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NOTE

Feeding by the heterotrophic dinoflagellate *Protoperidinium bipes* on the diatom *Skeletonema costatum*

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ABSTRACT: The heterotrophic dinoflagellate *Protoperidinium bipes* is a predominant heterotrophic dinoflagellate (maximum density = 137 cells ml^{-1}) during diatom blooms. To investigate its role as a grazer in the population dynamics of diatoms, we measured growth and ingestion rates of *P. bipes* when feeding on the common diatom *Skeletonema costatum*. We also calculated grazing coefficients by combining field data on abundances of *P. bipes* and co-occurring *S. costatum* with laboratory data on ingestion rates obtained in the present study. Specific growth rates of *P. bipes* increased continuously with increasing concentration of *S. costatum*. The maximum specific growth rate of *P. bipes* on *S. costatum* was 1.37 d^{-1} when data for the growth rate were fitted to a Michaelis-Menten equation. The threshold prey concentration (where net growth = 0) was 111 ng C ml^{-1} (4270 cells ml^{-1}). Maximum ingestion and clearance rates of *P. bipes* on this diatom were 2.9 ng C $grazer^{-1}$ d^{-1} (112 cells $grazer^{-1}$ d^{-1}) and 1.0 µl $grazer^{-1}$ h^{-1}, respectively. *P. bipes* exhibited the highest maximum swimming speed (ca. 8.3 mm s^{-1}) and maximum volume-specific clearance rate (5.4×10^6 h^{-1}) among *Protoperidinium* species so far reported. Calculated grazing coefficients by *P. bipes* on *S. costatum* (0.001 to 0.034 h^{-1}, i.e. 0.1 to 3.4 % of *S. costatum* populations were removed by a *P. bipes* population in 1 h) were much higher than those by co-occurring *Acartia* spp. (< 0.002 h^{-1}). The results of the present study suggest that *P. bipes* sometimes has a considerable grazing impact on populations of *S. costatum*.

KEY WORDS: Algal bloom · Growth · Grazing · Impact · Ingestion · Protist · *Arcartia* · Copepod

INTRODUCTION

Marine diatoms and heterotrophic protists are the major components of marine plankton communities. The abundance of heterotrophic protists often increases in the decline stage of diatom blooms (Kristiansen et al. 2001) and ingestion of diatom cells by heterotrophic protists has been observed (Jacobson & Anderson 1986, 1993, Hansen 1992, Strom & Buskey 1993, Buskey et al. 1994, Naustvoll 1998, Skovgaard & Hansen 2003). Thus, it has been suggested that grazing pressure by heterotrophic protists may sometimes play an important role in the population dynamics of diatoms (Jacobson & Anderson 1993).

Skeletonema costatum commonly dominates diatom abundance in coastal waters (Pratt 1965, Durbin & Durbin 1981, Reid et al. 1985, Marshall & Ranasinghe 1989, Ramaiah & Furuya 2002, Balkis 2003). While there are many studies on feeding by copepods on this diatom (i.e. Conover 1956, Martin 1965, Paffenhöfer 1976, Deason 1980), there are few studies on the growth of heterotrophic protists that graze on *S. costa-*

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tum (Hansen 1992), and no reports on ingestion rate and grazing impact. To understand the population dynamics of *S. costatum* and the total grazing pressure exerted by zooplankton on *S. costatum*, the grazing impact by co-occurring dominant heterotrophic protists on *S. costatum* should be explored.

The heterotrophic dinoflagellate *Protoperidinium bipes* is often abundant (reported maximum density = 137 cells ml^{-1}) in coastal waters when *Skeletonema costatum* is abundant (Jeong et al. 2000, Balkis 2003, Roberts et al. 2003). It has a wide distribution (Lessard & Swift 1986, Jacobson 1987, Nielsen et al. 1993, Bralewska & Witek 1995, Tiselius & Kuylendstierna 1996, Caroppo 2000, Tuschling et al. 2000, Johnson & Costello 2002, Lovejoy et al. 2002, Roberts et al. 2003), but its feeding habit is as yet unknown.

We established a monoclonal culture of *Protoperidinium bipes* and conducted experiments to examine its numerical and functional responses when grazing on *Skeletonema costatum* and measured its swimming speed. We also estimated grazing coefficients attributable to *P. bipes* on *S. costatum* using our data for ingestion rates obtained from the laboratory experiments and the abundances of predator and prey in the field. The results of the present study provide a basis for understanding the potential of *P. bipes* to influence the population dynamics of *S. costatum*.

MATERIALS AND METHODS

Culture of diatom prey. *Skeletonema costatum*, isolated from a coastal water off Kwangyang, Korea, were grown at 20°C in enriched f/2 seawater media (Guillard & Ryther 1962), under continuous illumination of 100 µE m^{-2} s^{-1} provided by cool white fluorescent lights. Only cultures in an exponential growth phase were used for the feeding experiments. The carbon content for *S. costatum* (0.026 ng C $cell^{-1}$) was estimated from cell volume (250 $µm^3$) according to Menden-Deuer & Lessard (2000).

Isolation and culture of *Protoperidinium bipes*. A 40 cm diameter, 20 µm mesh plankton net was used to collect samples from a coastal water off Kwangyang, Korea, during October 2003 when the water temperature and salinity were 17.6°C and 30.6 psu, respectively. The samples were screened gently through a 154 µm Nitex mesh and placed in 1 l polycarbonate (PC) bottles. The bottles were spiked with 50 ml of f/2 media, and *Skeletonema costatum* (density = ca. 40 000 cells ml^{-1}) were added as food. The bottles were placed on plankton wheels rotating at 0.9 rpm and incubated at 20°C under the continuous illumination of 20 µE m^{-2} s^{-1} of cool white fluorescent light. A week later, aliquots of the enriched water were transferred to 6-well tissue culture plates and a monoclonal culture of *Protoperidinium bipes* was established by 2 serial single cell isolations. Once dense cultures of *P. bipes* were obtained, they were transferred to 270 or 500 ml PC bottles of fresh *S. costatum* prey (density = ca. 50 000 cells ml^{-1}) every 3 d. Experiments were conducted when a large volume of *P. bipes* culture was available.

Cell volume. Cell length and maximum width of *Protoperidinium bipes* preserved in 5% acid Logol's solution were measured using a compound or inverted microscope at the beginning of the experiment and the end of each interval (n = 20 for each prey concentration). The shape of *P. bipes* was estimated to be 2 small cones (2 cones connected to each other in the bottom half of the cell: W shape) connected to a large cone (top half of the cell: Λ shape) at the cell equator (= maximum width of the cell). Cell volume of the preserved *P. bipes* was calculated according to the equation: volume = $1/3 \times [\pi\ (\text{cell width}/2)^2] \times [\text{cell length}/2] + 2 \times \{1/3 \times [\pi\ (\text{cell width}/4)^2] \times [\text{cell length}/2]\}$.

The carbon content for *Protoperidinium bipes* was estimated from cell volume according to Menden-Deuer & Lessard (2000).

Swimming speed. Swimming speeds of *Protoperidinium bipes* starved for 12 to 24 h were measured at 20°C using a video analyzing system. For each species, aliquots from a dense culture were added to multi-well plates and allowed to acclimate for 30 min. The video camera focused on 1 well (i.e. seen as 1 circle) in a multi-well plate and then swimming dinoflagellates were recorded at 40× magnification. The mean and maximum swimming velocities were analyzed for all swimming cells seen for the first 15 min. Average swimming speed was calculated based on the linear displacement of cells in 1 s during single-frame playback. Swimming speeds of 45 cells were measured.

Growth and ingestion rates. Experiments were designed to measure growth, ingestion, and clearance rates of *Protoperidinium bipes*, as a function of the prey concentration, when feeding on *Skeletonema costatum*.

Two days before these experiments were conducted, dense cultures of *Protoperidinium bipes* grazing on *Skeletonema costatum* were transferred into a 1 l PC bottle containing low concentrations of the target prey (ca. 5000 cells ml^{-1}). This was done to acclimate the predator to the target prey and minimize any possible residual growth resulting from the ingestion of prey during batch culture. The bottle was filled to capacity with filtered seawater and placed on a plankton wheel to incubate as above. A 10 ml aliquot was removed from the bottle and fixed with 5% Lugol's solution. The abundances of *P. bipes* and its prey were determined by enumerating cells in three 1 ml Sedgwick-Rafter counting chambers (SRCs).

Initial concentrations (cells ml^{-1}) of *Protoperidinium bipes* and the target prey were established using an autopipette to deliver predetermined volumes of known cell concentrations to the bottles. Triplicate 80 ml PC experiment bottles (mixtures of predator and prey) and triplicate control bottles (prey only) were set up at each predator-prey combination. Triplicate control bottles containing only *P. bipes* were also established at 1 predator concentration. Five ml of f/2 medium was added to all the bottles, which were then filled to capacity with freshly filtered seawater and capped. To determine the actual mean predator and prey densities (cells ml^{-1}) at the beginning of the experiment (6/94, 9/516, 11/990, 16/4997, 23/10590, 34/45240, 50/72150, 78/0) and after 24, 48, and 72 h incubation, 8 ml aliquots were removed from each bottle and fixed with 5% Lugol's solution, and all *P. bipes* cells and all or >200 prey cells in the three 1 ml SRCs were enumerated. Prior to taking subsamples, the condition of *P. bipes* and its prey was assessed under a dissecting microscope. The bottles were filled again to capacity with freshly filtered seawater, capped, and placed on the plankton wheel under the environmental conditions described above. The dilution of the cultures associated with refilling the bottles was taken into consideration in calculating growth and ingestion rates.

The specific growth rate of *Protoperidinium bipes* (μ, d^{-1}) was calculated by averaging the instantaneous growth rates (IGR) for each sampling interval, calculated as:

$$\mathrm{IGR} = \frac{\ln(S_{t_2} / S_{t_1})}{t_2 - t_1} \times 24 \quad (1)$$

where S_{t_1} and S_{t_2} = the concentration of *P. bipes* at consecutive samplings. The final t_2 for calculation was 48 h, which provided the highest specific growth rate. Mean prey concentrations for 48 h were also calculated by averaging the instantaneous mean prey concentrations at 0 to 24 h and at 24 to 48 h. The instantaneous mean prey concentration for each sampling interval was calculated using the equations of Frost (1972).

Data for *Protoperidinium bipes* growth rate were fitted to a Michaelis-Menten equation:

$$\mu = \frac{\mu_{\max}(x - x')}{K_{\mathrm{GR}} + (x - x')} \quad (2)$$

where μ_{max} = the maximum growth rate (d^{-1}); x = prey concentration (cells ml^{-1} or ng C ml^{-1}), x' = threshold prey concentration (the prey concentration where μ = 0), and K_{GR} = the prey concentration sustaining $^1/_2\ \mu_{max}$. Data were iteratively fitted to the model using DeltaGraph® (Delta Point).

Ingestion and clearance rates were calculated using the equations of Frost (1972) and Heinbokel (1978). The incubation time for calculating ingestion and clearance rates was the same as for estimating growth rate. Ingestion rate (IR) data were fitted to a Michaelis-Menten equation:

$$\mathrm{IR} = \frac{I_{\max}(x)}{K_{\mathrm{IR}} + (x)} \quad (3)$$

where I_{max} = the maximum ingestion rate (cells $grazer^{-1}\ d^{-1}$ or ng C $grazer^{-1}\ d^{-1}$); x = prey concentration (cells ml^{-1} or ng C ml^{-1}), and K_{IR} = the prey concentration sustaining $^1/_2\ I_{max}$.

Gross growth efficiency. Gross growth efficiency (GGE), defined as grazer biomass produced (+) or lost (–) per prey biomass ingested, as a function of mean prey concentration, was calculated from estimates of carbon content per cell based on cell volume at each mean prey concentration.

Grazing impact. With some assumptions (see Table 1), we estimated grazing coefficients attributable to *Protoperidinium bipes* feeding on *Skeletonema costatum* by combining field data on abundances of the grazer and the prey with ingestion rates of the grazer on the prey obtained in the present study. For comparison (of the grazing coefficients between *P. bipes* and *Acartia* spp. on *S. costatum*), we also estimated grazing coefficients attributable to co-occurring dominant copepods *Acartia* spp. feeding on *S. costatum* by combining field data on abundances of *Acartia* spp. and *S. costatum* with ingestion rates of the grazer on the prey obtained from the Michaelis-Menten equation in Fig. 4 of Deason (1980). The data on the abundances of *S. costatum*, *P. bipes*, and co-occurring *Acartia* spp. used in this estimation were obtained from water samples and net-towed samples collected from the coastal waters at the same stations off Kwangyang (1999–2001), Koheung (1997–1999), and Saemankeum (1999), Korea.

Grazing coefficients (g, h^{-1}) were calculated as:

$$g = (1/\Delta t)\ \{\ln[C_i/(C_i - C_e)]\} \quad (4)$$

where Δt (h) is a time interval, C_e (cells ml^{-1}) is the number of prey cells eaten by the *Protoperidinium bipes* or *Acartia* spp. population in 1 ml of seawater in 1 h, and C_i (cells ml^{-1}) is the initial prey cell concentration in a given hour. The values of C_e were calculated as:

$$C_e = \mathrm{PIR} \times 1\ \mathrm{h} = \mathrm{IR} \times G \times 1\ \mathrm{h} \quad (5)$$

where PIR is the population ingestion rate of *P. bipes* or *Acartia* spp. feeding on *S. costatum* in 1 ml of seawater (prey eaten $ml^{-1}\ h^{-1}$), IR is the ingestion rate (prey eaten $grazer^{-1}\ h^{-1}$) of *P. bipes* or *Acartia* spp. feeding on *S. costatum*, and G is the abundance (cells ml^{-1}) of *P. bipes* or *Acartia* spp. at the same time as C_i.

RESULTS

Swimming speed

The average (±SE, n = 45) and maximum swimming speeds of *Protoperidinium bipes* starved for 12 to 24 h were 4006 (±306) and 8269 µm s^{-1}, respectively. It was difficult to capture actively swimming *P. bipes* cells using a micropipette under a dissecting microscope.

Growth rates

The specific growth rates of *Protoperidinium bipes* feeding on a unialgal diet of *Skeletonema costatum* increased continuously with increasing mean prey concentration (Fig. 1). When the data were fitted to Eq. (2), the maximum specific growth rates (μ_{max}) of *P. bipes* were 1.37 d^{-1}. Threshold prey concentrations (where net growth = 0) were 111 ng C ml^{-1} (4270 cells ml^{-1}).

Ingestion and clearance rates

The ingestion rates of *Protoperidinium bipes* feeding on unialgal diets of *Skeletonema costatum* increased rapidly with increasing mean prey concentration below ca. 350 ng C ml^{-1} (13 460 cells ml^{-1}) and slowly, but continuously, increased at higher prey concentrations (Fig. 2). When the data were fitted to Eq. (3), the maximum ingestion rate of *P. bipes* was 2.9 ng C $grazer^{-1}$ d^{-1} (112 cells $grazer^{-1}$ d^{-1}).

The maximum clearance rate of *Protoperidinium bipes* was 1.0 µl $grazer^{-1}$ h^{-1}, which was measured at the lowest prey concentration. The maximum volume-specific clearance rate was 5.4×10^6 h^{-1} (1.0 µl $grazer^{-1}$ h^{-1} with a volume of 184 μm^3).

Cell volume

Cell volume of *Protoperidinium bipes* fed *Skeletonema costatum* after 48 h incubation did not change markedly with increasing mean prey concentrations between 3 and 17 ng C ml^{-1} (130 to 180 μm^3), but it increased rapidly up to 1160 μm^3 at mean prey concentrations between 17 and 183 ng C ml^{-1} and slowly at higher prey concentrations, reaching a maximum of 1430 μm^3 at a prey concentration of 1826 ng C ml^{-1} (Fig. 3). The cell volume of *P. bipes* without added prey for 48 h was 126 μm^3.

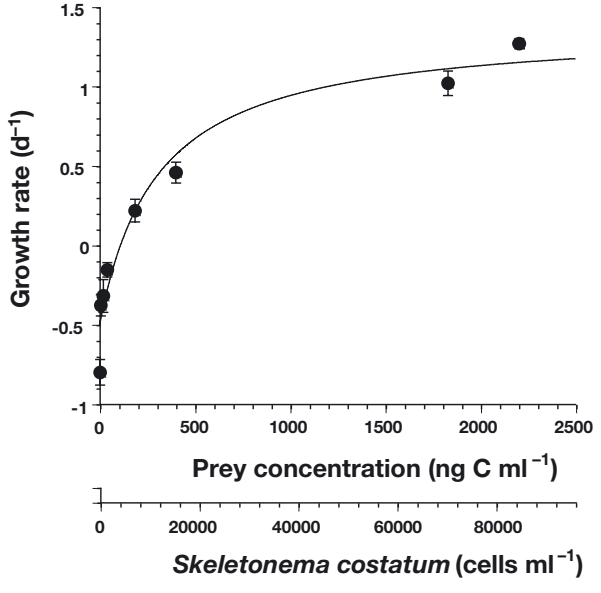

Fig. 1. Specific growth rates of *Protoperidinium bipes* feeding on *Skeletonema costatum* as a function of mean prey concentration (x, ng C ml^{-1}). Growth rates were calculated by averaging the instantaneous growth rates for 0 to 24 h and for 24 to 48 h. Symbols represent treatment means ± 1 SE. Curves are fitted by a Michaelis-Menten equation (Eq. 2) using all treatments in the experiment. Growth rate (GR, d^{-1}) = 1.37 {(x – 111)/[412 + (x – 111)]}, r^2 = 0.937

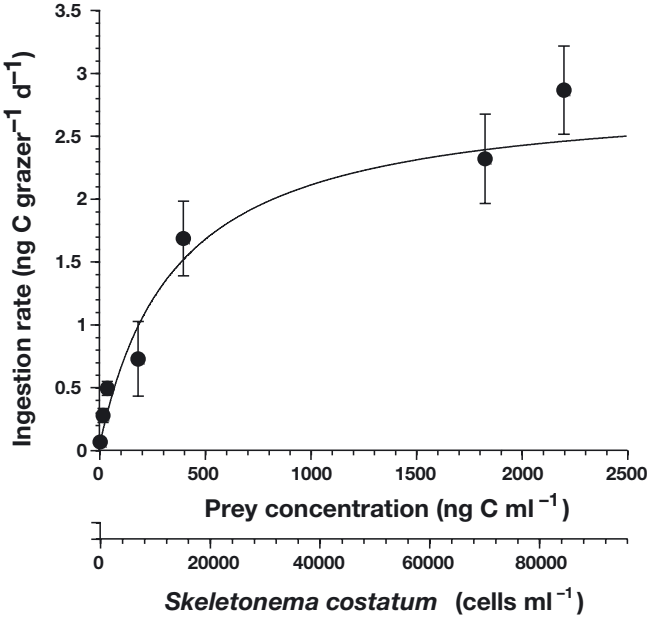

Fig. 2. Ingestion rates of *Protoperidinium bipes* feeding on *Skeletonema costatum* as a function of mean prey concentration (x, ng C ml^{-1}). Ingestion rates were calculated by averaging the instantaneous ingestion rates for 0 to 24 h and for 24 to 48 h. Symbols represent treatment means ± 1 SE. Curves are fitted by a Michaelis-Menten equation (Eq. 3) using all treatments in the experiment. Ingestion rate (IR, ng C $grazer^{-1}$ d^{-1}) = 2.9 [x/(355 + x)], r^2 = 0.794

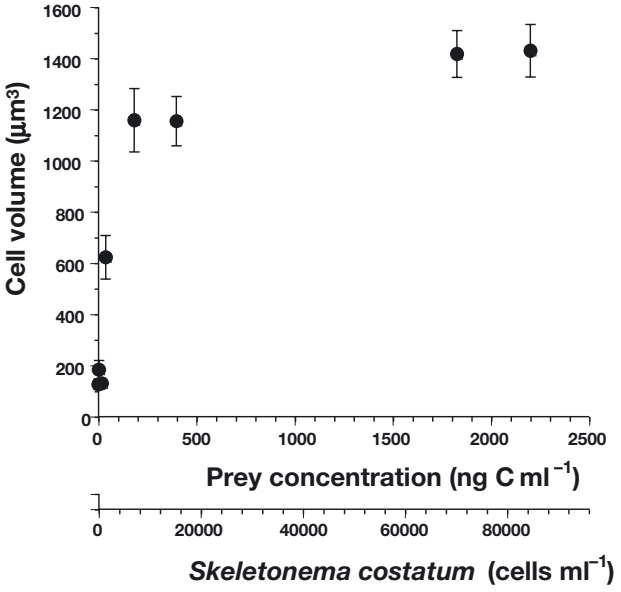

Fig. 3. Cell volume of *Protoperidinium bipes* feeding on *Skeletonema costatum* as a function of mean prey concentration. Symbols represent treatment means ±1 SE. N = 20 for each prey concentration

Gross growth efficiency

GGE of *Protoperidinium bipes* feeding on *Skeletonema costatum* were negative at mean prey concentrations ≤37 ng C ml^{-1} (1420 cells ml^{-1}), and they increased linearly up to 20% with increasing mean prey concentration (Fig. 4).

Grazing impact

Grazing coefficients (g) attributable to *Protoperidinium bipes* feeding on co-occurring *Skeletonema costatum* were 0.001 to 0.034 h^{-1} (Fig. 5A, Table 1). In general g increased with increasing *P. bipes* concentration. g attributable to *Acartia* spp. feeding on *S. costatum* were <0.002 h^{-1} (Fig. 5B, Table 1).

DISCUSSION

Protistan predators on *Skeletonema costatum*

While many metazoan grazers on *Skeletonema costatum* have been reported (Conover 1956, Martin 1965, Paffenhöfer 1976, Deason 1980, Jordana et al. 2001), until now few heterotrophic protists have been known, e.g. *Protoperidinium spinulosum* (Jacobson & Anderson 1986), *Diplopsalis lenticula* (Naustvoll 1998), and *P. pellucidum* (Hansen 1992) are known to feed on *S. costatum*. *P. huberi* is also able to feed on unknown *Skeletonema* species (cell volume of *Skeletonema* sp. = 200 µm^3) (Buskey et al. 1994). However, these studies on the heterotrophic protists have not measured growth and grazing rates of the grazers on *S. costatum* as a function of the prey concentration and there have been few studies to quantify grazing impact by the grazers on the prey. Based on the field data showing that *P. bipes* was abundant when *S. costatum* was abundant in most Korean coastal waters (Jeong et al. 2000, 2002, Yoo et al. 2002), it has been suggested that *S. costatum* (and possibly other co-occurring diatoms) might support the rapid growth of *P. bipes* when the concentration of the diatoms is high.

Swimming speed

Protoperidinium bipes has much higher average and maximum swimming speeds (4006 and 8269 µm s^{-1}) than any *Protoperidinium* species so far reported (see Table 2); *P.* cf. *quinquecorne* has a swimming speed of 1500 µm s^{-1} and *P. subinerme*, *P. ovatum*, *P. claudicans*, and *P. crassipes* have swimming speeds of 100 to 310 µm s^{-1} (Peters 1929 cited by Levandowsky & Kaneta 1987). Data from these previous studies and this study show that the swimming speeds of *Protoperidinium* spp. are not correlated with their cell volume. This relationship suggests that

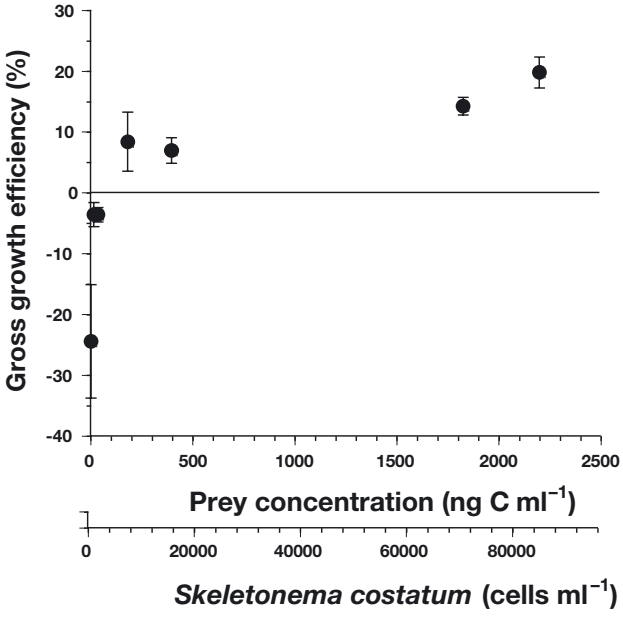

Fig. 4. Gross growth efficiency, defined as *Protoperidinium bipes* biomass produced (+) or lost (–) per *Skeletonema costatum* biomass ingested, as a function of mean prey concentration

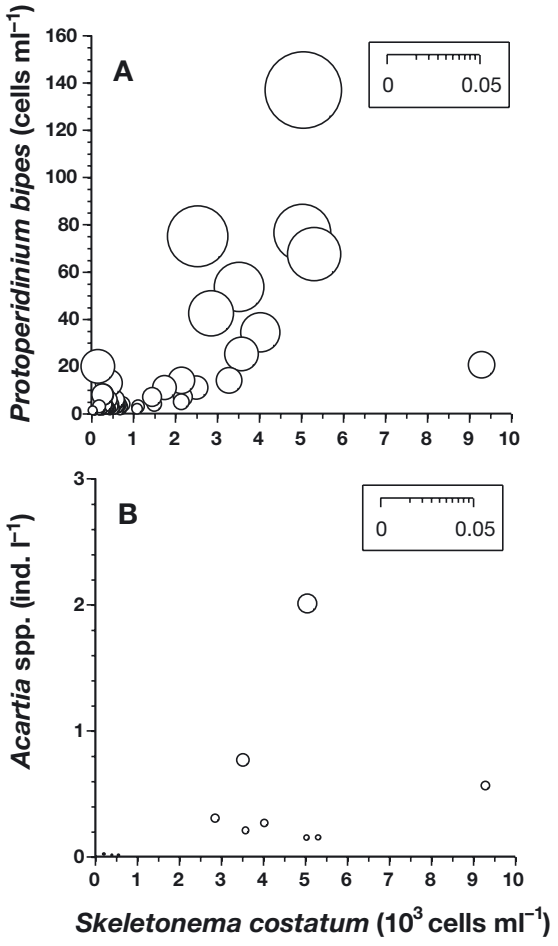

Fig. 5. Calculated grazing coefficients (g, h^{-1}) attributable to (A) *Protoperidinium bipes* and (B) *Acartia* spp. feeding on *Skeletonema costatum* (see text for calculation). N = 60

factors other than prey cell volume, such as cell shape, may have a marked effect on the swimming speed of *Protoperidinium* spp.; *P. bipes* has an elongated shape (ratio of cell length relative to cell width = ca. 1.5), while *P.* cf. *quinquecorne*, *P. subinerme*, *P. ovatum*, *P. claudicans*, and *P. crassipes* have almost spherical shapes (ratios = 0.80 to 1.0). The extremely high swimming speed of *P. bipes* may provide a high encounter rate between this grazer and its prey, particularly at low prey concentrations, but causes great energy loss (Crawford 1992).

Growth

The maximum growth rate (μ) of *Protoperidinium bipes* on *Skeletonema costatum* (1.37 d^{-1}) is the highest among the rates of the *Protoperidinium* species so far reported (see Table 2); *P. hirobis* had a maximum growth rate of 1.23 d^{-1} when feeding on the diatom *Leptocylindrus danicus* (Jacobson & Anderson 1986). The high growth rate of *P. bipes* might enable it to be abundant during diatom blooms in nature (our field data).

There are a few reports on the growth rates of other heterotrophic protists feeding on *Skeletonema costatum*, and this diatom has been known not to be the optimal prey for these grazers; *Gyrodinium dominans* has a maximum growth rate of 0.25 d^{-1} when fed *S. costatum*, while its highest maximum growth rate of 0.54 d^{-1} was observed when fed the dinoflagellate *Heterocapsa triquetra* (Nakamura et al. 1995), when corrected to 20°C using Q_{10} = 2.8 (Hansen et al. 1997). *Protoperidinium pellucidum* has a maximum growth rate of 0.55 d^{-1} when fed *S. costatum* (Hansen 1992), while its highest maximum growth rate of ca. 0.7 d^{-1} was observed when fed the diatoms *Ditylum brightwellii* or *Thalassiosira* sp. (Buskey 1997). Therefore, *P. bipes* exhibited the highest growth rate among heterotrophic protistan grazers of *S. costatum*.

Positive growth rates of *Protoperidinium bipes* feeding on *Skeletonema costatum* were maintained at mean prey concentrations >111 ng C ml^{-1}, which was much higher than those of *P. huberi* feeding on *Ditylum brightwelli* (>10 ng C ml^{-1}) (Buskey et al. 1994), *Diplopsalis lenticula* on *D. brightwelli* (24 ng C ml^{-1}) (Naustvoll 1998), or another pallium feeder, *Oblea rotunda*, on *D. brightwelli* (>10 ng C ml^{-1}) (Strom & Buskey 1993). Energy loss due to the high swimming speed of small *P. bipes* might cause negative growth at low prey concentrations.

Ingestion and clearance

There have been no reports on ingestion and clearance rates of heterotrophic protists feeding on *Skeletonema costatum*, while there are many studies on those of copepods feeding on this diatom.

The maximum ingestion rate (I_{max}) of *Protoperidinium bipes* grazing on *Skeletonema costatum* obtained in this study (2.9 ng C grazer^{-1} d^{-1}) was higher than that of *P. hirobis* grazing on *Leptocylindrus danicus* (0.8), but lower than that for other *Protoperidinium* species on phytoplankton so far reported (5.9 to 17.8), when corrected to 20°C using Q_{10} = 2.8 (Hansen et al. 1997) (Table 2). However, the ratios of I_{max} of *P. bipes* on *S. costatum* relative to cell volume were 6 to 15 times higher than those for other *Protoperidinium* species so far reported.

The maximum clearance rate (C_{max}) of *Protoperidinium bipes* grazing on *Skeletonema costatum* obtained in this study (1.0 μl grazer^{-1} d^{-1}) is higher than that of *Protoperidinium* cf. *divergens* (0.7) and *P. crassipes* (0.5) grazing on *Gonyaulax polyedra* (= presently *Lingulodinium polyedrum*), *P. hirobis* on *Leptocylindrus danicus* (0.5) and the pallium feeder *Oblea rotunda* on *Ditylum brightwellii* (0.7), when corrected

Table 1. Estimation of grazing impact by a *Protoperidinium bipes* population or *Acartia* spp. populations feeding on a *Skeletonema costatum* population using Eq. (3) of this study and the Michaelis-Menten equation in Fig. 4 of Deason (1980), and the abundances of co-occurring *S. costatum*, *P. bipes*, and *Acartia* spp. obtained from the water samples collected off Kwangyang, Korea in 2001. PPIR = *P. bipes*'s population ingestion rate; Pg = *P. bipes*'s grazing coefficient (h^{-1}), APIR = *Acartia* spp.'s population ingestion rate; Ag = *Acartia* spp.'s grazing coefficient (h^{-1})

S. costatum concentration (cells ml^{-1})	*P. bipes* concentration (cells ml^{-1})	PPIR (prey eaten ml^{-1} h^{-1})	Pg (h^{-1})	*Acartia* spp. density (ind. l^{-1})	APIR[a] (prey eaten ml^{-1} h^{-1})	Ag (h^{-1})
417	30	2	0.004	0.0037	0	0
2533	75	54	0.021	0.0002	0	0
2852	42	33	0.012	0.3062	1	0.0004
3520	54	50	0.014	0.7674	1	0.0002
3576	25	24	0.007	0.2088	3	0.0009
4022	34	36	0.009	0.2672	1	0.0003
5028	76	94	0.019	0.1531	1	0.0002
5049	137	169	0.034	2.0095	10	0.0021
5303	68	86	0.016	0.1531	1	0.0002
9290	21	38	0.004	0.5653	4	0.0004

[a]Assuming that the ingestion rate of *Acartia omorii*, the dominant copepod in the water samples collected off Kwangyang, on *Skeletonema cosatum* is the same as that of *A. hudsonica* obtained by Deason (1980) because the size of *A. omorii* is similar to that of *A. hudsonica*

Table 2. Comparison of growth, ingestion and clearance rates in the genus *Protoperidinium*. Rates are corrected to 20°C using Q_{10} = 2.8 (Hansen et al. 1997). PDV: predator volume ($10^3 \times \mu m^3$); DIA: diatom; DN: dinoflagellate; PYV: prey volume ($10^3 \times \mu m^3$); μ_{max}: maximum growth rate (d^{-1}); I_{max}: maximum ingestion rate (ng C $grazer^{-1}$ d^{-1}); C_{max}: maximum clearance rate (µl $grazer^{-1}$ h^{-1}); SC_{max}: maximum volume-specific clearance rate (10^5 h^{-1}); GGE_{max}: maximum gross growth efficiency (%); SS_{max}: maximum swimming speed (mm s^{-1})

Predator	PDV	Prey	PYV	μ_{max}	I_{max}	C_{max}	SC_{max}	GGE_{max}	SS_{max}	Source[a]
P. bipes	1	*Skeletonema costatum* (DIA)	0.25	1.37	2.9	1.0	54	20	8.3	This study
P. hirobis	4	*Leptocylindrus danicus* (DIA)	0.35	1.23	0.8	0.5	1.2	40	0.3[a]	Jacobson & Anderson (1993)
P. pellucidum	25	*Skeletonema costatum* (DIA)	0.25	0.55						Hansen (1992)
P. pellucidum	25	*Thalassiosira* sp. 1 (DIA)	3.6	0.7						Buskey (1997)
P. pellucidum	25	*Ditylum brightwellii* (DIA)	9.7	0.7	11.5				0.6	Buskey (1997)
P. pellucidum	25	*Prorocentrum micans* (DN)	18.4	0.4	7.7					Buskey (1997)
P. pellucidum	25	*Gonyaulax polyedra* (DN)	17.0	0.4						Buskey (1997)
P. huberi	39	*Ditylum brightwellii* (DIA)	9.7	0.72	17.8	23	5.9	59		Buskey et al. (1994)
P. cf. *divergens*	119	*Gymnodinium sanguineum* (DN)	25.0	0.28						Jeong & Latz (1994)
P. cf. *divergens*	119	*Gonyaulax polyedra* (DN)	28.5	0.53	13.3	0.74	0.088	41	1.4	Jeong & Latz (1994)
P. crassipes	204	*Gymnodinium sanguineum* (DN)	25.0	0.12						Jeong & Latz (1994)
P. crassipes	204	*Gonyaulax polyedra* (DN)	28.5	0.34	5.9	0.52	0.025	47		Jeong & Latz (1994)

[a]Mean swimming speed

to 20°C using Q_{10} = 2.8 (Hansen et al. 1997) (Table 2). However, it was lower than that of *P. huberi* grazing on *Ditylum brightwellii* (23) or another pallium feeder, *Diplopsalis lenticula* (4.5) (Buskey et al. 1994, Naustvoll 1998). The high swimming speed of *P. bipes* might increase the encounter rate between this grazer and its prey and thus enable this grazer to capture a prey cell at low prey concentrations.

The maximum volume-specific clearance rate of *Protoperidinium bipes* feeding on *Skeletonema costatum* obtained in this study (5.4×10^6 h^{-1}) was higher than that of any other heterotrophic protists so far reported (Hansen et al. 1997). A high maximum clearance rate of *P. bipes* with a small volume (184 μm^3) may account for this very high maximum volume-specific clearance rate.

Gross growth efficiency

Maximum GGE of *Protoperidinium bipes* feeding on *Skeletonema costatum* (20%) is lower than *P. hirobis* on *Leptocylindrus danicus* (40%), *P.* cf. *divergens* (41) and *P. crassipes* (47) on *Lingulodinium polyedrum*, or *P. huberi* on *Ditylum brightwellii* (59). The energy costs of motility in small, fast-moving protists (swimming at >100 body lengths s^{-1}) were calculated to be very high (>40% of total energy cost) (Crawford 1992). Therefore, energy loss due to the high swimming speed of *P. bipes* might be the cause of low maximum GGE. Otherwise, the nutritional value of *S. costatum* might be lower than that for *L. danicus*, *L. polyedrum*, or *D. brightwellii*.

Grazing impact

Grazing coefficients attributable to *Protoperidinium bipes* feeding on co-occurring *Skeletonema costatum* obtained in the present study were 0.001 to 0.034 h^{-1}; 0.1 to 3.4% of *S. costatum* populations were removed by a *P. bipes* population in 1 h (Table 1). However, grazing coefficients attributable to *Acartia* spp. feeding on *S. costatum* were <0.002 h^{-1}; <0.2% of *S. costatum* populations were removed by *Acartia* spp. populations in 1 h. An *Acartia* spp. density of 33 ind. l^{-1} would be necessary to produce the same grazing impact (0.034 h^{-1}) on *S. costatum* as *P. bipes* in the coastal waters off Kwangyang. The results of the present study suggest that *P. bipes* may sometimes have considerable grazing impact on the population of co-occurring *S. costatum*, and may be a primary zooplanktonic grazer of this diatom.

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