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Different ingestion patterns of ^{13}C-labeled bacteria and algae by deep-sea benthic foraminifera

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ABSTRACT: Benthic foraminiferal food sources were examined in the central part of Sagami Bay, Japan (water depth 1450 m) based on an *in situ* feeding experiment with ^{13}C-labeled food materials. In this study, 3 different ^{13}C-labeled food materials were used: the unicellular marine algae *Dunaliella tertiolecta*, the marine diatom *Chaetoceros sociale*, and the marine bacterium *Vibrio alginolyticus*. The first two are representatives of phytodetritus and the third of organic matter produced in the sediments. Each type of food material was injected into a series of *in situ* culture cores and incubated for up to 21 d. We observed that some benthic foraminiferal species selectively ingested ^{13}C-labeled algae from the sedimentary organic matter. On the other hand, benthic foraminifera ingested ^{13}C-labeled bacteria unselectively from sedimentary organic matter. Total benthic foraminifera assimilated 8.8 mg C m^{-2} d^{-1} of sedimentary organic matter without phytodetritus assimilation. Based on the assimilation rates estimated in this experiment, we recognized 3 types of feeding strategy among deep-sea benthic foraminifera in Sagami Bay. There are those that ingest (1) fresh phytodetritus selectively (phytophagous species: *Uvigerina akitaensis*, *Bolivina spissa*, *Bolivina pacifica*); (2) fresh phytodetritus selectively but sedimentary organic matter as well when phytodetritus is absent (seasonal-phytophagous species: *Bulimina aculeata*, *Textularia kattegatensis*, *Globobulimina affinis*); and (3) sedimentary organic matter at random (deposit feeders: *Cyclammina cancellata*, *Chilostomella ovoidea*). These different types of carbon utilization should be considered not only for understanding modern ecosystems on the deep-sea floor but also for paleoceanographic reconstructions using the abundance and distribution, or isotopic composition, of benthic foraminifera.

KEY WORDS: Benthic foraminifera · Feeding ecology · *In situ* tracer experiment · Carbon budget · Deep-sea

INTRODUCTION

Organic carbon fluxes from the photic zone to the seafloor are thought to be a major food source for deep-sea benthic organisms, excluding chemosynthetic communities (Gooday 1988, Graf 1989). Some benthic taxa in the deep-sea exhibit seasonal reproduction and/or growth that reflects seasonal deposition of phytodetritus (e.g. Campos-Creasey et al. 1994, Drazen et al. 1998), indicating that these groups utilize fresh phytodetritus. The conversion of phytodetritus by benthos is a key to understanding the carbon balance on the deep-sea floor.

Since benthic foraminifera constitute a large part of the biomass of the benthic community, especially in water depths greater than 1000 m (Heip et al. 2001), they have been thought to play a major role in carbon consumption from the short to long term in the surface sediments of the deep sea (Moodley et al. 2002, Witte et al. 2003a). Deep sea benthic foraminifera respond to

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elevated organic carbon content after algal blooms at the oceanic surface. Many studies have reported that benthic foraminiferal assemblages strongly correlate with surface ocean productivity and subsequent changes in organic carbon flux (e.g. Corliss & Emerson 1990, Gooday & Turley 1990, Pfannkuche 1993, Silva et al. 1996, Ohga & Kitazato 1997, Schmiedl et al. 1997, Drazen et al. 1998, Gooday & Rathburn 1999, Altenbach et al. 1999, Kitazato et al. 2000, Fontanier et al. 2002, 2003). It is evident that phytodetritus forms a suitable foraminiferal food source and affects foraminiferal fauna. On the other hand, in contrast to numerous investigations of faunal and abundance changes among benthic foraminifera, feeding ecologies in relation to phytodetritus processing are not well understood. Altenbach (1992) conducted a shipboard feeding experiment using benthic foraminifera collected from a water depth of 1240 m in the Norwegian Sea. Based on the temporal changes in foraminiferal biomass after food addition, it was found that the foraminiferal biomass of fed individuals had increased nearly 2-fold compared with that of unfed individuals 3 d after food supply. Linke et al. (1995) also reported the activation of benthic foraminiferal metabolism such as respiratory activity and ATP content after the foraminifers were fed. However, quantitative information on the ingestion rate of organic carbon by benthic foraminifera has remained scarce.

Foraminiferal responses to the input of organic matter should provide crucial information on the carbon budget of deep-sea sediments. Benthic foraminiferal feeding behavior is also an important consideration when reconstructing surface productivity using benthic foraminiferal assemblages (Loubere & Fariduddin 1999). Recently, ^{13}C-labeled feeding experiments have demonstrated the rapid ingestion of phytodetritus by benthic macrofauna, meiofauna, and bacteria (Blair et al. 1996, Levin et al. 1999, Middelburg et al. 2000, Moodley et al. 2002, 2005, Witte et al. 2003a,b, Nomaki et al. 2005a). Nomaki et al. (2005a) reported that benthic foraminifera showed a high assimilation rate of ^{13}C-labeled algae relative to metazoan meiobenthos. Most benthic foraminiferal species ingested notable amounts of labeled algae during 6 d incubation, suggesting that phytodetritus is a major food source for foraminifera. However, 2 of 8 tested species did not assimilate significant amounts of fresh algae, even though the labeled algae were potentially available to them. They must, therefore, require carbon sources other than phytodetritus.

Various types of feeding ecologies and food sources for benthic foraminifera other than phytodetritus were reported in previous studies (Lee et al. 1966, Lee & Muller 1973, Lee 1974, DeLaca et al. 1981, Lipps 1983, Goldstein & Corliss 1994, Goldstein 1999). DeLaca et al. (1981) and DeLaca (1982) reported that agglutinated foraminifera from Antarctica can utilize dissolved organic carbon (DOC) directly. Carnivory, omnivory, suspension feeding, scavenging, symbiotism, parasitism, cannibalism, and deposit feeding are also reported forms of benthic foraminiferal feeding (Lipps 1983, Goldstein 1999). In deep-sea settings, bacteria are one suitable alternative and/or complementary nutrition source to phytodetritus. Based on ^{32}P and ^{14}C tracer feeding experiments, Lee et al. (1966) found that most tested littoral foraminiferal species preferentially ingested certain food such as diatoms, flagellates, and bacteria. It has also been reported that some littoral benthic foraminifera require bacteria as essential reproductive energy sources (Muller & Lee 1969). Ingestion of bacteria was also shown for certain benthic foraminiferal species (Lee & Muller 1973, Bernhard & Bowser 1992). Benthic foraminifera from deep-sea areas are also expected to ingest bacteria from the sediments. Goldstein & Corliss (1994) found bacteria in the food vacuoles of 2 deep-sea benthic foraminiferal species.

Bacteria are able to utilize DOC and establish high levels of biomass in the deep-sea sediment from Sagami Bay, Japan (Shimanaga & Shirayama 2000). This can be an important carbon source in addition to phytodetritus settled from the water column. If deep-sea benthic foraminifera utilize bacteria as a carbon source in amounts comparable to those of phytodetritus, the link from DOC via bacteria and foraminifera to metazoan taxa must be an important path for the carbon cycle in deep-sea sediments. However, in *in situ* ^{13}C-labeled studies, only the ingestion of algal materials by deep-sea benthic foraminifera has been previously investigated.

In this study, the variability of organic carbon utilization by deep-sea benthic foraminifera was investigated based on an *in situ* feeding experiment using 3 types of ^{13}C-labeled food materials from the deep-sea floor in Sagami Bay. We chose the marine diatom *Chaetoceros sociale* and green algae *Dunaliella tertiolecta* as model organisms of phytodetritus. To investigate bacterial ingestion by benthic foraminifera, we also used the marine bacteria *Vibrio alginolyticus*. After incubation for 2 and 21 d, assimilation rates of these 2 algal and 1 bacterial species by benthic foraminifera were determined at species level. Based on these data, we were able to discuss the food source preferences of the benthic foraminifera investigated.

MATERIALS AND METHODS

^{13}C-labeled food materials. Three types of ^{13}C-labeled food materials were prepared for the *in situ* feeding experiment (Table 1). The unicellular algae *Dunaliella tertiolecta*, belonging to Chlorophyceae, was prepared as a representative of phytoplankton to facilitate compar-

isons with results of our previous studies (Nomaki et al. 2005a). The fast and active response of foraminifera to *Dunaliella* tertiolecta spp. has been shown in laboratory and *in situ* experiments in Sagami Bay (Kitazato et al. 2003a, Nomaki et al. 2005a,b) and in Mediterranean Sea assemblages (Heinz et al. 2002). Lee et al. (1966) also reported that littoral benthic foraminifera showed a strong preference for *Dunaliella parva*. Thus, *Dunaliella* spp. are clearly suitable food sources for benthic foraminifera. The marine diatom *Chaetoceros sociale*, belonging to Bacillariophyceae, was also prepared as an important representative of the common natural diatom community in Sagami Bay (Kanda et al. 2003). Since *D. tertiolecta* is not distributed in Sagami Bay, it is important to make comparisons between *Chaetoceros* and *Dunaliella*. According to Lee et al. (1966), diatoms were the most preferred food source for some foraminiferal species, closely followed by *D. parva*.

Vibrio alginolyticus was prepared as a representative of bacteria in the sediments. *Vibrio* sp. were chosen for the experiment because they are universally distributed in seawater and sediments, and are also found in the sediment of Sagami Bay (Urakawa et al. 1999). Benthic foraminifera from shallow water ingested this bacterium in amounts similar to those of *Dunaliella tertiolecta* under laboratory culture conditions (K. Larkin unpubl. data). Hence, benthic foraminifera potentially utilize this bacterium as a food source.

Production of ^{13}C-labeled algae and bacteria. *Dunaliella tertiolecta* used in this study was originally provided by the Institute of Biology, Tuebingen University, Germany. *Chaetoceros sociale* was provided by the Microbial Culture Collection, National Institute for Environmental Studies, Japan (strain no. NIES-377). Both algae were incubated at the Micropaleontological Laboratory of Shizuoka University at 20°C with sterilized seawater containing f/2 medium and approximately 0.1 mM $NaHCO_3$ enriched in ^{13}C (99.9%; Shoko Co.). The final concentration of ^{13}C in the algal carbon was 4.571 ± 0.047 atom% in *D. tertiolecta*, and 6.820 ± 0.072 atom% in *C. sociale* (Table 1). The algae were separated from the cultured seawater after centrifugation, and were rinsed with sterilized seawater and stored at –20°C until the feeding experiment.

Vibrio alginolyticus 138-2 that had been maintained in the Microbiology Laboratory, Ocean Research Institute at the University of Tokyo, was precultured in 1/4-ZoBell 2216E culture medium at 27°C overnight. This medium contained polypeptone (1.25 g l^{-1}) and yeast extract (0.25 g l^{-1}) as organic substrates with artificial seawater, which was composed of the following compounds (l^{-1} of distilled water [pH 7.5]): NaCl, 23.4 g; KCl, 0.8 g; $MgSO_4 \cdot 7H_2O$, 4 g; $CaCl_2$, 0.2 g; KBr, 100 mg; $SrCl_2 \cdot 6H_2O$, 26 mg; and H_3BO_3, 20 mg (Kogure et al. 1998). The precultured cells were separated by centrifugation and washed twice with artificial seawater to remove the 1/4-ZoBell medium. They were then transferred to artificial seawater composed of: glucose, 1.875 g; ^{13}C-labeled glucose (98.7%; Shoko Co.), 1.25 g; NH_4Cl, 15 mg; and Na_2HPO_4, 1.5 mg l^{-1}. After incubation at 25°C for 20 h, the centrifuged cells were washed twice with artificial seawater to remove glucose and ^{13}C-enriched inorganic carbon. They were stored at –80°C prior to the *in situ* feeding experiment. The final ^{13}C concentration of organic carbon in *V. alginolyticus* was 2.003 ± 0.026 atom% (Table 1). Because the biomass of precultured *V. alginolyticus* was large relative to the ^{13}C-labeled glucose, the final ^{13}C concentration was reduced from the proportion of ^{13}C-labeled to that of non-^{13}C-labeled glucose added to the artificial seawater.

Table 1. Different ^{13}C-labeled food materials used in this experiment

Food material	Added carbon (g C m^{-2})	^{13}C atom %
Dunaliella tertiolecta	0.431	4.571 ± 0.047
Chaetoceros sociale	0.211	6.820 ± 0.072
Vibrio alginolyticus	12.3	2.003 ± 0.026

Procedure for the *in situ* feeding experiment. A feeding experiment was carried out from October 14 to November 5, 2002, during the R/V 'Natsushima' NT02-10 training cruises. In Sagami Bay, the spring bloom occurs from mid-February to May, when the chlorophyll concentration is greater than 70 mg m^{-2} in the integrated upper 50 m of the water column (Kanda et al. 2003). Elevated fluxes of organic carbon (200–500 mg C m^{-2} d^{-1}) and pheopigment were subsequently observed in the sediment trap moored 20 m above the seafloor (Kitazato et al. 2000, 2003b, Nakatsuka et al. 2003). Our feeding experiment was conducted in a season when the amount of fresh phytodetritus should have been very small on the seafloor. In our previous experiments (Nomaki et al. 2005a), 2 benthic foraminiferal species may have exhibited seasonal variation in the assimilation of labeled algae, but the feeding patterns of foraminiferal species were stable between seasons.

On October 14, 6 culture cores (5 cm i.d.; Fig. 1) were placed on the seafloor (water depth 1450 m) using a manipulator arm of the manned submersible 'Shinkai 2000'. Each culture core was equipped with 10 ml of ^{13}C-labeled food materials that contained various amounts of organic carbon (Table 1). Two of 6 culture cores were equipped with 96" *Dunaliella tertiolecta* (D), 2 cores with *Chaetoceros sociale* (C), and the remaining 2 cores with *Vibrio alginolyticus* (V). After positioning the culture cores, ^{13}C-labeled food materials were introduced to the surface sediments. Immedi-

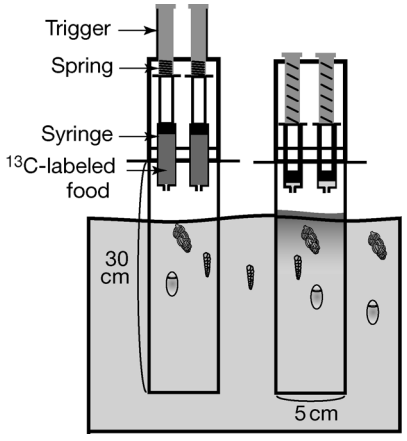

Fig. 1. Illustration of *in situ* feeding experiments. Each culture core had 2 syringes containing 5 ml of ^{13}C-labeled food. By removing triggers with the manipulator, ^{13}C-labeled food was injected into the core

ately after the injection of the food materials, one of each of these culture cores (referred to as D-0, C-0, and V-0, respectively) was recovered using the manipulator. They were sampled as 'time 0' controls. The other 3 cores (D-2, C-2, and V-2, respectively) were recovered on October 16, 2 d after the start of the experiment. On the same day, 3 additional cores with 3 different food materials were placed on the seafloor in the same area and then recovered after 21 d (D-21, C-21, and V-21, respectively). On October 14, 2 push-core samples (4.2 cm i.d.) were taken at the same station as a reference for the natural abundance, distribution, and carbon isotopic compositions of benthos and bulk sediments (B1 and B2, respectively). To test the 3 food materials in various time series in a limited payload, no replicate core was taken except for the natural background cores.

Sample preparation. The recovered culture cores were immediately sliced at 1 cm intervals from 0 to 5 cm in depth. Subsamples (0.5 cm^3) from the sliced sediments were used for the analysis of ^{13}C concentrations in the bulk sediments. These subsamples were kept frozen at –20°C until analysis. The remaining sediments were used for the determination of elemental and carbon isotopic compositions in benthic organisms. They were sieved on a 63 µm mesh with artificial seawater and then stored at –20°C until benthic organisms were removed. Metazoan meiofauna that had sufficient biomass for measurement (copepods, polychaetes, bivalves) and all foraminifera were removed from the sediment residues under a binocular microscope. Individual foraminifera in which the test cavity was filled with definite cytoplasm were selected and sorted into species. Eight common foraminiferal species at this site (Ohga & Kitazato 1997, Nomaki et al. 2005a)—*Cyclammina cancellata* (epifaunal to shallow infaunal species), *Bulimina aculeata*, *Uvigerina akitaensis*, *Bolivina spissa* (shallow infaunal species), *Bolivina pacifica*, *Textularia kattegatensis* (intermediate infaunal species), *Globobulimina affinis*, and *Chilostomella ovoidea* (deep infaunal species)—were analyzed at species level, and the remaining species were combined as 'others.' They were rinsed with artificial seawater to remove sediment particles attached to the test. These samples were kept frozen at –20°C until analysis.

Measurements of total organic carbon and carbon isotopic composition. Dried samples of both sediments and organisms were decalcified with 6 M HCl in silver cups. Total organic carbon (TOC) concentrations were determined using an elemental analyzer (EA1110, CE Instrument or FlashEA1112, ThermoQuest). Carbon isotopic compositions were determined using an isotope ratio monitoring mass spectrometer (Thermo Finnigan Delta plus or Delta plus XP). They were shown as δ-notations against the Vienna Pee Dee Belemnite standard ($\delta^{13}C = [(^{13}C/^{12}C)_{sample}/(^{13}C/^{12}C)_{PDB} - 1] \times 1000$) (Tables 2 & 3). Blank cup data were subtracted from the sample data for each measurement run.

To estimate the assimilation rate of ^{13}C-labeled food by benthic foraminifera, we calculated the fraction of carbon originating from added food material in the TOC (f) (Middelburg et al. 2000). The ^{13}C abundance in samples, food, and natural background were correlated with the following equation:

$$\phi_{sample} = f\phi_{food} + (1 - f)\phi_{bkgd} \; (0 \leq f \leq 1)$$

where ϕ_{sample}, ϕ_{food}, and ϕ_{bkgd} represent atomic fractions of ^{13}C (i.e. $^{13}C[^{12}C + {}^{13}C]^{-1}$) of sample, ^{13}C-labeled food, and natural background, respectively. Hence, the term f, the fraction of carbon originating from ^{13}C-labeled food) can be calculated using the following equation:

$$f^d_{food\ (D,C,V)} = (\phi_{sample} - \phi_{bkgd})\,(\phi_{food} - \phi_{bkgd})^{-1}$$

where f^d_{food} represents the fraction of carbon originating from ^{13}C-labeled food (D, *Dunaliella tertiolecta*; C, *Chaetoceros sociale*; and V, *Vibrio alginolyticus*) in a sample incubated for d days. Using this notation, we directly compared the foraminiferal food assimilation, even though the atomic fraction of ^{13}C was different among food materials. Since the value of f was small, we used $f \times 100$ (%). The carbon isotopic composition ($\delta^{13}C$) of benthic foraminifera and bulk sediments varied by about ± 2.5‰ and ± 0.3‰, respectively (Tables 2 & 3), within the background core samples. This potentially caused variation in f_{foram} of 0.08%, 0.05%, and 0.30%, and in f_{sed} of 0.01%, <0.01%, and 0.04% in cores D, C, and V, respectively.

RESULTS

Labeled carbon in incubation cores

The vertical distribution of added food materials varied greatly among incubation cores (Table 2, Fig. 2). The food materials were found in deep layers even in cores D-0 and V-0, suggesting that some disturbance had occurred. These transports were probably caused by both bioturbation by macrofauna and disturbances during core processing; be- cause previous experiments did not record such a high degree of disturbance in Sagami Bay (e.g. Kitazato et al. 2003a, Nomaki et al. 2005a), this was likely to be due to artificial disturbance during food injection and/or core recovery.

Added carbon was largely removed from the surface 5 cm sediments after 21 d incubation (Fig. 3). In cores D-2 and D-21, 1.5% and 16% (respectively) of added carbon was recovered from the sediments, whereas 80% was recovered at time 0 (D-0). In core C-0, only 8.6% of the added carbon was recovered from sediments. However, in core C-2, 96% of the added carbon was present in the surface 5 cm of sediments, which decreased to 7.5% after 21 d incubation. In cores V-0 and V-2, 88 and 100% of the added carbon was recovered but this drastically decreased in core V-21.

Added carbon concentrations in foraminiferal cells

In the time 0 control cores, all cellular carbon isotopic compositions of the foraminiferal species were within the range of those from the background cores (Table 3). We present data after 2 and 21 d of incubation in the following results and discussion. Since gradual mixing of food materials (e.g. Nomaki et al. 2005a) did not occur in this study, we examined the following results as integrated data of sediment depths from 0 to 5 cm. Values of f (mean ± SD) were calculated for the weighted average biomass at each depth.

Table 2. Carbon isotopic compositions ($\delta^{13}C$; % PDB) of sedimentary TOC; nd: not determined

Core name	0–0.5 cm	0.5–1 cm	1–1.5 cm	1.5–2 cm	2–3 cm	3–4 cm	4–5 cm
Dunaliella tertiolecta							
D-0	–6.5	–18.7	–17.5	–16.6	–18.2	–18.0	–15.3
D-2	–20.5	–20.8	–20.8	–21.1	–21.4	–21.4	–21.9
D-21	–17.9	–18.2	–21.4	–21.8	–20.6	–21.0	–19.6
Chaetoceros sociale							
C-0	–20.7	–21.4	–21.7	–21.8	–21.0	–21.4	–19.6
C-2	–15.5	–15.4	–15.4	–15.6	–17.8	–17.7	–16.1
C-21	–22.0	–19.3	–21.0	–20.6	–20.0	–21.4	–21.4
Vibrio alginolyticus							
V-0	93.9	61.9	44.7	–8.4	1.0	–8.2	–1.1
V-2	–11.8	–7.4	–15.8	3.3	1.0	80.0	13.8
V-21	9.6	–2.6	–0.6	–13.5	8.2	–13.9	–20.3
Background	–21.4	nd	nd	–20.7	–20.7	–20.7	–21.0

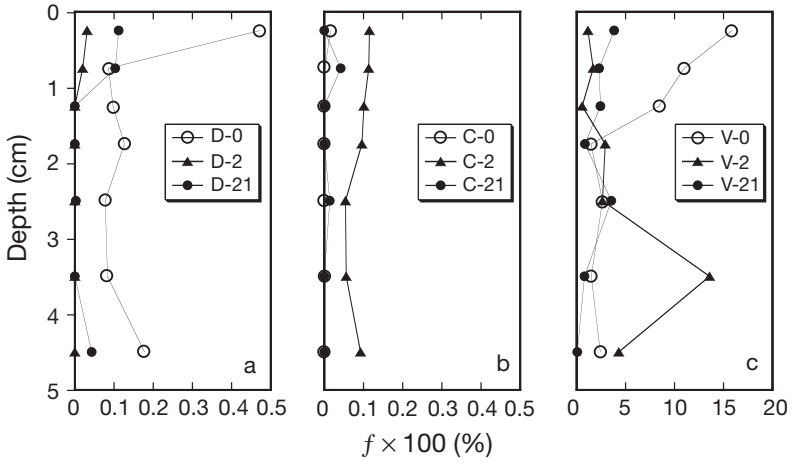

Fig. 2. Depth distribution of ^{13}C-labeled food concentration f within the sediment. (a) *Dunaliella tertiolecta* cores, (b) *Chaetoceros sociale* cores, (c) *Vibrio alginolyticus* cores

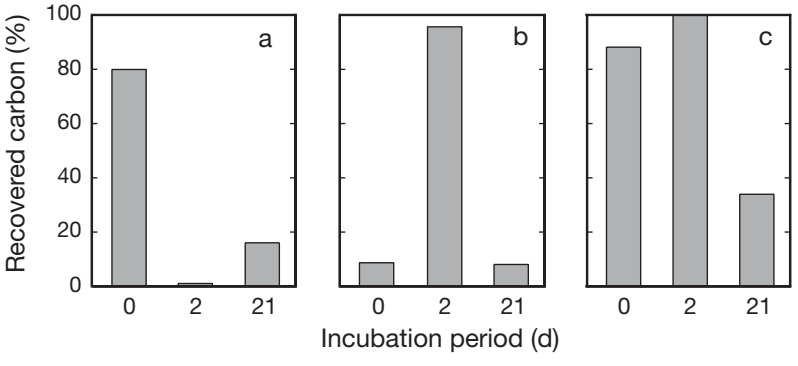

Fig. 3. Recovery of added carbon from surface 5 cm sediment. (a) *Dunaliella tertiolecta* cores, (b) *Chaetoceros sociale* cores, (c) *Vibrio alginolyticus* cores

Dunaliella tertiolecta and *Chaetoceros sociale* cores

Although sample numbers were small for the shallow infaunal species, similar trends were observed for *Dunaliella tertiolecta* and *Chaetoceros sociale* cores (Fig. 4). *Bulimina aculeata* exhibited the highest added carbon concentration (f^2_C) of 9.6%. It also had the highest carbon f^{21}_D value of 13.0%. Another shallow infaunal species, *Uvigerina akitaensis*, had moderate values of 1.0% for f^2_C and 2.9% for f^{21}_D, which were smaller than those of *B. aculeata*. Two intermediate infaunal species, *Bolivina pacifica* and *Textularia kattegatensis*, showed retarded reactions to algal supply in comparison with the above 2 shallow infaunal species. They recorded negligible (<0.1%) ingestion in the 2 d incubation. However, *B. pacifica* showed high a f^{21}_D value of 8.8% (no sample in C-21). *Textularia kattegatensis* also exhibited a high f^{21}_D value of 2.0%, although the f^{21}_C value was only 0.1%. *Globobulimina affinis*, a deep infaunal species, showed the highest or second highest f values (f^{21}_C = 7.3 ± 0.5%, f^{21}_D = 9.3 ± 3.9%) of all measured species after 21 d incubation, although this species had relatively low f values (f^{21}_C = 0.3 ± 0.2%, f^2_D = 0.2%) after 2 d incubation. Another deep infaunal species *Chilostomella ovoidea* and the shallow infaunal species *Cyclammina cancellata* showed very low f values throughout the entire experimental period. *Chilostomella ovoidea* did not assimilate added carbon, and had an apparent carbon content of less than 0.1% in cores C-2 and D-2. This species also exhibited low f values after the 21 d incubation, with an f^{21}_C value of 0.4 ± 0.2% and f^{21}_D value of 0.1 ± 0.0%. *Cyclammina cancellata* also showed little ingestion of fresh algae (f^{21}_C = 0.0%, f^{21}_D = 0.2%). These different response patterns between species confirmed results from our previous *in situ* experiments with *Dunaliella tertiolecta* as a food source (Nomaki et al. 2005a).

Bacterial cores

Most foraminiferal species assimilated no or only a small amount of ^{13}C-labeled bacteria in comparison with algae (Fig. 4). In the V-2 core, all examined species (*Uvigerina akitaensis*, *Textularia kattegatensis*, *Globobulimina affinis*, and *Chilostomella ovoidea*) showed

Table 3. Carbon isotopic compositions (δ^{13}C; % PDB) of benthic foraminiferal species collected from sediment cores of *in situ* feeding experiment

Species	Depth (cm)	B1	B2	D-0	D-2	D-21	C-0	C-2	C-21	V-0	V-2	V-21
Cyclammina cancellata	0–1	–20.0	–19.6			–14.6			–18.6			
	1–2			–17.3								–12.8
	2–3			–18.7								
Uvigerina akitaensis	0–1	–21.9	–23.0	–23.3	–22.7		–23.6	12.7		–22.5		–19.7
	1–2			–23.5	69.6	78.4	–23.1	26.8		–21.4		–19.8
	2–3					38.6					–21.1	
Bulimina aculeata	0–1	–21.1	–23.8	–28.2								3.3
	1–2		–18.9					487.4		–19.5		
	2–3					396.6						
Bolivina spissa	0–1		–27.8	–27.2								
	1–2	–23.3		–25.4				–25.5		–22.5		–23.1
	2–3					41.2						
Bolivina pacifica	0–1			–20.9						–24.0		
	1–2		–17.9	–17.9		268.1	–21.8			–19.5		–19.2
	2–3		–16.6				–15.7	–25.0				
	3–4		–17.5									
Textularia kattegatensis	0–1			–17.5	–15.5	80.2				–19.0	–22.3	–19.3
	1–2	–23.5	–17.2	–17.0	–16.0	7.2	–19.1	–19.0	7.4	–18.5	–22.6	–10.1
	2–3		–18.2	–18.1			–17.3	–21.7		–15.6	–20.3	–7.2
	3–4		–17.5	–16.8						–21.2	–15.4	
	4–5		–20.7	–22.9			–19.6			–19.8		
Globobulimina affinis	0–1				–20.0	484.9			363.8	–21.1	–21.9	–9.5
	1–2		–25.7	–26.7	–9.7	234.9	–24.2	–15.7	375.8	–20.0	–20.1	–11.2
	2–3		–23.6	–26.6	–10.3	234.1	–23.5	–17.6		–21.5	–23.5	–8.2
	3–4	–24.1		–24.4	–23.1	234.7	–25.0	–3.1	316.4	–22.2	–20.7	–8.1
	4–5	–24.8	–26.5	–25.1		507.1	–23.6	1.9		–22.8	–22.6	
Chilostomella ovoidea	0–1				–20.6					–19.7		
	1–2	–30.1		–20.1	–18.5	–18.0		–21.9		–19.5	–20.8	–8.4
	2–3		–23.9	–21.8	–25.4	–17.2	–24.3	–20.9	–4.5	–20.1	–20.5	–20.1
	3–4	–22.5	–25.4	–22.1	–20.0	–20.0	–23.9	–20.0	–18.2	–20.0	–19.8	
	4–5		–24.1	–21.6	–20.9		–20.8			–19.5	–20.6	

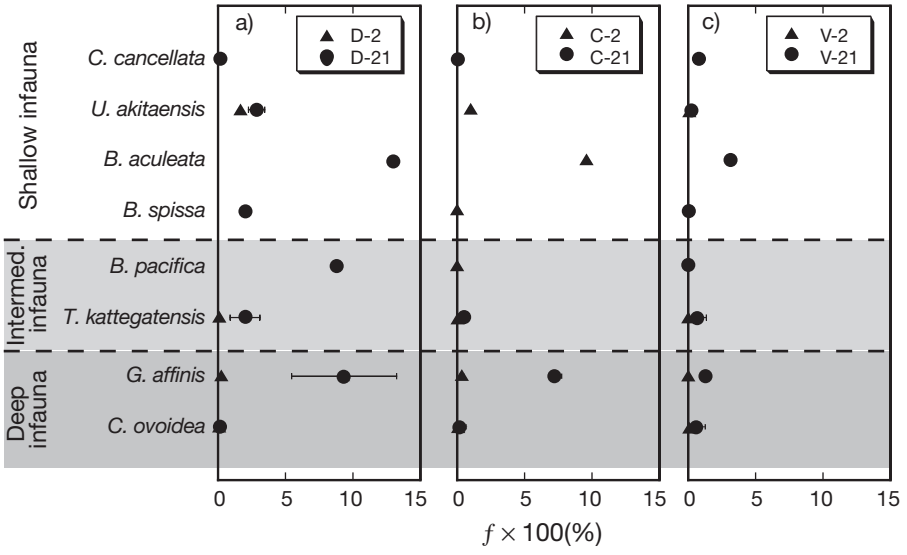

Fig. 4. Added carbon concentration (f^d_{food}) in the biomass of 8 foraminiferal species. (a) *Dunaliella tertiolecta* cores, (b) *Chaetoceros sociale* cores, (c) *Vibrio alginolyticus* cores. Refer to Table 3 for genus names

f values as low as 0.0 to 0.1%, which was in the range of background cores (≤0.3%). In core V-21, *U. akitaensis*, *Bolivina spissa*, and *B. pacifica* also had low $f^{21}{}_V$ values of 0.2 ± 0.1%, 0.0%, and 0.0%, respectively, even though they had elevated f^{21} values in algal cores D-21 and C-21. *Bulimina aculeata* exhibited the highest algal ingestion rate and had the highest f_V value (3.2%) of all tested foraminiferal species in core V-21. *Globobulimina affinis* also showed elevated f_V values (1.3 ± 0.1%) in the same core. Except for *C. ovoidea* and *Cyclammina cancellata*, all foraminiferal species exhibited low added carbon concentrations in *Vibrio alginolyticus* cores relative to those in algal cores, even though the labeled materials were present in sufficient amounts (Fig. 2). *Chilostomella ovoidea* and *C. cancellata* exhibited $f^{21}{}_V$ values (0.6% and 0.8%, respectively) somewhat higher than $f^{21}{}_D$ (0.1 ± 0.0% and 0.2%, respectively) and $f^{21}{}_C$ (0.4 ± 0.2% and 0.0%, respectively).

Response of metazoans to labeled materials

Copepods, polychaetes, and bivalves were examined in the isotopic measurements. Other metazoan animals were not found or were not found in sufficient biomass to facilitate measurements. Bivalvia was the only taxon that showed apparent ingestion of labeled materials among measured taxa in *Dunaliella tertiolecta* and *Chaetoceros sociale* cores (Table 4). Added carbon concentrations of copepods and polychaetes were all within the range of nonsignificant ingestion. Such low assimilation rates by metazoans were within the same range reported by Nomaki et al. (2005a). Although bivalves were not sampled from *Vibrio alginolyticus* cores, there were no metazoan samples that had assimilated significant amounts of labeled bacteria.

Table 4. Fractions of carbon from added food materials ($f \times 100$) in metazoans in experimental cores; nd: not determined; *significant ingestion of labeled materials

	V-0	V-2	C-2	D-21
Copepod	−0.1 ± 0.1	−0.1 ± 0.0	0.0 ± 0.0	0.0
Polychaete	nd	0.2	0.0	nd
Bivalve	nd	nd	0.1*	0.3*

DISCUSSION

Selective and random ingestion of labeled materials

We estimated the added carbon concentrations (f^d_{food}) in benthic foraminiferal cells to determine foraminiferal assimilation of ^{13}C-labeled food materials (Fig. 4). To understand foraminiferal feeding behavior, the added amounts of each food material must be considered in addition to the added ^{13}C concentration of each food material. For example, the f values of *Cyclammina cancellata* in the bacterial cores were higher than those in the algal cores after 21 d incubation. However, this was thought to be caused by the high labeled food concentrations in the sediment of *Vibrio alginolyticus* cores relative to algal cores (Fig. 2). To avoid this, we normalized the f^d_{food} value of foraminifera by dividing it by the f^d_{food} value of sedimentary organic matter (Fig. 5). If benthic foraminifera ingested the labeled materials selectively, the ratio of f^d_{food} to f^d_{sed} should be greater than 1 (selective ingestion), whereas if they ingested sediments at random, the ratio would correspond to unity (random ingestion). If they did not ingest any labeled materials, the ratio of f^d_{food} to

f^d_{sed} of foraminifera should be 0 (no ingestion). Here, the concentrations were normalized by the 21 d data, because 2 d data strongly reflected the response time (feeding velocity) in addition to the feeding ecology. Nomaki et al. (2005a) reported that the response time varied among 5 algae-feeding species during 2-, 4-, 6-, and 11 d incubations. After 11 d incubation, all the algae-feeding species exhibited an apparent isotopic signature for the ingestion of ^{13}C-labeled algae, while non-algae-feeding species did not. We therefore consider that a 21 d period is sufficiently long to neglect the influence of the response time.

Fig. 5 clearly indicates distinct selective ingestion of algae by *Uvigerina akitaensis*, *Bolivina spissa*, *B. pacifica*, *Bulimina aculeata*, *Textularia kattegatensis*, and *Globobulimina affinis*. On the other hand, no species showed selective ingestion of bacteria. *Bulimina aculeata*, *T. kattegatensis*, *G. affinis*, *C. ovoidea*, and *C. cancellata* ingested bacteria at random. *Bolivina spissa*, *B. pacifica*, and *U. akitaensis* did not ingest labeled bacteria. In this study, *Chilostomella ovoidea* and *Cyclammina cancellata* constantly showed no or random ingestion of both labeled algae and bacteria. We concluded that they ingest bulk sediments at random and thus had approximately the same level of labeled carbon as sedimentary organic matter.

Strictly speaking, this analysis of selective and random ingestion can only be achieved in a steady state, i.e. an environment where labeled materials are constantly supplied to the sediments. Because our experiment was not in the steady state, the interpretation of our results requires consideration of the factor of time, as mentioned above. Foraminiferal species selectively ingesting the labeled carbon but at a slow rate (i.e. small eaters) are also expected to have low f_{foram}/f_{sed} ratios in the early stage of the experiment. If we assume that the low ingestion of labeled food by *Chilostomella ovoidea* was due to a slow ingestion rate, this species' total carbon uptake would be 10- to 100-fold lower than that of selective species. However, in Sagami Bay, all the foraminiferal species were expected to have similar levels of carbon ingestion. We estimated the respiration rate of carbon for each benthic foraminiferal species by measuring oxygen consumption in the laboratory (H. Nomaki et al. unpubl. data). The respiration rate of carbon (i.e. carbon output per unit biomass) by *C. ovoidea* was about 4- to 5-fold lower than that of *Uvigerina akitaensis* and *Globobulimina affinis*, but not 10- to 100-fold lower. Although the carbon demand of *C. ovoidea* was somewhat lower, it must utilize carbon sources other than labeled materials. Thus, we concluded that *C. ovoidea* is not only a small eater, but a deposit feeder that utilizes sedimentary organic matter including both bacteria and algae.

In Sagami Bay, we recognized the following 3 types of benthic foraminifera in terms of feeding ecology. There were those that ingested (1) fresh phytodetritus alone (phytophagous species), (2) fresh phytodetritus selectively, but sedimentary organic matter as well when the phytodetritus was absent (seasonal-phytophagous species), and (3) sedimentary organic matter (deposit feeders) (Fig. 5). Such food preferences should affect the foraminiferal role in the degradation of

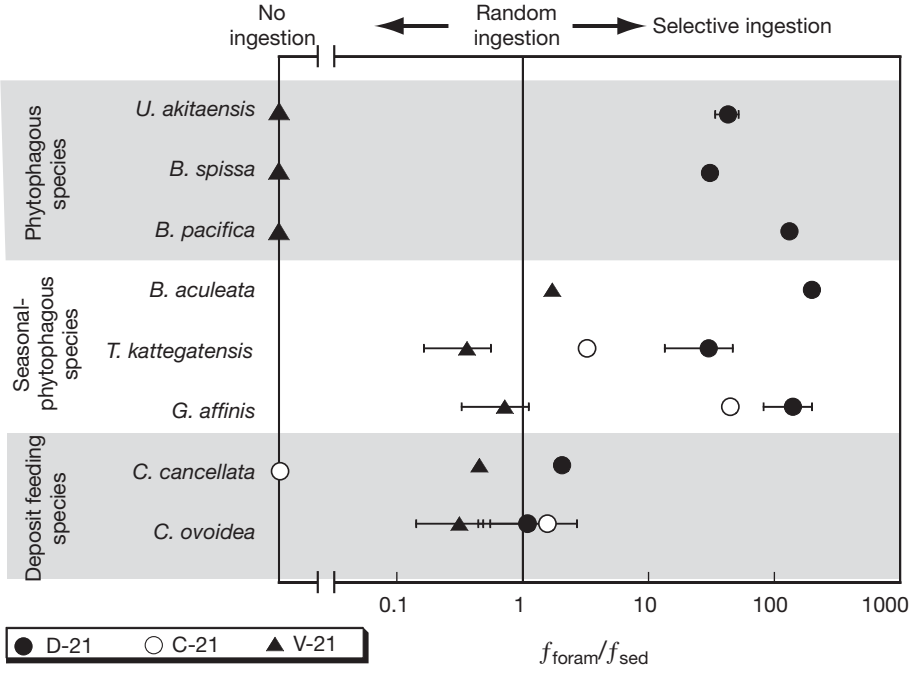

Fig. 5. Three types of feeding behavior of benthic foraminifera in Sagami Bay. Ratio of f_{foram} to f_{sed} represents preference for each food material by benthic foraminifera. Refer to Table 3 for genus names

organic matter in surface sediments. Since the phytophagous species *Uvigerina akitaensis*, *Bolivina spissa*, and *B. pacifica* exclusively ingest algal material settling from the water column, they could be partly responsible for early degradation of phytodetritus. This selective feeding on algal materials possibly contributes to age-dependent bioturbation by the rapid accumulation of phytodetritus, even though these effects may be much smaller than those caused by macrobenthic animals (Fornes et al. 1999). The seasonal-phytophagous species *Bulimina aculeata*, *Globobulimina affinis*, and *Textularia kattegatensis* also selectively ingest algal materials when phytodetritus exists. On the other hand, they also ingest sedimentary organic matter under conditions of limited algal material (summer to winter in Sagami Bay). Although deposit-feeding species like *Chilostomella ovoidea* and *Cyclammina cancellata* only ingest sedimentary organic matter at random, they have different microhabitats; *C. ovoidea* is part of the deep infauna, whereas *C. cancellata* is epifaunal to shallow infaunal. As a result, deposit-feeding species do not contribute to the degradation of newly supplied phytodetritus to the extent of other feeding types. They mainly utilize sedimentary organic matter already degraded by other foraminiferal species, heterotrophic bacteria, and metazoan animals in the sediments.

Our categorization of feeding types corresponds with results of ultrastructural observations (Goldstein & Corliss 1994), laboratory feeding experiments (Nomaki et al. 2005b), and long-term monitoring of foraminiferal densities on the deep-sea floor (Ohga & Kitazato 1997, Kitazato et al. 2000). Goldstein & Corliss (1994) observed cross-sections of the food vacuoles of *Globobulimina pacifica* and *Uvigerina peregrina*. They found that fragments of diatom frustules were common in *U. peregrina* but not in *G. pacifica*. They speculated that *U. peregrina* selectively ingested these materials, whereas *G. pacifica* did not. Although these species were different to those in our study and that of Goldstein & Corliss (1994), *Uvigerina* spp. are more likely to ingest algal materials.

Based on the results of laboratory feeding experiments, Nomaki et al. (2005b) observed ingestion of *Dunaliella tertiolecta* by all phytophagous and seasonal-phytophagous species, and upward migration to the sediment surface where *D. tertiolecta* was injected. However, in the case of deposit-feeding species, such ingestion or migration was not observed. They also reported that the numbers of *Bolivina pacifica* (phytophagous) and *Textularia kattegatensis* (seasonal-phytophagous) increased after the supply of *D. tertiolecta*, while *Chilostomella ovoidea* (deposit feeder) did not. Different types of feeding may affect both the abundance and vertical microhabitat of benthic foraminifera. Long-term monitoring of foraminiferal densities in deep-sea sediments (Ohga & Kitazato 1997) had also shown that phytophagous and seasonal-phytophagous species like *B. pacifica*, *T. kattegatensis*, and *Globobulimina affinis* exhibit seasonal fluctuation in their numbers, along with phytodetritus deposition. Individual numbers of *Bulimina aculeata*, *Uvigerina akitaensis*, and *Bolivina spissa* also tended to increase in spring, but not in all cases. This may have been caused by a patchy effect resulting from small sample numbers. In contrast, the abundance of *C. ovoidea* did not show any seasonal variations associated with phytodetritus deposition, although it was as abundant as *G. affinis* and *B. pacifica*. Deposition of fresh phytodetritus did not stimulate these deposit feeders, whereas phytophagous and seasonal-phytophagous species were strongly influenced by phytodetritus deposition in terms of their abundance and vertical distribution.

Effects of prolonged incubation

After 21 d, 16% (D-21), 8% (C-21), and 34% (V-21) of the added carbon remained as organic carbon in the surface 5 cm of the cores (Fig. 3). Although 21 d of incubation on the deep-sea floor is not long (Levin et al. 1999, Witte et al. 2003a), it was expected that some (or most) of the remaining labeled organic carbon was degraded or resynthesized during the 21 d incubation. Some previous ^{13}C-labeling experiments using algal materials found that certain parts of added algae were degraded after a few hours to days (Middelburg et al. 2000, Thomas & Blair 2002, Moodley et al. 2005). Degradation during incubation impedes the evaluation of what the heavy isotopic composition indicates: e.g. did benthic foraminifera ingest the labeled food itself? Although time lags in food assimilation within the same feeding type (e.g. *Uvigerina akitaensis* assimilates food more rapidly than *Bolivina spissa* among phytophagous species) probably reflect the response time and ingestion speed of each species, there is the additional possibility that they could reflect different pathways from food materials to foraminifera. Species that show slower ingestion might ingest carbon degraded or altered by other animals, while quick-response species may ingest labeled algae directly. Utilization of bacteria or DOC has been reported for benthic foraminifera (Lee et al. 1966, DeLaca et al. 1981, DeLaca 1982, Goldstein 1999). Slow-assimilation species might uptake DOC that was degraded from labeled food material, not from the food itself. They also might ingest 'newly labeled' organisms that had first ingested and assimilated labeled food materials.

If benthic foraminifera utilized the quickly degraded and mediated carbon in the surface sediments, then ben-

thic foraminifera in bacterial cores should show heavier carbon isotopic values than algal cores. Added excess ^{13}C in bacterial cores was nearly 10-fold greater than that in algal cores (D: 14.9, C: 12.0, V: 109.8 mg C; see also Fig. 2). However, in our experiment, most foraminifera showed lower labeled carbon concentrations in bacterial rather than algal cores. This indicates that most added carbon, in particular labeled bacteria, was not incorporated into benthic carbon pathways very quickly. Based on a laboratory experiment, Nagata et al. (1998) observed that only 20% of the disrupted cell membrane of *Vibrio alginolyticus* was degraded within 4 d at room temperature. This means that the degradation rate of an intact cell should be much slower than that of a disrupted cell. Moreover, the degradation rate may have been suppressed by the low temperature of our experiment on the deep-sea floor. *V. alginolyticus* added in our experiments were available as bacterial cells for the benthic foraminifera during the 21 d incubation. In the case of *Dunaliella tertiolecta* cores, species that had a heavy carbon isotopic composition (phytophagous and seasonal-phytophagous) commonly exhibited a green cytoplasmic color. This indicated the occurrence of at least some ingestion of algal chlorophyll-containing components that had not been processed by any other organism (Kitazato et al. 2003a).

Although some degradation and processing in the experimental cores could be expected, our results clearly indicated the fate of added organic carbon that had originated from food materials. Concentrations of labeled carbon in foraminifera were clearly different between algal and bacterial cores, and suggested that algal carbon was better assimilated by benthic communities than bacterial carbon, in both the short and long terms.

Assimilation of food materials

In this study, 2 algal species and 1 bacterial species were labeled and investigated as foraminiferal carbon sources. It has been reported that benthic foraminifera from shallow water show different ingestion rates of species of algae and bacteria (Lee et al. 1966). In our study, benthic foraminifera showed comparable responses to the addition of *Chaetoceros sociale* and *Dunaliella tertiolecta*.

Dunaliella tertiolecta was selectively ingested by dominant foraminiferal species, except *Cyclammina cancellata* and *Chilostemella ovoidea*. A similar trend was also observed for the addition of *Chaetoceros sociale*, a common natural diatom at the surface water of Sagami Bay (Kanda et al. 2003) (Figs. 4 & 5). However, a direct comparison of foraminiferal responses to the 2 algae is somewhat problematic, because foraminifera were unfortunately not found in sufficient densities in cores D-2 and C-21. Nevertheless, for a better comparison, we used data from cores C-2 and D-2 (present study) as well as data from Nomaki et al. (2005a) of November 2001 (Fig. 6), where sediments were incubated with *D. tertiolecta* for 2 d in a comparable *in situ* experiment (same station, same experimental procedure, same food material, and similar amount of added organic carbon). The foraminiferal responses to *D. tertiolecta* and *C. sociale* were comparable after the 2 d incubation, with the highest f values exhibited by *Bulimina aculeata* and relatively high f values by *Uvigerina akitaensis*, but low f values characterising other species. This was also found for the 4 species examined in core D-2 in this experiment. Similar foraminiferal responses to labeled food were also observed in cores D-21 and C-21 (Figs. 4 & 5). Therefore, we can conclude that foraminiferal responses to *D. tertiolecta* were similar to those to *C. sociale*, which is a natural diatom in this area, in both the short (2 d) and long (21 d) terms. These 2 algae belong to different classes: *D. tertiolecta* is a Chlorophyceae while *C. sociale* is a Bacillariophyceae. This may indicate that phytophagous and seasonal-phytophagous foraminifera can potentially utilize various types of algal materials.

Suhr et al. (2003) reported selective feeding on phytodetritus by benthic foraminifera based on lipid composition. In Sagami Bay, we also observed that phytol was abundant in the cells of *Uvigerina akitaensis* (phytophagous) and *Globobulimina affinis* (seasonal-phytophagous), whereas it was less abundant in the cells of *Chilostomella ovoidea* (deposit feeder) (unpubl. data). The concentrations of phytol in the cells of *U. akitaensis* and *G. affinis* were higher than those in the surrounding sediments. Phytol derives from the side chain of chlorophyll and is used as a biomarker for phytoplankton. Since phytol is contained in various phytoplankton materials, this observation supports our hypothesis that *C. ovoidea* does not ingest natural phytodetritus in large amounts, whereas *U. akitaensis* and *G. affinis* preferentially consume phytodetritus. Nakatsuka et al. (2003) reported that a significant amount of chlorophyll *a* (0.7 µg mg^{-1} dry sediment) was observed in a sediment trap moored 20 m above the seafloor during the spring bloom season at a station close to our site. This suggests that a portion of the phytoplankton is rapidly transported to the seafloor (1450 m in depth) before microbial degradation, and forms an important food source for benthic organisms. We observed that some benthic foraminiferal species quickly ingested similar amounts of fresh algal material. This indicated that benthic foraminifera react specifically to the signature of phytoplankton, and preferentially consume phytodetritus on the deep-sea floor.

We propose that the bacteria in this study were unselectively incorporated into food vacuoles and were

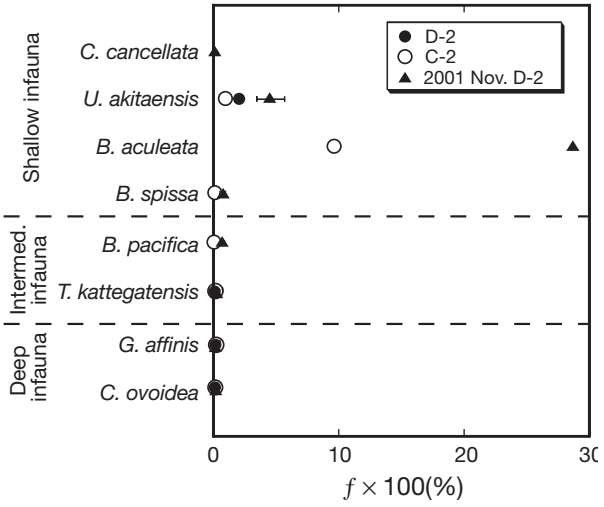

Fig. 6. Comparison of added carbon fractions (f^d_{food}) in benthic foraminifera among C-2 and D-2 (this experiment) and the 2 d incubation with *Dunaliella tertiolecta* as the food source in November, 2001 (Nomaki et al. 2005a). Refer to Table 3 for genus names

present as a specific carbon source. However, no benthic foraminiferal species showed selective ingestion of *Vibrio alginolyticus*, which was in contrast to selective ingestion of the 2 algal species (Fig. 5). *Chilostomella ovoidea* and *Cyclammina cancellata*, which exhibited nearly negligible assimilation of algae, ingested bacteria unselectively from the sediments. It has been speculated that these species, especially members of the deep infauna, could be dependent on anaerobic bacterial stocks (Kitazato et al. 2003a, Fontanier et al. 2003). We found that these species actually ingested bacteria, but that this was in association with other sediment particles rather than selectively from the sediments. Bacteria in food vacuoles were reported among deep-sea benthic foraminifera (Goldstein & Corliss 1994). At the same time, algae and sediment particles were also found inside foraminiferal food vacuoles. It was difficult to distinguish whether bacteria in food vacuoles were selectively incorporated or unselectively ingested with other sediment particles. In our study area, seasonal-phytophagous and deposit-feeding foraminifera unselectively ingested bacteria with sedimentary organic matter. It is also probable that the deep-sea benthic foraminifera examined by Goldstein & Corliss (1994) ingested bacteria associated with other sediment particles as a deposit-feeding strategy. Numerous deep-sea benthic foraminifera appear to have the potential to utilize bacteria as their trophic source. However, these deep-sea foraminiferal species feed on bacteria without selection, whereas some other species show a strong preference for algae.

Another explanation for the 'passive ingestion' of bacteria observed in this study is that the natural density of bacteria was sufficiently high in the sediment so that foraminiferal species did not need to selectively ingest newly added ^{13}C-labeled bacteria. In the laboratory, some investigators reported that benthic foraminifera ingest bacteria selectively (Lee et al. 1966, Lee & Muller 1973). These studies and another experiment (K. Larkin unpubl. data), which showed significant ingestion of *Vibrio alginolyticus* by *Ammonia beccarii*, were tested in an artificial environment where other food sources were absent. Their results may indicate potential bacterial assimilation by benthic foraminifera. Further investigations, such as direct observation of food vacuoles in nature, or lipid biomarker analysis, will hopefully elucidate the importance of bacteria as a foraminiferal food source.

Fate of added carbon

In algal cores, only a small portion of the injected carbon was recovered from sediments (Fig. 2). Generally, the recovery rate decreased over the incubation period. The low recovery in core C-0 immediately after the injection of labeled *Chaetoceros sociale* may be explained by the sedimentation process. Based on laboratory observations in a glass bottle, *C. sociale* has a slower deposition rate relative to that of *Dunaliella tertiolecta*. Most *C. sociale* cells may not have been deposited on the sediment surface or may have been resuspended during the recovery of core C-0, resulting in the low recovery of added carbon. After 2 d, the added *C. sociale* had settled and 96% of the added carbon was recovered. This time lag might explain the slow reactions to *C. sociale* by benthic foraminifera.

Low recovery rates of added carbon were also observed in the sedimentary organic matter from cores D-2, D-21, and C-21. Some of the loss may have been caused by transport to sediment layers deeper than 5 cm. The proportion of unrecovered carbon to added carbon in core D-2 (98.5%) was much greater than that in 2 previous experiments (60 to 70%) (Nomaki et al. 2005a). We observed rapid mixing during this experiment, which was not observed in the previous experiments; therefore, some of the rapid removal (30 to 40%) may have been due to transport to deeper sediment layers, as suggested by elevated δ^{13}C values at 5 cm sediment depth (Fig. 2). On the other hand, high concentrations of added carbon were observed in the phytophagous and seasonal-phytophagous foraminiferal species in these cores. This strongly suggests that ^{13}C-labeled carbon was rapidly ingested by benthic communities, including benthic foraminifera, and then respired as CO_2. As a result, only a minor fraction remained as 'sedimentary organic matter' in these cores, especially after longer incubation.

After 21 d, 84% (D-21) and 92% (C-21) of the added carbon was removed from the core. These decreases correspond to 10 to 18 mg C m^{-2} d^{-1} of carbon respiration for the first 21 d. Assuming that 40% of added carbon was transported to the layer deeper than 5 cm (as in core D-2), carbon respiration became 6 to 11 mg C m^{-2} d^{-1}. According to Moodley et al. (2002), supplied ^{13}C-labeled algal carbon is rapidly respired as CO_2 in comparison with sedimentary organic carbon. They reported that labeled carbon accounts for only 0.7% TOC in the sediments, but that 16% of total respired carbon originated from the labeled carbon. Unfortunately, we did not measure the sediment community oxygen consumption (SCOC) in this experiment. However, Yamaoka (2000) estimated a SCOC of about 100 to 400 mg C m^{-2} d^{-1} at this site. Respired labeled carbon composed 1.5 to 11% of total respiration, although the added algal carbon consisted of only 0.09 to 0.18% of the surface 5 cm sediment layer. We have estimated a benthic foraminiferal respiration rate of 45.6 mg C m^{-2} d^{-1} (H. Nomaki et al. unpubl. data), which corresponds to 11 to 46% of SCOC. Therefore, we conclude that the added algal carbon was preferentially ingested and respired by the benthic community, especially by benthic foraminifera.

In *Vibrio alginolyticus* cores, organic carbon was removed from the surface 5 cm of sediments at the rate of 390 mg C m^{-2} d^{-1} during the 21 d incubation. Assuming that the rate of transport to deeper layers was similar to those observed for cores C and D, the respiration rate was 234 to 273 mg C m^{-2} d^{-1}. This respiration rate by the total benthic community corresponds to approximately 60 to 70% of the SCOC value estimated by Yamaoka (2000), which implies that the benthic community exclusively utilizes labeled bacterial carbon and then respires it as CO_2. Benthic foraminifera and metazoan meiofauna showed nonselective ingestion of labeled bacteria. Bacterial activity may dominate this high respiration rate. We have observed high bacterial activity after addition of *Dunaliella tertiolecta* to sediments at the same station at Sagami Bay (unpubl. data). The abundance of some bacterial fatty acid biomarkers increased 2-fold under natural conditions, indicating that bacteria reproduced within 2 to 4 d. Although we did not measure the incorporation rate of labeled food by bacteria in this study, the respiration of labeled bacteria may be due to the bacterial community in the sediments.

Carbon assimilation rate of sedimentary organic carbon

Nomaki et al. (2005a) reported that benthic foraminifera assimilated carbon at rates of 1.0 ± 0.6 mg C m^{-2} d^{-1} in autumn, and 2.9 ± 2.4 mg C m^{-2} d^{-1} in spring. However, these rates were calculated for 'fresh algal carbon', not for total carbon. In this study, we observed that some species ingested sedimentary organic matter as well as fresh phytodetritus. Therefore, we should take into account the 'old' carbon source when discussing the benthic foraminiferal role in carbon budgets of surface sediments. By assuming that labeled bacteria and sedimentary organic carbon were proportionally incorporated by the seasonal-phytophagous and deposit-feeding foraminifera, the foraminiferal assimilation rate of sedimentary organic carbon can be estimated using the following equation:

$$R_{sed} = m(f^{21}{}_{V\,foram} \times f^{21}{}_{V\,sed})t^{-1}$$

where R_{sed} is the ingestion rate of sedimentary organic carbon (mg C m^{-2} d^{-1}), m is the biomass of benthic foraminifera in a unit area (mg C m^{-2}), and t is the incubation period (days). Estimated ingestion rates are shown in Table 5. We used data from core V-21 for this calculation because only 4 species were measured in core V-2, and no species assimilated significant amounts of bacteria in that time. However, if the incubation period exceeds foraminiferal turnover time, the ingestion rate of sedimentary organic carbon will be underestimated. This is probably true in the case of *Bulimina aculeata*, which showed almost identical values of $f^{21}{}_{V\,foram}$ relative to $f^{21}{}_{V\,sed}$. On the other hand, for all other species, $f^{21}{}_{V\,foram}$ never exceeded $f^{21}{}_{V\,sed}$, which suggests that carbon ingestion rates were not underestimated. One other possible source of underestimation of the carbon ingestion rate is carbon respiration. Ingested carbon can partly be respired as CO_2 over 21 d However, we have estimated the amount of respired carbon to be less than 10% (H. Nomaki et al. unpubl. data). Therefore, we did not take the respired carbon into account in the following discussion.

The highest sediment ingestion rate was calculated for the seasonal-phytophagous species *Globobulimina*

Table 5. Estimated ingestion rate of sedimentary organic carbon by benthic foraminifera

Species	Feeding type	Ingestion rate (mg C m^{-2} d^{-1})
Bulimina aculeata	Seasonal-phytophagous	0.3 ± 0.2
Textularia kattegatensis	Seasonal-phytophagous	0.2 ± 0.1
Globobulimina affinis	Seasonal-phytophagous	6.5 ± 7.8
Chilostomella ovoidea	Deposit feeder	0.3 ± 0.2
Cyclammina cancellata	Deposit feeder	1.0 ± 1.1
Others	–	0.4 ± 0.2
Total	–	8.8 ± 9.4

affinis (6.5 ± 7.8 mg C m^{-2} d^{-1}). The deposit feeder *Cyclammina cancellata* consumed 1.0 ± 1.1 mg C m^{-2} d^{-1} of sedimentary organic carbon. In total, benthic foraminifera ingested 8.8 ± 9.4 mg C m^{-2} d^{-1} of sedimentary organic carbon, while phytophagous and seasonal-phytophagous species selectively ingested 1.0 ± 0.6 to 2.9 ± 2.4 mg C m^{-2} d^{-1} of algal carbon. The benthic foraminifera processed more sedimentary organic carbon than algal carbon. While they make substantial contributions to the consumption of fresh phytodetritus (2.7% of TOC flux at this site), they also consume a large amount of 'old' carbon from the bulk sediments, even though the ingested carbon (8.8 ± 9.4 mg C m^{-2}) was negligible (0.0038%) relative to TOC in the surface 5 cm of sediment (230 g C m^{-2}).

CONCLUSIONS

Most deep-sea benthic foraminiferal species selectively ingested fresh algal material in this study, although some species did not. The ingestion patterns were comparable between *Chaetoceros sociale* and *Dunaliella tertiolecta* cores. Foraminiferal species such as *Uvigerina akitaensis*, *Bolivina spissa*, *B. pacifica*, *Textularia kattegatensis*, and *Globobulimina affinis* preferentially ingested phytodetritus transported from the surface ocean. In contrast, no benthic foraminiferal species selectively ingested bacteria in our experiment. Bacteria in the sediment are utilized by benthic foraminifera through passive ingestion with other sedimentary organic matter.

We identified the following 3 types of benthic foraminiferal feeding pattern in Sagami Bay: (1) selective ingestion of fresh phytodetritus (phytophagous species), (2) selective ingestion of fresh phytodetritus in addition to sedimentary organic matter (seasonal-phytophagous species), and (3) sedimentary organic matter at random (deposit feeders). These different types of feeding patterns should affect the role of foraminifera in organic matter degradation in surface sediments. These types are reflected by foraminiferal density and vertical distribution in the sediment column of the deep-sea.

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