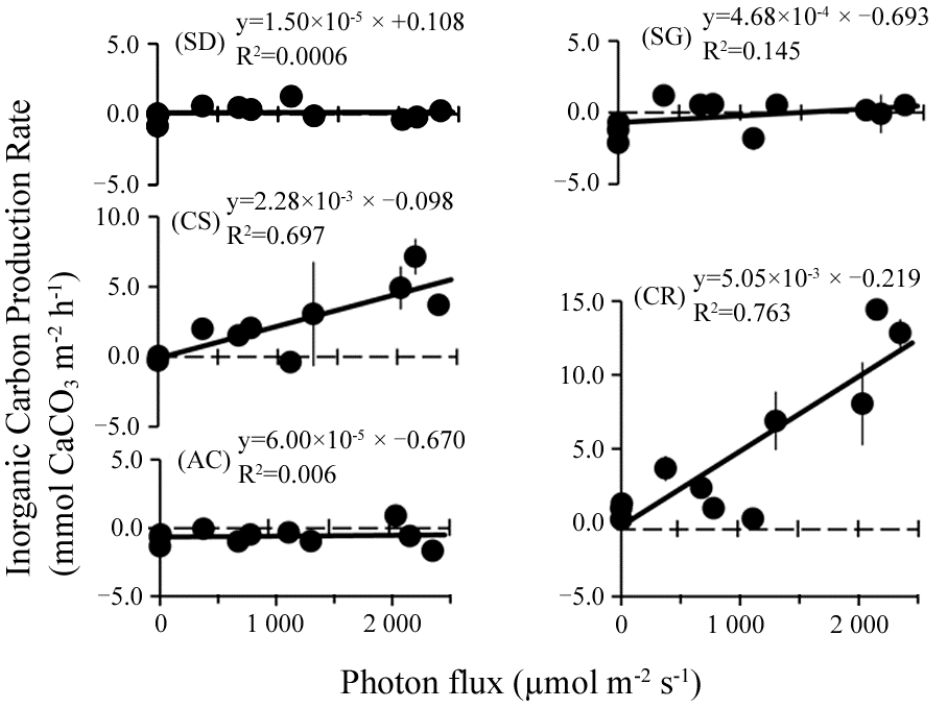

Figure 5 Comparisons of the inorganic carbon production rate with photon flux in the sand (SD), seagrass (SG), coral-seagrass (CS), coral (CR), and sand-acorn worm (AC) habitats. Inorganic carbon production values are ± SE (n=3, each). Regression lines are fitted to the experimental data and shown with correlation of R^2

Table 1 Summary of the carbon metabolisms of gross production, 24-hour respiration and calcification in the sand (SD), seagrass (SG), coral-seagrass (SC), coral (CR), and sand-acorn worm (AC) habitats. Different letters in each column indicate a significant difference between habitats (Tukey-Kramer HSD, P<0.05)

Habitat	Photosynthesis–Respiration (mmol m^{-2} d^{-1})			Calcification (mmol m^{-2} d^{-1})		
	P_{gross}	R_{24h}	P_{gross}/R_{24h}	G_{light}	G_{dark}	G_{net}
SD	57.9±0.89^{c}	64.1±11.1^{c}	0.95±0.14^{ab}	3.6±1.1^{c}	−3.1±1.0^{b}	0.5±1.7^{c}
SG	296.9±16.7^{a}	267.4±15.3^{a}	1.11±0.02^{a}	2.4±0.6^{c}	−15.0±3.0^{c}	−12.5±2.5^{c}
CS	252.5±14.4^{ab}	227.4±7.0^{ab}	1.11±0.07^{a}	38.9±5.6^{b}	−1.7±2.6^{b}	37.2±7.9^{b}
CR	223.1±3.5^{b}	219.7±5.3^{b}	1.02±0.03^{a}	80.4±5.1^{a}	9.2±1.5^{a}	89.6±6.6^{a}
AC	53.2±1.2^{c}	83.2±1.4^{c}	0.64±0.02^{b}	−6.9±1.0^{c}	−9.1±0.6^{bc}	−16.0±1.3^{c}

3.4 Nitrate and nitrite flux

Nitrate and nitrite (NO_x) uptake rate in each habitat was linearly correlated with NO_x concentration with a relatively strong relationship ranging from R^2=0.40 in the SG habitat to 0.83 in the AC habitat (Figure 6). The higher concentrations corresponded to low tide. The NO_x concentration in each habitat was significantly higher during low tide (n=18) than high tide (n=15) (Student's t-test; p<0.01). However there was no evidence of a strong relationship between the NO_x uptake rates and photon fluxes in each habitat, ranging from R^2=0.09 in the AC to 0.17 in the SG. These data clearly suggest that the NO_x uptake rate in each habitat is highly dependent on the NO_x concentration. The AC habitat showed the highest uptake rate of 906 μmol m^{-2} h^{-1} due to the high NO_x concentration of 32 μmol l^{-1}. Since the NO_x concentration was different among all habitats, we used the uptake rate constant to compare NO_x acquisition ability. Figure 8a shows the NO_x uptake rate constant derived from the slope of the regression line in Figure 6. The estimated rate constant in the CS habitat was the highest and significantly different from all other habitats (ANCOVA; p<0.05) except for the coral habitat.

3.5 Ammonium flux

Figure 7 shows the relationship between the NH_4^+ uptake rate and concentration with the regression line at each habitat. The correlations were relatively high in the SD (R^2=0.37), SG (R^2=0.47), and AC (R^2=0.81) habitats while they were lower in the CS and CR habitats. The NH_4^+ concentration in each habitat was significantly higher during low tide (n=18) than high tide (n=15) (Student's t-test; p<0.01) although the differences of the averaged concentration between low and high tide were very small, ranging from 0.17 in the SG to 0.32 μmol l^{-1} in the AC habitat. These results indicate that the ammonium concentrations in Bise moat are likely affected by tidally-driven groundwater inflow and the uptake rate is primarily controlled by the concentration. In the SD and AC habitats, there was no relationship between the concentration and photon flux (R^2=0.12 and 0.01, respectively). More than half of the uptake rate data in the CR habitat were negative, demonstrating the release of NH_4^+ from coral. This caused a low correlation between the uptake rate and concentration of NH_4^+ in both the CR and CS habitats. Figure 8b shows the uptake rate constants for NH_4^+in all the habitats. Although there were no significant differences between them, the rate constant tended to be the highest in the SG habitat. The lack of a significant difference by ANCOVA statistics is likely due to the low correlation in the CS and CR habitats shown in Figure 7.

4 Discussions

4.1 Carbon production

In the sand habitat, the organic carbon production (OP) was low (Figure 4). This suggests that the amount of phytoplankton in the benthos and water column in the Bise area is also low. Conversely, the OP in the coral and seagrass habitats was higher in comparison to the sand habitat. The OP was correlated with photon flux in the seagrass, coral-seagrass and coral habitats (R^2= 0.72, 0.70, and 0.85, respectively). Similar to previous studies which have reported a photosynthesis-respiration vs irradiance curve (P-I curve) (e.g. Gattuso et al., 1999), our study also supports this relationship. In addition, the carbon production of coral and seagrass in the present study were within the range of the metabolic data of macroalgal-dominated coral reef community as shown in Bensoussan and Gattuso (2007), although the photosynthetic gross production and respiration rates in the seagrass habitat were higher than in the other habitats in the Bise area (Table 1).

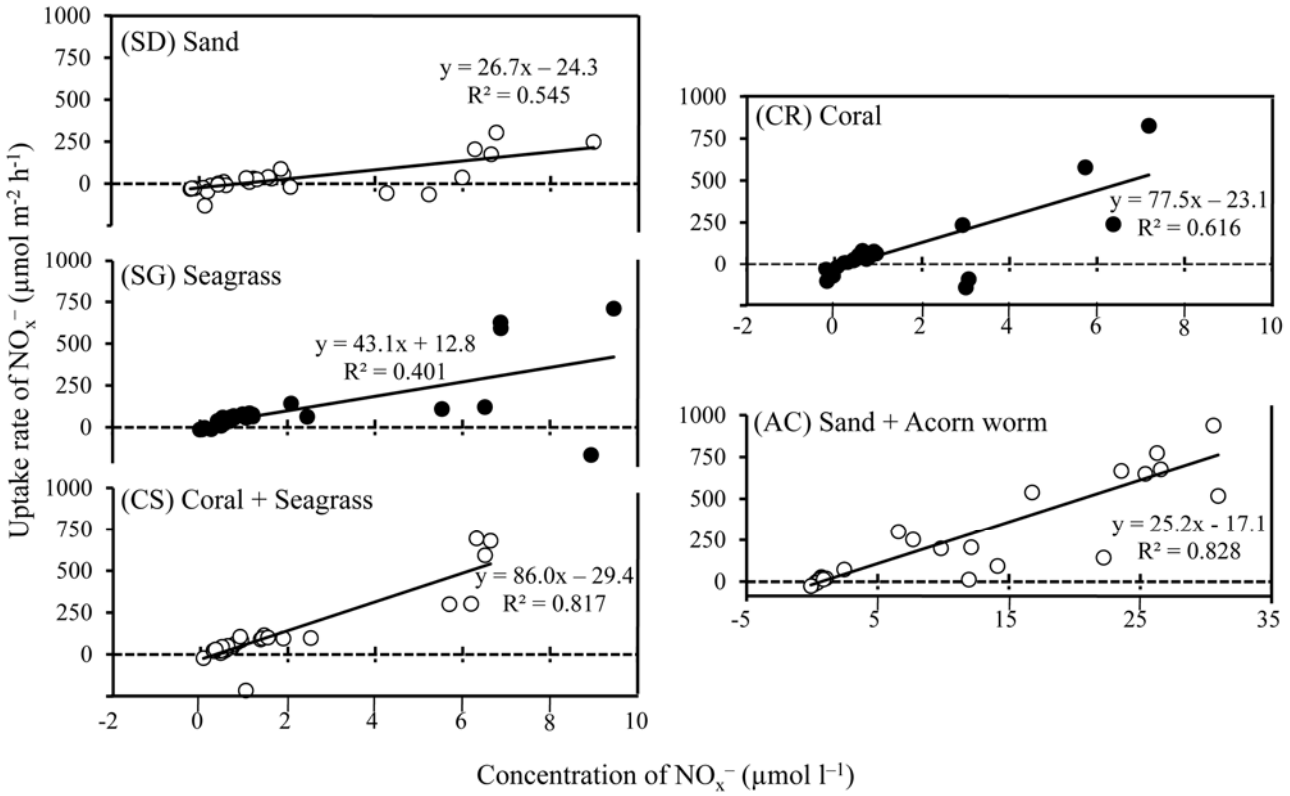

Figure 6 Correlations between the uptake or release rate and the total (initial + inflow) concentration of NO_x in the sand (SD), seagrass (SG), coral-seagrass (CS), coral (CR), and sand-acorn worm (AC) habitats.

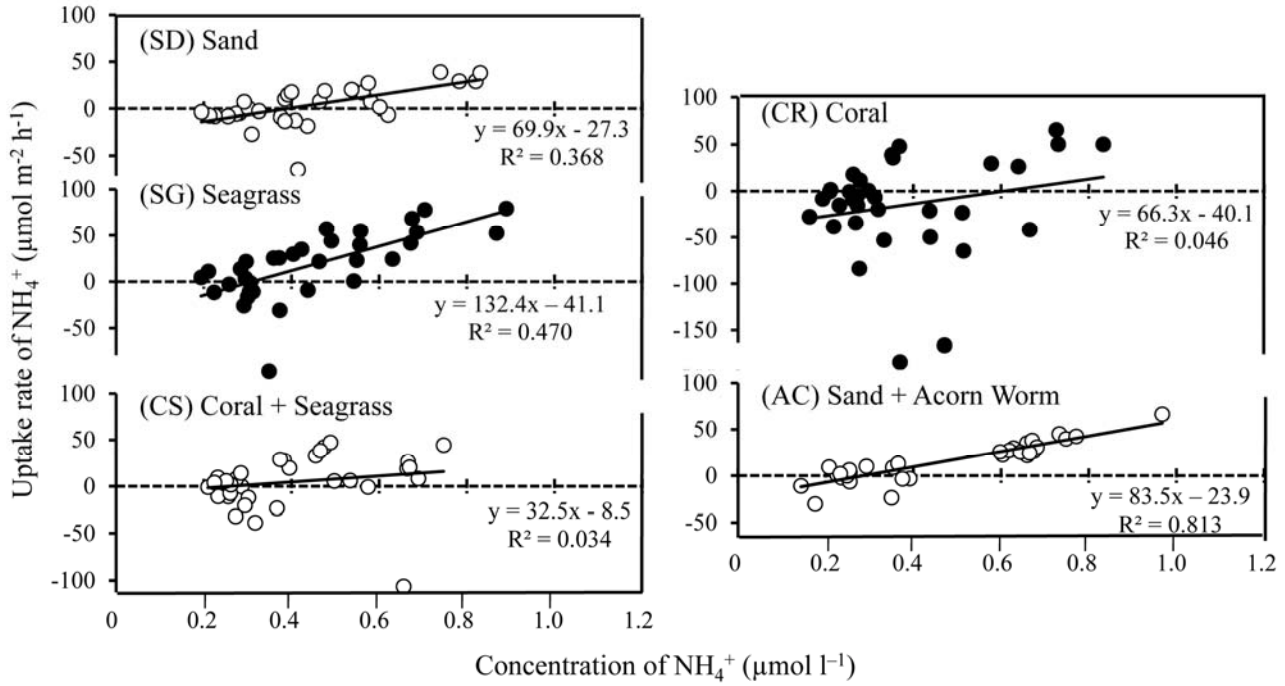

Figure 7 Correlations between the uptake or release rate and the total (initial + inflow) concentration of NH_4^+ in the sand (SD), seagrass (SG), coral-seagrass (CS), coral (CR), and sand-acorn worm (AC) habitats.

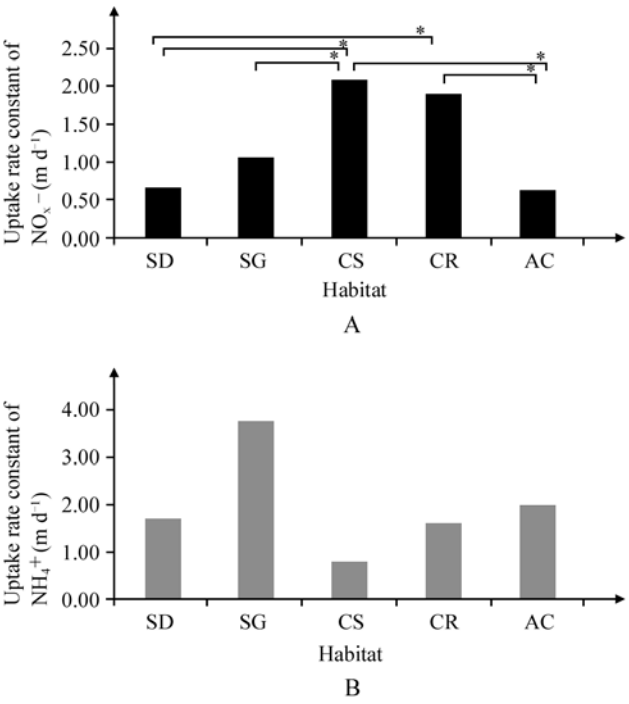

Figure 8 Uptake rate constants of NO_x (A) and NH_4^+ (B) in the sand (SD), seagrass (SG), coral-seagrass (CS), coral (CR), and sand-acorn worm (AC) habitats. * indicates a significant difference between habitats (ANCOVA, Bonferroni adjustment, $p<0.05$). There are no significant differences among the NH_4^+ rate constants

Using the P_{gross} and R_{24h} values calculated from the sand, coral, and seagrass habitats respectively, we can calculate the expected P_{gross} and R_{24h} values in the coral-seagrass mixed habitat based on the sand:coral:seagrass ratio found in the chambers. By applying this ratio (5% sand + 45% coral + 50% seagrass) and summing each parameter, the values are 251.7 for P_{gross} and 235.8 for R_{24h}, respectively. These values were similar to the measured photosynthesis and respiration rates in the coral-seagrass habitat (t-test: $p = 0.97$ and 0.41 respectively). If a synergistic effect were present, we would expect the observed rates to be higher than those calculated in the coral-seagrass habitat. Therefore, there was no synergistic effect of photosynthesis and respiration processes in the coral-seagrass habitat and hence no beneficial relationship between seagrass and coral co-inhabitation with respect to the organic carbon metabolism in the Bise reef moat.

In comparison to the other habitats, the P_{gross}/R_{24h} ratio in the acorn worm habitat had a lower ratio, indicating a higher respiration rate was found in the acorn worm community (Table 1). As bioturbating organisms often create a more oxic environment due to their burrowing activities (Kogure and Wada, 2005), it is possible that the oxygen consumption of microorganisms living in the burrowed layer affected the incubation water as well as respiration of the acorn worm. This is further confirmed in a study conducted by Papaspyrou et al. (2007) which showed that the bioturbating polychaete, *Arenicola marina* positively affected the O_2 flux in the sediment-water interface. In the case of the acorn worm, previous research has stated that the species, *Ptychodera flava* shows an increase in respiratory activity in response to lowered salinity (Azariah et al., 1975). Therefore, it is possible that both the bioturbation activity and the influx of groundwater in the acorn worm habitat concurrently contribute to the higher respiration rate. Our results also indicate that bioturbation of sediments leads to increased dissolution rates. It was considered that more advection of seawater into sediments drives faster rates of dissolution (Rao et al., 2012; Cyronak et al., 2013).

With the exception of the acorn worm habitat, P_{gross}/R_{24h} ratios were 1.0±0.1 which indicates no net production or net consumption, and that the community is slightly autotrophic (Table 1). As the acorn worm habitat is limited to a narrow area along the coastal beach (~10% of the total reef area), the net organic carbon produced by seagrass seems to sustain the nutritional needs of the acorn worm habitat and balances the overall reef ecosystem. This is in agreement with Kinsey (1985) that reported the value of a whole reef system was ~1.0 even if the P_{gross}/R_{24h} ratio varied in different benthic reef environments.

In the sand habitat, the inorganic carbon production (IP) was also low (Figure 5). This suggests that the contribution of micro-calcifying organisms, i.e. foraminiferans or coccolithophorids, to inorganic production in this area is consequently small. In contrast, the coral-seagrass and coral habitats demonstrated that IP was correlated with the photon flux (R^2= 0.70 and 0.76, respectively, Figure 5) which is consistent with the light-enhanced calcification theory of coral reviewed in Gattuso et

al. (1999). The slope between IP vs photon flux in the coral-seagrass habitat was exactly 45% of the slope of coral habitat, indicating a direct relationship between the calcification rate and the amount of coral in the habitat. Thus, in regards to calcification, there is no synergistic effect between coral coexisting with seagrass. On the other hand, $CaCO_3$ dissolution was found in the seagrass and coral-seagrass habitat at night (Table 1). This suggests that respiration by seagrass decreases the pH in the surrounding seawater which causes a decrease in the saturation state of $CaCO_3$. This agrees with Nakamura and Nakamori (2009) who indicated that the dissolution of $CaCO_3$ was observed in seagrass communities. It should be noted however that some dissolution could occur due to the biological processes of bacteria or epilithic and endolithic algae (Islam et al., 2012). The dissolution of $CaCO_3$ in the seagrass habitat is likely not beneficial to the coral with respect to the calcification-dissolution process and the formation of a robust reef framework. On the other hand, the root systems of seagrass elevate the release of alkalinity from sediments, which would elevate the carbonate saturation state of the overlying water column (Burdige and Zimmerman, 2002; Burdige et al., 2008). In the case of the Bise area, the seagrass community seems to provide a moderate dissolution environment for *M. digitata*. Here, the most basal part of *M. digitata* is fragile while the living parts of the coral are healthy. This suggests that the respiration of seagrass roots or stems that are entwined with the basal portion of *M. digitata* could actually decrease the saturation state of $CaCO_3$ by lowering the pH of seawater around that part of the coral. By utilizing this environment, *M. digitata* may contribute to the development of the coral-seagrass and coral habitat through the natural dispersion of fragments as more fragile skeletons allow for easier fragmentation. $CaCO_3$ dissolution was also found in the sand and acorn worm habitat due to a higher respiration rate than photosynthesis (Table 1). However, this habitat does not have any substrate that could be entwined by seagrass, and it is difficult for coral fragments to survive in the sandy habitats. This is evident in a study which reported more than 70% mortality of *Acropora palmata* fragments transplanted onto a sand substrate (Lirman, 2000). Therefore, it is possible that the respiration of the seagrass which can entwine the basal part of coral may be beneficial to coral by encouraging the dispersal of fragments via the partial dissolution of calcium carbonate in the Bise area.

4.2 Inorganic nitrogen dynamics

The inorganic nitrogen dynamics in the Bise area are highly impacted by groundwater inflow during spring low tide. This is evident in the decrease in salinity and the increase in inorganic nitrogen concentrations in each chamber area during low vs. high tide (Table 2). The strong correlations found between concentration and uptake rate in each habitat indicate each of the habitats are likely nitrogen limited and that groundwater inflow stimulates microbial activity. Thus the habitats with groundwater inflow will demonstrate higher inorganic nitrogen uptake rates. This is seen in the acorn worm habitat, which is located nearest to the coast, exposed directly to groundwater inflow, and exhibits the highest NO_x uptake rate. It should be noted however, that although the high uptake rate in the acorn worm habitat during low tide was prominently due to the correlative increase in the NO_x concentration, the uptake rate constant was not significantly different from the sand area (Figure 8). The lack of a significant difference between the NO_x uptake rate constants in the acorn worm and sand habitats indicate that the impact of acorn worm activity on the coupled nitrification-denitrification processes are negligible. This is surprising as burrowing macrofauna generally stimulate nitrification and denitrification processes by increasing sediment oxygen concentration, redox potential, and bacteria-mediated processes (Krantzberg, 1985; Kogure and Wada, 2005).

We attribute our inability to see the impact of acorn worm activity on the inorganic nitrogen dynamics to two possibilities. First, there is a lower population density of acorn worms at this site (maximum 24 individuals m^{-2}), and second, in line with recent studies, the sediment characteristics could be masking the impact of the acorn worm. Mermillod-Blondin and Rosenberg (2006) demonstrated that in diffusion dominated sediments, burrowing macrofauna have the potential to increase oxygen consumption up to 3-fold, whereas

in advection dominated sediments, their impact is much lower (-20% to +30%). The coral rubble that is the primary foundation of the benthos in the Bise ecosystem, and the high influx of groundwater implies that it is an advection dominated system. Thus the coastal sands of Bise are neither nitrogen nor oxygen limited and the impact of the acorn worm on the inorganic nitrogen cycle in Bise is less apparent. Instead, the NO_x uptake in the sand and acorn worm habitats may be dominated by autochthonous microorganisms and epilithic algae. Although less effective, it is possible that the NO_x uptake by sand-related microbes in the acorn worm habitat may suppress further increases in the NO_x concentration found in the nearby seagrass, coral, and coral-seagrass habitats. By mitigating NO_x increases, the sand in the acorn worm and sand habitats may aid in maintaining a healthy coral community.

The coral-seagrass and coral habitats demonstrated a significantly higher uptake rate constant than the other habitats (Figure 8). These rate constants are comparable to estimates (4.4±0.4 m d^{-1} for NO_3^-) derived from a coral reef in Biosphere 2 (Atkinson et al. 2001). When the rate constants of the sand, seagrass, and coral habitats (0.6, 1.0 and 1.9 m d^{-1}, respectively) were used to calculate the expected rate constant for a 5% sand, 50% seagrass and 45% coral mixed habitat, we estimated 1.4 m d^{-1} which is lower than the observed constant of the coral-seagrass habitat (Figure 8). This estimate indicates that the co-existence of coral and seagrass can remove more NO_x from the water column than would be expected.

Moreover, because of the significant difference in rate constants between the seagrass and coral-seagrass habitats, it is possible that the coexistence of coral and seagrass may benefit seagrass with respect to NO_x. It is known that the most important factors of nutrient exchange between water and the benthic community are the nutrient concentration, water velocity, and friction with the benthos (Atkinson, 2011). Falter and Atkinson (2004) formulated the rate constant which is a molar mass-transfer coefficient as a function of the roughness of a community, the diffusivity of the nutrients and the ambient flow conditions. Since the diffusivity and flow conditions are similar across all the habitats enclosed by the chambers, the coexistence of coral and seagrass may increase this friction by creating the complex structure of entwined coral and seagrass. Additionally, since this system is not nutrient deficient, it is not necessary for coral and seagrass to compete for nutrient acquisition. If the system were to exist under nutrient deficient conditions, different mechanisms would likely be involved in the competitive co-existence between seagrass and coral.

Overall, the ammonium uptake rates were relatively low due to the low concentration of ammonium in Bise. Previous studies in the seagrass area indicated the ambient concentration of NH_4^+ ranged between 0.5 – 5.4 μM (Romero et al., 2006). Although the present study found a lower average of 0.43 ± 0.20 μM than that of a general seagrass area, the value was within the range of a typical coral reef area which was reported as 0.7 ± 0.7 μM at the One Tree reef lagoon, Australia (Steven and Atkinson, 2003) and 0.76 ± 0.49 at Rukan-sho atoll off Okinawa Island, Japan (Ohde and van Woesik, 1999). The uptake rate constant of the seagrass habitat seemed to be the highest among all the habitats, however there were no significant differences between habitats (Figure 8). This may be due to a lower correlation between the uptake rate and concentration in the coral-seagrass and coral habitats (R^2=0.03 and 0.05, respectively, Figure 7) which is mainly caused by the release of ammonium from coral.

More than half of the ammonium flux values in the coral habitat were negative, indicating a net release of ammonium. Most of these rates corresponded to samples collected during the night and morning incubations. Muscatine and D'Elia (1978) and Muller et al. (2009) reported ammonium release by the coral host and uptake/ retention by zooxanthellae, resulting in a net uptake of ammonium via the coral-zooxanthellae symbiosis. They also demonstrated that under normal seawater conditions, which are nitrogen limited, a net release was produced by *Pocillopora damicornis* after more than 12 hours incubation in the dark, resulting in a depletion of the photosynthetically produced energy in zooxanthellae. However, due to the high NO_x

availability in this study site, our results contradict these findings and suggest that when inorganic nitrogen supplies are sufficient, zooxanthellae do not need to take up ammonium, resulting in a net release from coral-zooxanthellae symbiosis.

The ammonium concentration in the coral-seagrass habitat however was lower than that in the coral habitat (Figure 7 and Table 2). Stapel et al. (1996) demonstrated *T. hemprichii* leaves have a high capacity for ammonium uptake. As we found a strong linear relationship between ammonium uptake rates and concentration in the seagrass habitat, the lower ammonium concentrations in the coral-seagrass habitat are attributed to the ability of the coexisting seagrass to take up and retain the excess ammonium released by coral. Thus, the coral-seagrass habitat shows a possible beneficial relationship in terms of ammonium. Although the lack of a strong correlation between concentration and uptake rate in the coral-seagrass and coral habitats demonstrate that the coral community does not benefit from increased ammonium concentrations, this indicates a somewhat "commensalistic" biogeochemical relationship between seagrass and coral in regards to nitrogen nutrient dynamics in the Bise area.

5 Conclusion

While this study found no synergistic effects between coral and seagrass with respect to the photosynthesis-respiration and calcification processes, it is possible that $CaCO_3$ dissolution driven by seagrass could be facilitating coral fragmentation and fragment distribution. This could benefit *M. digitata* by furthering habitat development.

In contrast, the NO_x results demonstrated that there is a synergistic effect between coral and seagrass in the coral-seagrass habitat. This is evident in the higher uptake rate constant of NO_x in the coral-seagrass habitat than that estimated from each habitat alone. In addition to this, significant differences between the rate constants of the coral-seagrass and seagrass habitats demonstrated that the coral-seagrass habitat benefit seagrass with respect to NO_x uptake. In the acorn worm habitat however, the high NO_x uptake possibly aids in maintaining a lower nitrate concentration in this coral reef ecosystem. Overall, the coral reef ecosystem in Bise assimilates the high input of NO_x by demonstrating continual uptake regardless of the benthic composition. On the other hand ammonium was released by coral because of the sufficient nitrogen source as nitrate. As ammonium is the most easily assimilated by all organisms, the released ammonium was taken up by seagrass in the coral-seagrass habitat. Therefore, on a strictly biogeochemical scale, the coral-seagrass relationship is more beneficial for the seagrass in terms of inorganic nitrogen dynamics.

However, continued research (such as microbial activity, abundance of microbes in the sediments, stable isotope analyses or ex-situ experiments) needs to be conducted to clarify both the physical and biogeochemical relationships/ mechanisms occurring within the coral-seagrass habitats.

Acknowledgements

This study was part of the 2008 COE (Center of Excellence) Summer Program supported by the 21st Century COE Program (University of the Ryukyus). The authors wish to thank the people of the Bise, Okinawa municipality for their support and cooperation during the research period. We also thank 3 anonymous reviewers for valuable comments to improve the manuscript.

References

Atkinson M.J., Falter J.L., and Hearn C.J., 2001, Nutrient dynamics is the Biosphere 2 coral reef mesocosm: water velocity controls NH_4 and PO_4 uptake. Coral Reefs, 20: 341-346
http://dx.doi.org/10.1007/s00338-001-0184-7

Atkinson M.J., 2011, Biogeochemistry of nutrients, in: Dubinsky, Z., Stambler, N. (Eds.), Coral Reefs: An Ecosystem in Transition. Springer, Dordrecht, pp. 199-206
http://dx.doi.org/10.1007/978-94-007-0114-4_13
PMCid:PMC3151229

Azariah J., Ismail M.M., and Najib M.A., 1975, Investigation on the ecology and respiratory responses of the hemichordate *Ptychodera flava* to tidal cycles and salinity changes. The biological bulletin, 149: 455-466
http://dx.doi.org/10.2307/1540379

Badgley B.D., Lipschultz F., and Sebens K.P., 2006, Nitrate uptake by the reef coral *Diploria strigosa*: effects of concentration, waterflow and irradiance. Marine Biology, 149: 327-338
http://dx.doi.org/10.1007/s00227-005-0180-5

Bellwood D.R., Hughes T.P., Folke C., and Nyström M., 2004, Confronting the coral reef crisis. Nature, 429: 827-833
http://dx.doi.org/10.1038/nature02691 PMid:15215854

Bensoussan N., and Gattuso J.P., 2007, Community primary production and calcification in a NW Mediterranean ecosystem dominated by calcareous macroalgae. Marine Ecology Progress Series, 334: 37-45
http://dx.doi.org/10.3354/meps334037

Burdige D.J., and Zimmerman R.C., 2002, Impact of sea grass density on carbonate dissolution in Bahamian sediments. Limnology and Oceanography, 47: 1751-1763
http://dx.doi.org/10.4319/lo.2002.47.6.1751

Burdige D.J., Zimmerman R.C., and Hu X., 2008, Rates of carbonate dissolution in permeable sediments estimated from pore-water profiles: the role of sea grasses. Limnology and Oceanography, 53: 549-565
http://dx.doi.org/10.4319/lo.2008.53.2.0549

Cyronak T., Santos I.R., McMahon A., and Eyre B.D., 2013, Carbon cycling hysteresis in permeable carbonate sands over a diel cycle:

implications for ocean acidification. Limnology and Oceanography, 58: 131-143

Falter J.L., Atkinson M.J., and Merrifield M.A., 2004, Mass-transfer limitation of nutrient uptake by a wave-dominated reef flat community. Limnology and Oceanography, 49: 1820-1831
http://dx.doi.org/10.4319/lo.2004.49.5.1820

Gattuso J.P., Allemand D., and Frankignoulle M., 1999, Photosynthesis and calcification at cellular, organismal and community levels in coral reefs: a review on interactions and control by carbonate chemistry. American Zoologist, 39: 160-183

Grover R., Maguer J., Allemand D., and Ferrier-Pagès C., 2003, Nitrate uptake in the scleractinian coral *Stylophora pistillata*. Limnology and Oceanography, 48: 2266-2274
http://dx.doi.org/10.4319/lo.2003.48.6.2266

Ishikawa Y., Fujimura H., and Suzuki Y., 2007, Closed-chamber system for primary production of benthic communities in sub-tropical lagoon. Eco-Engineering, 19: 173-177

Islam N.M., Casareto B.E., Higuchi T., Niraula M.P., and Suzuki Y., 2012, Contribution of coral rubble associated microbial community to the dissolution of calcium carbonate under high pCO_2. Galaxea JCRS, 14: 119-131

Jompa J., and McCook L.J., 2003, Coral-algal competition: macroalgae with different properties have different effects on corals. Marine Ecology Progress Series, 258: 87-95
http://dx.doi.org/10.3354/meps258087

Kawahata H., Yukino I., and Suzuki A., 2000, Terrestrial influences on the Shiraho fringing reef, Ishigaki Island, Japan: high carbon input relative to phosphate. Coral Reefs 19: 172-178
http://dx.doi.org/10.1007/s003380000093

Kinsey D.W., 1985, Metabolism, calcification, carbon production I: System level studies. Proceedings of the fifth international coral reef congress, 4: 505-526

Kogure K., and Wada M., 2005, Impacts of Macrobenthic Bioturbation in Marine Sediment on Bacterial Metabolic Activity. Microbes and Environments, 20: 191-199
http://dx.doi.org/10.1264/jsme2.20.191

Krantzberg G., 1985, The influence of bioturbation on physical, chemical and biological parameters in aquatic encvironments: A review. Environmental Pollution (Series A), 39: 99-122
http://dx.doi.org/10.1016/0143-1471(85)90009-1

Langdon C., Broecker W.S., Hammond D., Glenn E., Fitzsimmons K., Nelson S.G., Peng T.H., Hajdas I., and Bonani G., 2003, Effect of elevated CO_2 on the community metabolism of an experimental coral reef. Global Biogeochemical Cycles, 17: 11-1-11-14

Lirman D., 2000, Fragmentation in the branching coral *Acropora palmate* (Lamarck): growth, survivorship, and reproduction of colonies and fragments. Journal of Experimental Marine Biology and Ecology, 251: 41-57
http://dx.doi.org/10.1016/S0022-0981(00)00205-7

Marbà N., Holmer M., Garcia E., and Barrón C., 2006, Seagrass beds and coastal biogeochemistry. In: Larkum AWD, Orth RJ, Duarte CM, editors. Seagrasses: Biology, Ecology and Their Conservation., Kluwer Academic Publishers BV Dordrecht, The Netherlands: 135-157
http://dx.doi.org/10.1007/1-4020-2983-7_6
http://dx.doi.org/10.1007/978-1-4020-2983-7_6

Manzello D.P., Enochs I.C., Melo N., Gledhill D.K., and Johns E.M., 2012, Ocean Acidification Refugia of the Florida Reef Tract. PLoS ONE, 7: e41715
http://dx.doi.org/10.1371/journal.pone.0041715

Mermillod-Blondin F., and Rosenberg R., 2006, Ecosystem engineering: the impact of bioturbation on biogeochemical processes in marine and freshwater benthic habitats. Aquatic Sciences, 68: 434-442
http://dx.doi.org/10.1007/s00027-006-0858-x

Miyajima T., Suzumura M., Umezawa Y., and Koike I., 2001, Microbiological nitrogen transformation in carbonate sediments of coral-reef lagoon and associated seagrass beds. Marine Ecology Progress Series, 217: 273-286
http://dx.doi.org/10.3354/meps217273

Moberg F., and Folke C., 1999, Ecological goods and services of coral reef ecosystems. Ecological Economics, 29: 215-233
http://dx.doi.org/10.1016/S0921-8009(99)00009-9

Muller E.B., Kooijman S.A.L.M., Edmunds P.J.E., Doyle F.J., and Nisbet R.M., 2009, Dynamic energy budgets in syntrophic symbiotic relationships between heterotrophic hosts and photoautotrophic symbionts. Journal of Theoretical Biology, 259: 44-57
http://dx.doi.org/10.1016/j.jtbi.2009.03.004 PMid:19285512

Muscatine L., and D'Elia C.F., 1978, The uptake, retention, and release of ammonium by reef corals. Limnology and Oceanography, 23: 725-734
http://dx.doi.org/10.4319/lo.1978.23.4.0725

Naeem S., 1998, Species redundancy and ecosystem reliability. Conservation Biology, 12: 39-35
http://dx.doi.org/10.1111/j.1523-1739.1998.96379.x
http://dx.doi.org/10.1046/j.1523-1739.1998.96379.x

Nakamura T., and Nakamori T., 2009, Estimation of photosynthesis and calcification rates at a fringing reef by accounting for diurnal variations and the zonation of coral reef communities on reef flat and slope: a case study for the Shiraho reef, Ishigaki Island, southwest Japan. Coral Reefs, 28: 229-250
http://dx.doi.org/10.1007/s00338-008-0454-8

Ninomiya S., Kurahashi S., Masumoto T., Iwashita T., and Nakano Y., 2006, A study on comprehensive management of coral reef areas: Restoration technologies focusing on a symbiotic relationship between corals and seagrasses in moats. Proceedings of 10th International Coral Reef Symposium: 1617-1626

Ohde S., and van Woesik R., 1999, Carbon dioxide flux and metabolic processes of a coral reef, Okinawa. Bulletin of Marine Science, 65: 559-576

Papaspyrou S., Kristensen E., and Christensen B., 2007, *Arenicola marina* (Polychaeta) and organic matter mineralisation in sandy marine sediments: *In situ* and microcosm comparison. Estuarine, Coastal and Shelf Science, 72: 213-222
http://dx.doi.org/10.1016/j.ecss.2006.10.020

Rao A.M.F., Polerecky L., Ionescu D., Meysman F. J. R., and De Beer D., 2012, The influence of pore-water advection, benthic photosynthesis, and respiration on calcium carbonate dynamics in reef sands. Limnology and Oceanography, 57: 809-825
http://dx.doi.org/10.4319/lo.2012.57.3.0809

Romero J., Lee K., Pérez M., Mateo M.A., and Alcoverro T., 2006, Nutrient dynamics in seagrass ecosystems, in: Larkum AWD, Orth RJ, Duarte CM. editors. Seagrasses: Biology, Ecology and Their Conservation., Kluwer Academic Publishers BV Dordrecht, The Netherlands: 227-254
http://dx.doi.org/10.1007/1-4020-2983-7_9

Smith S.V., 1973, Carbon dioxide dynamics: a record of organic carbon production, respiration, and calcification in the Eniwetok reef flat community. Limnology and Oceanography, 18: 106-120
http://dx.doi.org/10.4319/lo.1973.18.1.0106
http://dx.doi.org/10.4319/lo.1973.18.6.0953

Smith S.V., and Kinsey D.W., 1978, Calcification and organic carbon metabolism as indicated by carbon dioxide. In: Stoddart DR, Johannes RE (eds), Coral reefs: research methods, UNESCO, Paris, pp. 469-484

Stapel J., Aarts T.L., van Duynhoven B.H.M., de Groot J.D., van den Hoogen P.H.W., and Hemminga M.A., 1996, Nutrient uptake by leaves and roots of the seagrass *Thalassia hemprichii* in the Spermonde Archipelago, Indonesia. Marine Ecology Progress Series, 134: 195-206
http://dx.doi.org/10.3354/meps134195

Steven A.D.L., and Atkinson M.J., 2003, Nutrient uptake by coral-reef microatolls. Coral Reefs, 22: 197-204
http://dx.doi.org/10.1007/s00338-003-0303-8

Tenore K.R., 1988, Nitrogen in Benthic Food Chains. In: Blackburn TH, Sørensen J, editors. Nitrogen Cycling in Coastal Marine Environments. Essex: John Wiley and Sons Ltd, pp. 191-206

Tribollet A., Langdon C., and Atkinson M.J., 2006, Effects of elevated pCO_2 on epilithic and endolithic metabolism of reef carbonates. Global Change Biology, 12: 1-9

Umezawa Y., Miyajima T., Kayanne H., and Koike I., 2002, Significance of groundwater nitrogen discharge into coral reefs at Ishigaki Island, southwest of Japan. Coral Reefs, 21: 346-356

Table 2 Summary of the data collected for each sand (SD), seagrass (SG), coral-seagrass (CS), coral (CR), and sand-acorn worm (AC) habitat chamber during each incubation trial. OP, IP, and nutrients uptake rates are averages ± SE (n=3, each). The range of values indicates the values at the beginning and end of each 2-h incubation, respectively.

Date & Time	Temp. (ºC)	Photon flux ($\mu mol\ m^{-2}\ s^{-1}$)	Water level (cm)	Habitat	pH	Salinity	Alkalinity ($\mu mol\ kg^{-1}$)	NO_X^- (μM)	NH_4^+ (μM)	ΔC_{inflow} Alkalinity ($\mu mol\ kg^{-1}$)	ΔC_{inflow} NO_X^- (μM)	ΔC_{inflow} NH_4^+ (μM)	Carbon production Organic ($mmol\ m^{-2}\ h^{-1}$)	Carbon production Inorganic ($mmol\ m^{-2}\ h^{-1}$)	Uptake rate NO_X^- ($\mu mol\ m^{-2}\ h^{-1}$)	Uptake rate NH_4^+ ($\mu mol\ m^{-2}\ h^{-1}$)
1-Aug	31.2	2153	12 – -4	SD	8.332–8.378	33.43–32.96	2442–2493	4.27–4.33	0.53–0.36	45	3.19	0.11	2.6±0.7	-0.2±0.1	243±37	22±4
12–14			(Low tide)	SG	8.311–8.600	33.26–33.08	2461–2480	6.53–3.42	0.65–0.45	17	1.20	0.04	27.3±2.4	-0.1±0.2	643±35	35±23
				CS	8.352–8.519	33.31–33.27	2444–2352	6.24–2.09	0.66–0.85	3	0.24	0.01	20.2±4.3	7.1±2.1	656±32	-26±40
				CR	8.429–8.481	33.55–33.46	2401–2216	4.69–1.60	0.66–0.35	8	0.59	0.02	14.9±1.1	14.4±1.1	548±171	49±10
				AC	8.295–8.223	32.81–30.95	2543–2740	10.95–16.33	0.72–0.19	182	12.79	0.03	-1.0±0.8	-0.6±0.2	575±47	44±11
1-Aug	30.3	1109	70–158	SD	8.649–8.600	32.69–32.93	2486–2430	4.47–2.58	0.78–0.36	-23	-1.64	-0.05	-2.3±2.8	1.3±0.4	19±99	28±6
16–18			(Low tide)	SG	8.534–8.472	33.65–34.06	2397–2382	2.48–0.27	0.69–0.23	-40	-2.83	-0.09	-8.3±2.2	-1.8±0.8	-93±78	54±8
				CS	8.499–8.409	33.87–34.20	2370–2343	1.30–0.10	0.55–0.20	-32	-2.27	-0.07	-9.3±2.0	-0.4±0.7	-160±32	41±4
				CR	8.389–8.354	34.22–34.29	2349–2338	0.36–0.32	0.37–0.08	-7	-0.50	-0.02	-2.9±1.0	0.3±0.3	-68±21	41±4
				AC	8.344–8.365	29.02–29.75	2819–2774	32.82–17.40	0.64–0.30	-53	-3.76	-0.01	0.0±0.1	-0.3±0.1	906±68	25±1
1-Aug	28.7	4	182–189	SD	8.341–8.300	34.21–34.28	2369–2364	0.72–0.40	0.30–0.32	-6	-0.45	-0.01	-1.8±0.6	0.0±0.0	-10±8	-3±2
19–21			(High tide)	SG	8.358–8.223	34.25–34.28	2371–2378	0.76–0.38	0.31–0.40	-3	-0.20	-0.01	-10.9±0.2	-0.7±0.5	28±19	-14±7
				CS	8.321–8.249	34.29–34.28	2365–2366	0.53–0.40	0.28–0.33	1	0.10	0.00	-5.4±1.5	0.0±0.2	34±6	-8±4
				CR	8.301–8.183	34.30–34.28	2360–2345	0.51–0.28	0.24–0.20	2	0.12	0.00	-7.4±0.4	1.3±0.1	52±1	6±6
				AC	8.383–8.351	34.22–34.28	2374–2381	0.77–0.32	0.24–0.23	-6	-0.41	0.00	-1.8±0.2	-0.5±0.0	3±13	1±1
4-Aug	31.0	674	20–92	SD	8.323–8.360	32.78–33.21	2543–2488	8.11–5.51	0.70–0.59	-42	-2.95	-0.10	1.4±0.7	0.5±0.2	-27±32	1±4
16–18			(Low tide)	SG	8.344–8.399	32.93–33.28	2520–2479	9.42–6.84	0.78–0.43	-35	-2.43	-0.08	4.2±2.2	0.5±0.4	23±94	39±9
				CS	8.374–8.401	33.07–33.33	2482–2437	6.07–3.44	0.75–0.49	-25	-1.76	-0.06	2.7±1.9	1.5±0.2	129±172	30±7
				CR	8.409–8.399	33.13–33.34	2466–2414	5.60–4.13	0.76–0.63	-21	-1.46	-0.05	0.2±0.7	2.4±0.5	2±120	12±28
				AC	8.244–8.214	31.95–31.40	2635–2714	20.92–18.39	0.65–0.26	54	3.79	0.01	-1.3±0.3	-1.0±0.1	491±173	31±7
4-Aug	28.6	0	172–195	SD	8.250–8.202	34.28–34.27	2380–2380	0.38–0.62	0.40–0.56	0	0.03	0.00	-1.7±0.7	0.0±0.2	-16±15	-12±10
20–22			(High tide)	SG	8.253–8.116	34.29–34.28	2374–2391	0.46–0.38	0.47–0.31	0	0.03	0.00	-10.4±0.4	-1.2±0.2	17±4	24±16
				CS	8.248–8.061	34.29–34.28	2363–2367	0.43–0.38	0.25–0.37	1	0.05	0.00	-12.5±0.2	-0.2±0.2	14±4	-17±8
				CR	8.226–8.048	34.29–34.29	2375–2362	0.44–0.28	0.38–1.02	0	0.01	0.00	-10.6±1.2	1.0±0.2	24±3	-94±46
				AC	8.282–8.220	34.27–34.29	2323–2356	0.64–0.36	0.21–0.25	-2	-0.13	0.00	-3.4±0.3	-1.3±0.0	12±4	-3±4
5-Aug	28.7	777	142–196	SD	8.147–8.135	34.28–34.17	2383–2384	0.40–0.79	0.29–0.38	11	0.74	0.02	0.0±0.0	0.3±0.0	27±3	-5±3
7–9			(High tide)	SG	8.151–8.275	34.28–34.21	2380–2379	0.34–0.37	0.39–0.24	7	0.48	0.02	9.4±1.6	0.6±0.2	68±7	24±27
				CS	8.174–8.244	34.29–34.21	2384–2364	0.32–0.36	0.21–0.20	7	0.52	0.02	6.7±0.8	2.1±0.9	72±7	4±3
				CR	8.172–8.181	34.30–34.23	2378–2372	0.29–0.35	0.24–0.45	7	0.46	0.02	1.6±0.7	1.0±0.5	60±2	-30±13
				AC	8.154–8.147	34.25–34.19	2389–2407	0.48–0.68	0.33–0.34	6	0.42	0.00	-0.6±0.1	-0.5±0.3	17±2	0±5

Table 2 Continued

Date & Time	Temp. (ºC)	Photon flux ($\mu mol\ m^{-2}\ s^{-1}$)	Water level (cm)	Habitat	pH	Salinity	Alkalinity ($\mu mol\ kg^{-1}$)	NO_X^- (μM)	NH_4^+ (μM)	ΔC_{inflow} Alkalinity ($\mu mol\ kg^{-1}$)	ΔC_{inflow} NO_X^- (μM)	ΔC_{inflow} NH_4^+ (μM)	Carbon production Organic ($mmol\ m^{-2}\ h^{-1}$)	Carbon production Inorganic ($mmol\ m^{-2}\ h^{-1}$)	Uptake rate NO_X^- ($\mu mol\ m^{-2}\ h^{-1}$)	Uptake rate NH_4^+ ($\mu mol\ m^{-2}\ h^{-1}$)
5-Aug	29.5	2031	193–132	SD	8.189–8.255	34.18–34.21	2417–2423	0.68–0.41	0.42–0.23	-3	-0.20	-0.01	2.2±0.3	-0.4±0.1	5±3	15±3
10–12			(High tide)	SG	8.195–8.446	34.18–34.21	2416–2412	0.72–0.27	0.39–0.20	-3	-0.22	-0.01	20.1±1.1	0.1±0.2	34±5	27±1
				CS	8.188–8.417	34.20–34.22	2430–2363	0.63–0.29	0.31–0.22	-2	-0.12	0.00	21.7±3.2	4.9±0.4	33±10	13±8
				CR	8.190–8.341	34.20–34.22	2435–2324	0.58–0.26	0.25–0.39	-2	-0.17	-0.01	17.5±0.5	8.1±0.3	22±7	-23±8
				AC	8.186–8.284	34.19–34.19	2437–2413	0.64–0.41	0.34–0.41	-1	-0.04	0.00	4.5±0.3	0.9±0.1	15±1	-6±10
5-Aug	31.6	2349	94–32	SD	8.333–8.417	34.24–34.08	2422–2431	0.30–1.10	0.17–0.29	16	1.15	0.04	4.1±0.2	0.2±0.0	27±9	-6±1
13–15			(Low tide)	SG	8.358–8.517	34.25–34.02	2417–2433	0.33–1.30	0.22–0.32	22	1.58	0.05	15.4±2.4	0.5±0.2	91±26	-6±5
				CS	8.356–8.488	34.25–34.02	2416–2390	0.34–1.26	0.27–0.51	23	1.62	0.05	15.0±1.7	3.7±0.9	103±6	-27±6
				CR	8.407–8.492	34.28–34.21	2386–2220	0.27–0.49	0.31–0.57	7	0.48	0.02	16.5±1.5	12.8±1.6	39±8	-37±24
				AC	8.349–8.367	34.21–33.10	2429–2581	0.40–4.78	0.19–0.30	108	7.62	0.02	1.2±0.7	-1.7±0.3	252±29	-7±12
6-Aug	29.3	0	59–49	SD	8.204–8.094	34.23–34.12	2455–2488	0.91–0.96	0.38–0.79	11	0.78	0.03	-4.5±0.4	-0.8±0.1	57±18	-30±17
3–5			(Low tide)	SG	8.181–8.020	34.25–34.21	2454–2487	0.88–0.64	0.38–0.69	4	0.25	0.01	-12.1±1.5	-2.1±0.5	73±8	-45±26
				CS	8.166–7.999	34.25–34.18	2457–2467	0.98–0.80	0.52–0.51	7	0.46	0.02	-10.6±0.5	-0.3±0.5	96±3	4±2
				CR	8.104–7.982	34.28–34.27	2442–2441	0.78–0.42	0.47–1.01	2	0.11	0.00	-9.4±0.7	0.2±0.3	70±4	-80±44
				AC	8.156–7.974	33.95–32.54	2503–2657	3.00–11.37	0.66–0.24	138	9.72	0.02	-5.2±0.2	-0.6±0.2	105±56	35±5
6-Aug	29.0	374	80–148	SD	8.066–8.078	34.06–34.20	2460–2431	1.67–0.61	0.65–0.25	-14	-0.97	-0.03	0.7±0.3	0.6±0.2	7±14	29±5
6–8			(Low tide)	SG	8.032–8.124	34.13–34.22	2461–2436	1.07–0.15	0.49–0.16	-9	-0.63	-0.02	6.6±1.0	1.2±0.8	43±8	45±6
				CS	8.040–8.109	34.14–34.24	2462–2425	1.19–0.19	0.43–0.22	-10	-0.71	-0.02	5.8±0.7	2.0±0.6	44±37	29±5
				CR	8.049–8.070	34.22–34.27	2462–2408	0.51–0.16	0.31–0.33	-5	-0.34	-0.01	4.3±0.4	3.7±0.1	2±8	-5±9
				AC	8.084–8.081	34.00–34.09	2476–2468	1.90–0.87	0.71–0.28	-10	-0.67	0.00	-0.3±0.3	-0.1±0.3	27±23	33±5
6-Aug	29.5	1300	172–183	SD	8.122–8.276	34.19–34.31	2467–2459	0.63–0.20	0.37–0.24	-11	-0.79	-0.03	5.5±0.3	-0.1±0.1	-28±1	8±6
9–11			(High tide)	SG	8.146–8.389	34.22–34.27	2451–2439	0.40–0.11	0.28–0.16	-5	-0.35	-0.01	19.0±2.3	0.5±0.3	-9±3	16±3
				CS	8.165–8.353	34.26–34.26	2452–2412	0.29–0.15	0.25–0.20	0	0.03	0.00	16.8±1.7	3.0±0.3	24±3	8±3
				CR	8.190–8.276	34.32–34.26	2462–2375	0.26–0.12	0.22–0.25	5	0.38	0.01	12.0±0.9	6.9±0.5	77±3	-2±3
				AC	8.154–8.245	34.21–34.27	2441–2460	0.30–0.23	0.29–0.19	-6	-0.39	0.00	2.5±0.1	-1.0±0.1	-25±1	7±3