Research Article

Algae 2020, 35(1): 61-78
https://doi.org/10.4490/algae.2020.35.2.28
Open Access

Feeding by common heterotrophic protist predators on seven *Prorocentrum* species

Ji Hyun You[1], Hae Jin Jeong[1,2,*], Hee Chang Kang[1], Jin Hee Ok[1], Sang Ah Park[1] and An Suk Lim[3]

[1]*School of Earth and Environmental Sciences, College of Natural Sciences, Seoul National University, Seoul 08826, Korea*
[2]*Research Institute of Oceanography, Seoul National University, Seoul 08826, Korea*
[3]*Division of Life Science, Gyeongsang National University, Jinju 52828, Korea*

Species belonging to the dinoflagellate genus *Prorocentrum* are known to cause red tides or harmful algal blooms. To understand the dynamics of a *Prorocentrum* sp., its growth and mortality due to predation need to be assessed. However, there are only a few *Prorocentrum* spp. for which heterotrophic protist predators have been reported. We explored feeding by the common heterotrophic dinoflagellates *Gyrodinium dominans*, *Oxyrrhis marina*, *Pfiesteria piscicida*, *Oblea rotunda*, and *Polykrikos kofoidii* and the naked ciliate *Strombidinopsis* sp. (approx. 90 µm cell length) on the planktonic species *Prorocentrum triestinum*, *P. cordatum*, *P. donghaiense*, *P. rhathymum*, and *P. micans* as well as the benthic species *P. lima* and *P. hoffmannianum*. All heterotrophic protists tested were able to feed on the planktonic prey species. However, *O. marina* and *O. rotunda* did not feed on *P. lima* and *P. hoffmannianum*, while *G. dominans*, *P. kofoidii*, and *Strombidinopsis* sp. did. The growth and ingestion rates of *G. dominans* and *P. kofoidii* on one of the seven *Prorocentrum* spp. were significantly different from those on other prey species. *G. dominans* showed the top three highest growth rates when it fed on *P. triestinum*, *P. cordatum*, and *P. donghaiense*, however, *P. kofoidii* had negative growth rates when fed on these three prey species. In contrast, *P. kofoidii* had a positive growth rate only when fed on *P. hoffmannianum*. This differential feeding on *Prorocentrum* spp. between *G. dominans* and *P. kofoidii* may provide different ecological niches and reduce competition between these two common heterotrophic protist predators.

Key Words: ciliate; dinoflagellate; harmful algal bloom; protist; red tide

Abbreviations: ESD, equivalent spherical diameter; HAB, harmful algal bloom; HSD, honestly significant difference; HTD, heterotrophic dinoflagellate; I_{max}, maximum ingestion rate; K_{GR}, prey concentration sustaining 1/2 μ_{max}; K_{IR}, prey concentration sustaining 1/2 I_{max}; μ_{max}, maximum growth rate; OA, okadaic acid; SRC, Sedgewick Rafter chamber

INTRODUCTION

Dinoflagellates are ubiquitous in marine environments (Kudela and Gobler 2012, Jeong et al. 2013, Lee et al. 2018, 2019*b*, Leles et al. 2019). They play diverse roles in marine food webs as primary producers, prey, and predators (Mallin et al. 1991, Hansen 1992, Psarra et al. 2000, Jeong et al. 2010, Anderson and Menden-Deuer 2017, Kang et al. 2019). They often form red tides or harmful algal blooms (HABs), causing human illness and the large-scale mortality of finfish and shellfish (Hallegraeff 1992, Khan et al. 1997, Azanza et al. 2005, Backer

Received January 27, 2020, **Accepted** February 28, 2020
***Corresponding Author**
E-mail: hjjeong@snu.ac.kr
Tel: +82-2-880-6746, Fax: +82-2-874-9695

and McGillicuddy 2006, Jeong et al. 2017). To minimize losses due to red tides or HABs, the population dynamics of red-tide dinoflagellate species should be elucidated (Wyatt and Zingone 2014, Jeong et al. 2015). The growth and mortality of red-tide dinoflagellates due to predation are two critical parameters in their population dynamics (Franklin et al. 2006, Jeong et al. 2015). While there have been many studies reporting the growth rates of red-tide dinoflagellates, there have been fewer studies on their mortality due to predation (Smayda 1997, Matsubara et al. 2007, Jeong et al. 2010, Lee et al. 2019*a*); the mortality of red-tide dinoflagellates due to predation should be explored.

Species in the dinoflagellate genus *Prorocentrum* are known to cause red tides or HABs (Labib 1996, Heil et al. 2005, Pearce et al. 2005, Chen et al. 2006, Ingarao et al. 2009, Kang et al. 2013). To understand the dynamics of a *Prorocentrum* sp., its growth and mortality due to predation need to be assessed. However, there are only a few *Prorocentrum* spp. for which heterotrophic protist predators have been reported (Jeong et al. 2010); many mixotrophic and heterotrophic protist predators of *Prorocentrum cordatum* (Ostenfeld) J. D. Dodge (= *P. minimum*) and *P. micans* Ehrenberg have been found (Strom and Buskey 1993, Nakamura et al. 1995, Lee 1998, Jeong et al. 1999*b*, 2001*b*, Kim and Jeong 2004); *P. donghaiense* D. Lu is known to be eaten by *Oxyrrhis marina* Dujardin (An et al. 2016) and *P. lima* (Ehrenberg) F. Stein is fed on by *Polykrikos kofoidii* Chatton (Matsuoka et al. 2000). Some *Prorocentrum* spp. are planktonic, whereas others are benthic (Faust 1990, Nagahama et al. 2011, Hoppenrath et al. 2013). Furthermore, some species are toxic, and others are not (Murakami et al. 1982, Yasumoto et al. 1987, Morton et al. 1994, Dam and Colin 2005, Sierra-Beltrán et al. 2005, Zingone et al. 2006, Sugahara et al. 2011). *Prorocentrum* spp. vary widely in size (10-50 µm) (Hoppenrath et al. 2013, Lim et al. 2013, Jeong et al. 2015). Thus, a heterotrophic protist predator may respond differently to different *Prorocentrum* spp. Meanwhile, there are several heterotrophic dinoflagellate and ciliate predators that are commonly found in diverse marine environments (Lewis 1990, Strom and Buskey 1993, Nakamura et al. 1995, Jeong et al. 1999*b*, 2006, Coyne et al. 2001, Lowe et al. 2005, Claessens et al. 2008, Watts et al. 2010, Calbet et al. 2013, Tillmann and Hoppenrath 2013). Thus, to understand the population dynamics of a *Prorocentrum* sp., feeding by these common heterotrophic protists on *Prorocentrum* spp. should be explored.

In this study, we explored feeding by the common heterotrophic dinoflagellates (HTDs) *Gyrodinium dominans* Hulbert, *Oblea rotunda* (Lebour) Balech ex Sournia, *O. marina*, *Pfiesteria piscicida* Steidinger & J. M. Burkholder, and *Polykrikos kofoidii* and the naked ciliate *Strombidinopsis* sp. (approximately 90 µm in cell length) on *P. cordatum*, *P. donghaiense*, *P. hoffmannianum* M. A. Faust, *P. lima*, *P. micans*, *P. rhathymum* A. R. Loeblich III, Sherley & R. J. Schmidt, and *P. triestinum* J. Schiller. *Prorocentrum cordatum*, *P. triestinum*, *P. donghaiense*, and *P. rhathymum* are planktonic species, while *P. lima* and *P. hoffmannianum* are benthic species (Hoppenrath et al. 2013, Jeong et al. 2017). *P. cordatum*, *P. donghaiense*, and *P. triestinum* are considerably smaller in size than are *P. rhathymum* and *P. micans*. Additionally, *P. hoffmannianum* and *P. lima* are larger than the other five *Prorocentrum* spp. Thus, several factors affecting feeding by heterotrophic protists on *Prorocentrum* spp. can be examined. HTDs feed on prey cells in various ways: *O. marina* and *Gyrodinium dominans* feed by direct engulfment, *Polykrikos kofoidii* feeds by engulfment after anchoring a prey cell using a taeniocyst-nematocyst complex, *Oblea rotunda* feeds using a pallium after anchoring prey cells using a tow filament, and *Pfiesteria piscicida* feeds using a peduncle. In addition, the naked ciliate *Strombidinopsis* sp. feeds by direct engulfment after generating a feeding current (Barker 1935, Westfall et al. 1983, Gaines and Taylor 1984, Hansen 1991, Burkholder and Glasgow 1997, Verni and Gualtieri 1997). Thus, whether the feeding mechanisms of the heterotrophic protist predators affect feeding or not can be explored.

Using clonal cultures of these *Prorocentrum* spp. and the heterotrophic protists, we investigated whether each predator feeds on each *Prorocentrum* sp. Furthermore, we measured the growth and ingestion rates of *G. dominans* on *P. donghaiense* and *P. kofoidii* on *P. hoffmannianum* as functions of prey concentration. We also measured the growth and ingestion rates of *G. dominans* and *P. kofoidii* on the other *Prorocentrum* spp. at single high prey concentrations. The results of this study may provide a basis for understanding the feeding by heterotrophic protist predators on *Prorocentrum* spp. in marine food webs and the dynamics of these predators and prey species.

MATERIALS AND METHODS

Preparation of experimental organisms

Cells of *Prorocentrum triestinum*, *P. cordatum*, *P. donghaiense*, *P. lima*, *P. rhathymum*, and *P. micans* were isolat-

ed from Korean coastal waters, and clonal cultures were established using two serial single-cell isolations (Table 1). A culture of *P. hoffmannianum* (CCMP683) isolated from the Caribbean Sea was obtained from the National Center for Marine Algae and Microbiota (NCMA), USA (Table 1). All cultures were maintained in 250-mL flasks containing f/2-Si or L1-Si medium (Guillard and Ryther 1962, Guillard and Hargraves 1993), placed on a shelf at 20°C under 20 µmol photons m^{-2} s^{-1} illumination provided by cool-white fluorescent light with a 14 : 10 h light / dark cycle. The carbon content of each *Prorocentrum* sp. was estimated from the cell volume according to Strathmann (1967).

To establish cells of *G. dominans*, *Oxyrrhis marina*, *Oblea rotunda*, and *Polykrikos kofoidii*, plankton samples were collected from water samples of Korean coastal waters during 2001-2019 (Table 2). A clonal culture of each species was established by two serial single-cell isolations. The culture of *Pfiesteria piscicida* was obtained from the NCMA. Moreover, to obtain cells of the ciliate *Strombidinopsis* sp. (approximately 90 µm in cell length), plankton samples were collected using a 10 µm mesh net from the waters of Masan Bay, Korea, in January 2018, when the water temperature and salinity were 5.6°C and 33.0, respectively (Table 2). A clonal culture of *Strombidinopsis* sp. was established by two serial single-cell isolations. The cell volumes of the predators were estimated using the methods of Kim and Jeong (2004) for *G. dominans*, Ok et al. (2017) for *O. rotunda*, Jeong et al. (2001*a*) for *O. marina*, Jeong et al. (2001*b*) for *P. kofoidii*, Jeong et al. (2007) for *P. piscicida*, and Kang et al. (2018) for *Strombidinopsis* sp. (Table 2).

Feeding by heterotrophic protists on each *Prorocentrum* species

Experiment (Expt.) 1 was designed to investigate the predator–prey relationship between each of the seven *Prorocentrum* spp. and each of the six heterotrophic protists after one prey species was provided to one predator species.

A dense culture of each *Prorocentrum* sp. was added to a 42-mL polycarbonate (PC) bottle containing each of the HTDs and added to a well of a 6-well plate chamber containing the ciliate (Table 3). One predator control bottle or well (without *Prorocentrum* prey species) and one prey control bottle or well (without heterotrophic protist predators) were set up for each experiment. The bottles were placed on a plankton wheel rotating at 0.9 rpm (0.00017 g) and incubated at 20°C, under 20 µmol photons m^{-2} s^{-1} illumination provided by cool-white fluorescent light with a 14 : 10 h light / dark cycle. The 6-well

Table 1. Information on the isolation and maintenance of seven *Prorocentrum* spp.

Organisms	Strain name	ESD	CV	Location	Time	T	S
Prorocentrum triestinum	PTMS0205	11.8	0.9	Masan, Korea	May 2002	-	-
P. cordatum (= *P. minimum*)	PMKS9906	12.1	1.1	Gunsan, Korea	Jun 1999	21.1	30.1
P. donghaiense	PDYS1407-1	13.3	1.2	Yeosu, Korea	Jul 2014	-	-
P. rhathymum	PRJJ0907	25.3	9.4	Jeju, Korea	Jun 2012	11.4	13.3
P. micans	PMSH0910	26.0	9.2	Shiwha, Korea	Oct 2009	16.8	27.0
P. lima	DF-114	37.1	17.8	Yeosu, Korea	Nov 2012	16.5	32.4
P. hoffmannianum	CCMP683	43.4	25.5	Caribbean sea, USA	Jan 1985	-	-

ESD, equivalent spherical diameter (µm); CV, cell volume (×10^3 µm^3); T, temperature (°C); S, salinity.

Table 2. Information on the isolation and maintenance of the six potential predator species

Organisms	Type	FM	ESD	CV	Location	Time	T	S	PS	PC
Pfiesteria piscicida (CCMP 2091)	HTD	PD	13.5	1.3	Neuse River, USA	Jan 1998	-	-	Ac	~5.0
Oxyrrhis marina	HTD	EG	15.6	2.0	Gunsan, Korea	May 2001	16.0	27.7	Ac	10.0-20.0
Gyrodinium dominans	HTD	EG	20.0	2.0	Jeongok, Korea	Jul 2019	25.2	31.9	Ac	10.0-20.0
Oblea rotunda	HTD	PA	21.6	5.3	Jinhae bay, Korea	Apr 2015	12.6	31.2	Ac	~10.0
Polykrikos kofoidii	HTD	EG	43.5	43.0	Jangheung bay, Korea	Jul 2016	23.6	26.4	Am	~5.0
Strombidinopsis sp.	NC	FF	68.5	168.1	Masan bay, Korea	Jan 2018	5.6	33.0	Lp	~5.0

HTD, heterotrophic dinoflagellate; NC, naked ciliate; FM, feeding mechanism; PD, peduncle feeder; EG, engulfment feeder; PA, pallium feeder; FF, filter feeder; ESD, equivalent spherical diameter (µm); CV, cell volume (×10^3 µm^3); T, temperature (°C); S, salinity; PS, prey species; Ac, *Amphidinium carterae*; Am, *Alexandrium minutum* CCMP1888 (= *A. lusitanicum*); Lp, *Lingulodinium polyedra*; PC, prey concentration (×10^3 cells mL^{-1}).

plate chamber was placed on the shelf under the same conditions.

Three milliliter aliquots were removed from each bottle after 2, 24, and 48 h incubations and transferred into 6-well plate chambers. Approximately 100 cells of each HTD predator and 10 cells of the ciliate in the plate chambers were observed under a dissecting microscope (or inverted microscope), at a magnification of 10-63× (or 100-400×) to determine whether the predator could feed on the *Prorocentrum* sp. Cells of the predator containing ingested *Prorocentrum* cells were photographed on a confocal dish (SPL100350; SPL Life Sciences Co., Ltd., Pocheon, Korea) using a digital camera (Zeiss AxioCam HRc 5; Carl Zeiss Ltd., Göttingen, Germany) attached to a microscope at 200-400× magnification.

Growth and ingestion rates of *Gyrodinium dominans* on *Prorocentrum donghaiense* as a function of prey concentration

Expt. 2 was designed to measure the growth and ingestion rates of *G. dominans* feeding on *P. donghaiense* as a function of the prey concentration (Table 3). In a preliminary test, among the *Prorocentrum* spp., *P. donghaiense* supported the highest growth rate of *G. dominans*.

In this experiment, a dense culture of *G. dominans* growing on the dinoflagellate *Amphidinium carterae* Hulburt was transferred into a 250-mL culture flask (Falcon; Corning Inc., New York, NY, USA). The flask was filled to capacity with freshly filtered seawater, capped, and placed on a rotating plankton wheel, and incubated under the conditions described above. To monitor the conditions and interactions between predator and prey species, the flask was periodically removed from the rotating wheel, examined through the surface of the capped bottle using a dissecting microscope, and then returned to the rotating wheel. Once the prey cells were no longer detectable, the flask was maintained without added prey for 2 d. This was carried out to minimize possible residual growth resulting from the ingestion of prey during batch culture. Three 1-mL aliquots from the flask were counted, using a compound microscope, to determine the cell concentration of the predator, and the culture was then used to conduct the experiment.

For Expt. 2, the initial concentrations of *G. dominans* and *P. donghaiense* were established using an autopipette to deliver predetermined volumes of known cell concentrations to the bottles. Triplicate 42-mL PC experiment bottles (mixtures of predator and prey) and triplicate control bottles (prey only) were set up for each predator–prey combination. Triplicate control bottles containing only predators were also established at one predator concentration. To obtain similar water conditions, the water of the predator culture was filtered through a 0.2-

Table 3. Design of experiments

Expt. No.	Prey		Predators	
	Species	Density (cells mL^{-1})	Species	Density (cells mL^{-1})
1	Each of *Prorocentrum triestinum*, *P. donghaiense*, *P. micans*, *P. rhathymum*, *P. lima*, and *P. hoffmannianum*	See Table 4	Each of *Oxyrrhis marina*, *Gyrodinium dominans*, *Polykrikos kofoidii*, *Pfiesteria piscicida*, *Oblea rotunda*, and *Strombidinopsis* sp.	See Table 4
2	*P. donghaiense*	88, 170, 498, 969, 4,642, 7,816, 14,105, 19,203, 32,707	*G. dominans*	10, 7, 7, 17, 41, 49, 44, 51, 88 (243)
3	*P. triestinum*	15,475	*G. dominans*	73
4	*P. cordatum*	12,779	*G. dominans*	118
5	*P. rhathymum*	3,019	*G. dominans*	58
6	*P. micans*	2,020	*G. dominans*	117
7	*P. lima*	1,683	*G. dominans*	95
8	*P. hoffmannianum*	1,318	*G. dominans*	192
9	*P. hoffmannianum*	8, 31, 86, 164, 324, 739, 1,276, 1,814	*P. kofoidii*	7, 20, 17, 36, 55, 76, 76, 96 (83)
10	*P. triestinum*	10,977	*P. kofoidii*	52
11	*P. donghaiense*	8,616	*P. kofoidii*	45
12	*P. rhathymum*	1,903	*P. kofoidii*	35
13	*P. lima*	1,019	*P. kofoidii*	84

The numbers in the prey and predator columns are the actual initial densities (cells mL^{-1}) of prey and predators. The values within parentheses in the predator column are the predator densities in the control bottles.

μm disposable syringe filter (DISMIC-25CS type, 25 mm; Advantec, Toyo Roshi Kaisha Ltd., Chiba, Japan) and then added to the prey control bottles in the same amount as that added to the experiment bottles for each predator–prey combination. Similarly, the water of the prey culture was filtered through a 0.2-μm disposable syringe filter and then added to the predator control bottles in the same amount as that of the prey culture added into the experimental bottles. All bottles were filled to capacity with freshly filtered seawater and capped. To determine the actual predator and prey densities at the beginning of the experiment, a 5-mL aliquot was removed from each bottle, fixed with 5% acidic Lugol's solution, and examined with a light microscope to determine predator and prey abundances by enumerating the cells in three 1-mL Sedgewick Rafter chambers (SRCs). The bottles were refilled to capacity with f/2-Si medium, capped, and placed on a rotating wheel under the conditions described above. The dilution of the cultures associated with refilling the bottles was considered when the growth and ingestion rates were calculated. A 10-mL aliquot was taken from each bottle at 48 h and fixed with 5% acidic Lugol's solution. The abundances of predators and prey were determined by counting all or >300 cells in three 1-mL SRCs at 48 h. The conditions of the predators and prey were assessed using a dissecting microscope, as described above, prior to subsampling.

The specific growth rates of *G. dominans* [μ (d^{-1})] were calculated using the following formula:

$$\mu\ (d^{-1}) = [\mathrm{Ln}(G_t\ /\ G_0)]\ /\ t \quad (1)$$

, where G_0 and G_t are the concentrations of the predator at 0 and 2 d, respectively.

Data for the growth rates of *G. dominans* were fitted to a modified Michaelis–Menten equation:

$$\mu\ (d^{-1}) = \mu_{max}(x - x')\ /\ [K_{GR} + (x - x')] \quad (2)$$

, where μ_{max} is the maximum growth rate (d^{-1}), x is the prey concentration (cells mL^{-1} or ng C mL^{-1}), x' is threshold prey concentration (the prey concentration where $\mu = 0$), and K_{GR} is the prey concentration sustaining 1/2 μ_{max}. Data were iteratively fitted to the model using DeltaGraph (Red Rock Software Inc., Salt Lake, UT, USA).

The ingestion and clearance rates and mean prey concentrations were calculated using the equations of Frost (1972) and Heinbokel (1978). The incubation time for calculating the ingestion and clearance rates was the same as that for estimating the growth rate. The data for ingestion rates (IRs, cells $predator^{-1}\ d^{-1}$ or ng C $predator^{-1}\ d^{-1}$) of *G. dominans* were fitted into a modified Michaelis-Menten equation:

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

, where I_{max} is the maximum ingestion rate (cells $predator^{-1}\ d^{-1}$ or ng C $predator^{-1}\ d^{-1}$), x is the prey concentration (cells mL^{-1} or ng C mL^{-1}), and K_{IR} is the prey concentration sustaining 1/2 I_{max}.

Growth and ingestion rates of *Gyrodinium dominans* on all *Prorocentrum* spp. at single prey concentrations

Expts. 3-8 were designed to measure the growth and ingestion rates of *G. dominans* on each of *P. triestinum*, *P. cordatum*, *P. rhathymum*, *P. micans*, *P. lima*, and *P. hoffmannianum* at single high prey concentrations (Table 3).

In these experiments, 9-10 different prey concentrations for each *Prorocentrum* sp. were originally set up, but the abundances of *G. dominans* and the target prey at two or three of the highest prey concentrations were quantified. The growth and ingestion rates of *G. dominans* on each *Prorocentrum* sp. and mean prey concentrations were determined as those in Expt. 2. To compare the growth and ingestion rates of *G. dominans* on each *Prorocentrum* sp., the growth and ingestion rates of *G. dominans* on each *Prorocentrum* sp. at a high mean prey concentration, which was similar to that on the other *Prorocentrum* spp., were selected. Moreover, the growth and ingestion rates of *G. dominans* on *P. donghaiense* at a similar mean prey concentration to those in Expt. 2 were also selected for this comparison.

Growth and ingestion rates of *Polykrikos kofoidii* on *Prorocentrum hoffmannianum* as a function of prey concentration

Expt. 9 was designed to measure the growth and ingestion rates of *P. kofoidii* on *P. hoffmannianum* as a function of the prey concentration (Table 3). In a preliminary test, among the *Prorocentrum* spp., only *P. hoffmannianum* supported a positive growth rate of *P. kofoidii*.

In this experiment, dense cultures of *P. kofoidii* growing on the dinoflagellate *Alexandrium minutum* Halim CCMP1888 (= *A. lusitanicum*) were transferred into two 250-mL culture flasks. The flasks were filled to capacity with freshly filtered seawater, capped, placed on a rotating plankton wheel, and incubated under the conditions

described above. To monitor the conditions and interactions between the predator and prey species, the bottles were periodically removed from the rotating wheel, examined, and then returned. Once the prey cells were no longer detectable, the flasks were maintained without added prey for 1 d. Three 1-mL aliquots from each flask were counted to determine the cell concentration of the predator.

The process of establishing triplicate experiments and prey and predator control bottles, subsampling, determining the abundances of the predator and prey, and calculating the growth and ingestion rates in this experiment was the same as those in Expt. 2.

Growth and ingestion rates of *Polykrikos kofoidii* on all *Prorocentrum* spp. at single prey concentrations

Expts. 10-13 were designed to measure the growth and ingestion rates of *P. kofoidii* on each of *P. triestinum*, *P. donghaiense*, *P. rhathymum*, and *P. lima* at single high prey concentrations (Table 3). Data on the growth and ingestion rates of *P. kofoidii* on *P. cordatum* and *P. micans* at single high prey concentrations were obtained from Jeong et al. (2001*b*) for comparison.

In these experiments, 9-10 different prey concentrations for each of *P. lima* and *P. rhathymum* and two different high prey concentrations for each of *P. donghaiense* and *P. triestinum* were originally set up, but the abundances of *P. kofoidii* and the target prey at two of the highest prey concentrations were quantified. The growth and ingestion rates of *P. kofoidii* on each *Prorocentrum* sp. and mean prey concentrations were determined using the same method as those in Expt. 9. To compare the growth and ingestion rates of *P. kofoidii* on all *Prorocentrum* spp., the growth and ingestion rates of *P. kofoidii* on each *Prorocentrum* sp. at a high mean prey concentration, which was similar to those on the other *Prorocentrum* spp., were selected. Moreover, the growth and ingestion rates of *P. kofoidii* on *P. hoffmannianum* at a similar mean prey concentration to those in Expt. 9 were also selected for this comparison.

Swimming speed

The (previously unrecorded) swimming speeds of *P. rhathymum*, *P. lima*, and *P. hoffmannianum* were measured. The swimming speeds of the four other *Prorocentrum* spp., *G. dominans*, and *P. kofoidii* were obtained from previous studies (Jeong et al. 1999*a*, 2001*b*, 2002, 2015, Kim and Jeong 2004, Berdalet et al. 2007).

A dense culture of a target *Prorocentrum* sp. growing autotrophically in f/2-Si or L1-Si medium was transferred to a 250-mL culture flask. An aliquot from the flask was added to a 38-mL cell culture flask (BD Biosciences, Bedford, MA, USA) and allowed to acclimatize for 30 min. A video camera was focused on one field (appearing as a circle in the cell culture flask) under a dissecting microscope (SZX10; Olympus, Tokyo, Japan) at 20°C, and the swimming movement of the cells of the target *Prorocentrum* sp. was then recorded at 20× magnification using a video analysis system (SRD-1673DN; Samsung Techwin, Seongnam, Korea). The mean and maximum swimming velocities observed after the first 10 min were analyzed for all swimming cells moving randomly. The average swimming speed was calculated based on the linear displacement of cells in 1 s during single-frame playback. The swimming speeds of 20 cells were measured.

Statistical analysis

Univariate analyses and *post-hoc* tests were performed to investigate the effects of the seven different *Prorocentrum* spp. as prey on the growth and ingestion rates of two heterotrophic protists (*G. dominans* and *Polykrikos kofoidii*) at single high mean prey concentrations. Prior to the analyses, normality and homogeneity of variance were tested. A one-way ANOVA with Tukey's honestly significant difference (HSD) *post-hoc* test was performed when both assumptions of normality and homogeneity of variance were satisfied. However, Welch's ANOVA with Games-Howell *post-hoc* test was conducted when the homogeneity of variance was not satisfied. Moreover, a non-parametric Kruskal-Wallis test and Mann-Whitney *U* test with Bonferroni correction was conducted when the normality assumption was not satisfied. To assess the differential effects of *Prorocentrum* spp. on the specific growth and ingestion rates of those two heterotrophic protists, a multivariate analysis of variance (MANOVA) was conducted. Pillai's trace for MANOVA was selected, as this test is robust to the violation of assumptions (Scheiner 1993). The simple linear regression was used to examine relationships between variables (i.e., equivalent spherical diameters [ESD] of *Prorocentrum* species, and the growth and ingestion rates of a predator feeding on each *Prorocentrum* species). All analyses were performed using SPSS ver. 25.0 (IBM-SPSS Inc., Armonk, NY, USA). A 0.05 significance criterion was chosen.

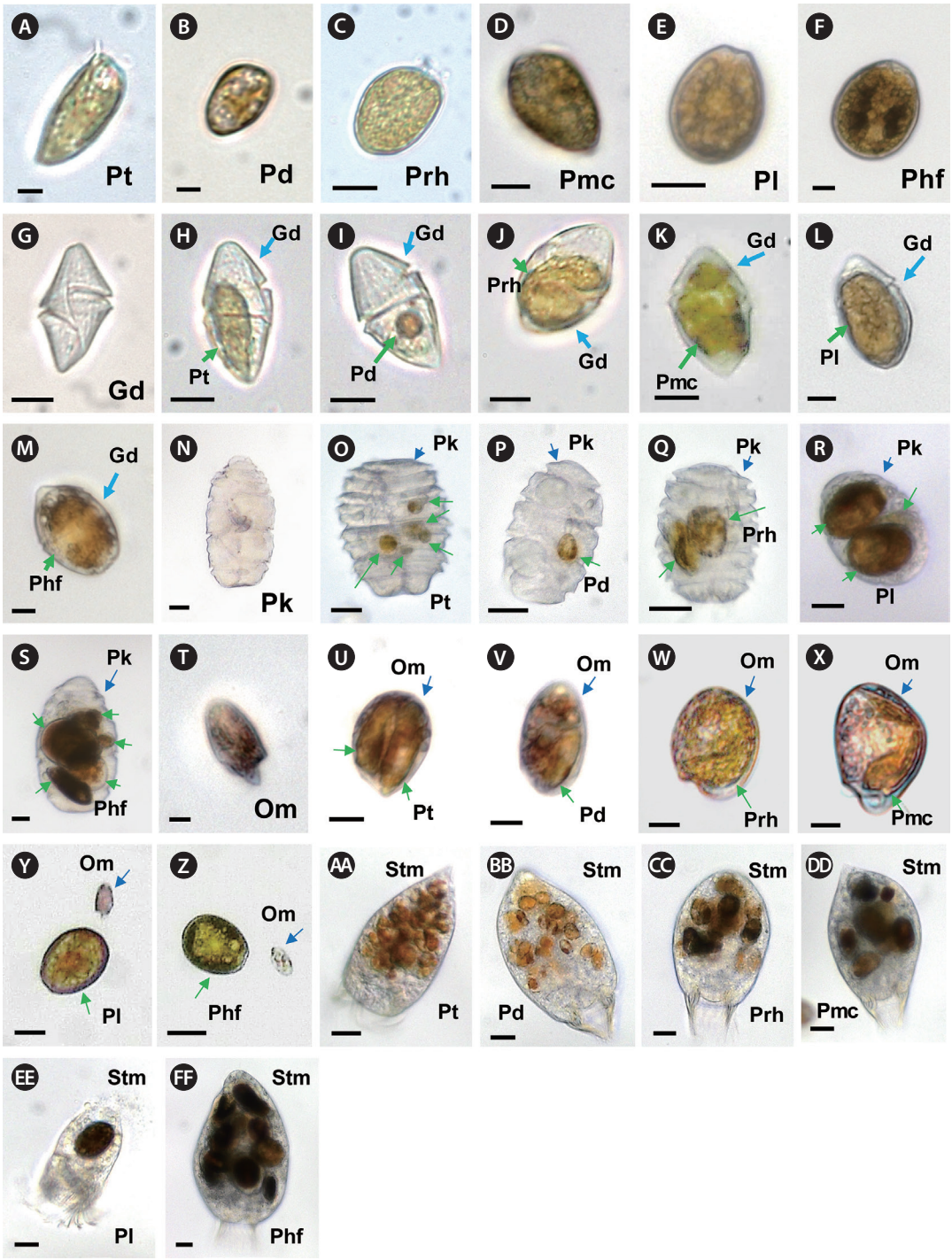

Fig. 1. Feeding by the heterotrophic dinoflagellates *Gyrodinium dominans* (Gd), *Polykrikos kofoidii* (Pk), and *Oxyrrhis marina* (Om) and the naked ciliate *Strombidinopsis* sp. (Stm; AA-FF) on *Prorocentrum* prey species (A-F). Intact *Prorocentrum triestinum* (Pt) (A), *Prorocentrum donghaiense* (Pd) (B), *Prorocentrum rhathymum* (Prh) (C), *Prorocentrum micans* (Pmc) (D), *Prorocentrum lima* (Pl) (E), and *Prorocentrum hoffmannianum* (Phf) (F). Unfed Gd cell (G). Gd with an ingested Pt cell (H). Gd with an ingested Pd cell (I). Gd with an ingested Prh cell (J). Gd with an ingested Pmc cell (K). Gd with an ingested Pl cell (L). Gd with an ingested Phf cell (M). Unfed Pk cell (N). Pk with five ingested Pt cells (O). Pk with an ingested Pd cell (P). Pk with two ingested Prh cells (Q). Pk with two ingested Pl cells (R). Pk with five ingested Phf cells (S). Unfed Om cell (T). Om with two ingested Pt cells (U). Om with an ingested Pd cell (V). Om with an ingested Prh cell (W). Om with an ingested Pmc cell (X). Om did not feed on Pl cell (Y). Om did not feed on Phf cell (Z). Stm with many ingested Pt cells (AA). Stm with many ingested Pd cells (BB). Stm with several ingested Prh cells (CC). Stm with several ingested Pmc cells (DD). Stm with an ingested Pl cell (EE). Stm with several ingested Phf cells (FF). Scale bars represent: A & B, 5 μm; C-M & T-Z, 10 μm; N-S & AA-FF, 20 μm.

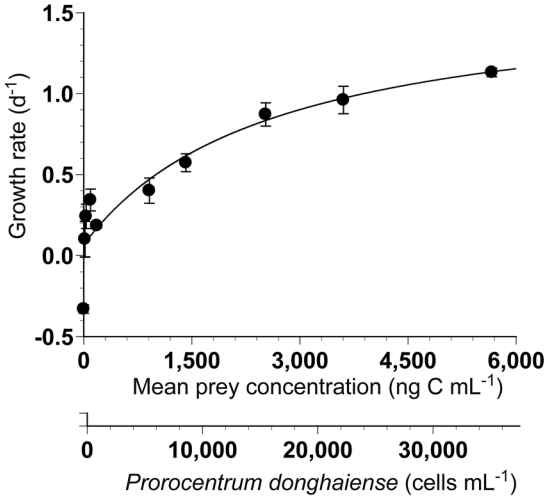

Fig. 2. Specific growth rate (GR) of *Gyrodinium dominans* on *Prorocentrum donghaiense* as a function of mean prey concentration (*x*). Symbols represent treatment means ± standard error. The curve is fitted according to a modified Michaelis-Menten equation [Eq. (2)] using all treatments in the experiment. GR (d^{-1}) = 1.62 {[*x* + 92.5] / [2,510 + (*x* + 92.5)]}, r^2 = 0.866.

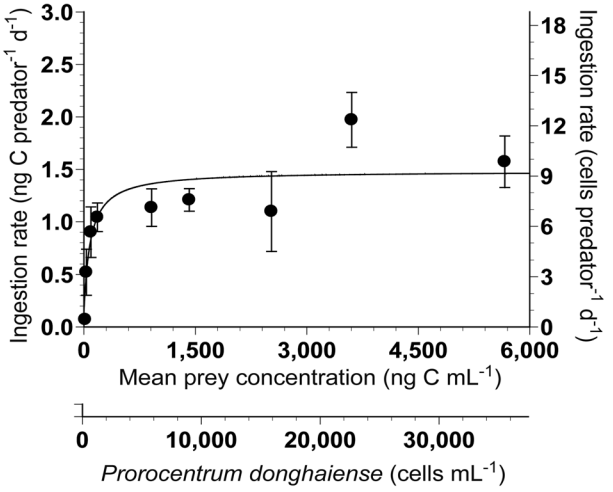

Fig. 3. Ingestion rate (IR) of *Gyrodinium dominans* on *Prorocentrum donghaiense* as a function of the mean prey concentration (*x*). Symbols represent treatment means ± standard error. The curve is fitted according to a modified Michaelis-Menten equation [Eq. (3)] using all treatments in the experiment. IR (ng C predator^{-1} d^{-1}) = 1.48 {[*x*] / [80.1 + (*x*)]}, r^2 = 0.573.

RESULTS

Feeding by heterotrophic protists on each *Prorocentrum* species

All the heterotrophic protists tested were able to feed on *P. triestinum*, *P. cordatum*, *P. donghaiense*, *P. rhathymum*, and *P. micans*, although *Pfiesteria piscicida* was only able to feed on motionless prey cells (Fig. 1). Furthermore, *Gyrodinium dominans*, *Polykrikos kofoidii*, and *Strombidinopsis* sp. were able to feed on *P. lima* and *P. hoffmannianum*, and *P. piscicida* only on motionless prey cells (Fig. 1). However, *Oxyrrhis marina* did not feed on *P. lima* and *P. hoffmannianum*, although it did attempt to engulf them. Moreover, *Oblea rotunda* failed to deploy a pallium on *P. lima* and *P. hoffmannianum*, although it did anchor the cells using a tow filament.

Growth and ingestion rates of *Gyrodinium dominans* on *Prorocentrum donghaiense* as a function of prey concentration

The specific growth rates of *G. dominans* on *P. donghaiense* increased rapidly with increasing mean prey concentrations <2,260 ng C mL^{-1} (14,100 cells mL^{-1}), but increased slowly at higher concentrations (Fig. 2). The specific growth rate of *G. dominans* without added prey was -0.329 d^{-1}. When the data were fitted to Eq. (2), the calculated maximum growth rate (μ_{max}) of *G. dominans* on *P. donghaiense* was 1.62 d^{-1}.

The ingestion rates of *G. dominans* on *P. donghaiense* increased rapidly with increasing mean prey concentrations <155 ng C mL^{-1} (970 cells mL^{-1}), but became saturated at higher concentrations (Fig. 3). When the data were fitted to Eq. (3), the calculated maximum ingestion rate (I_{max}) of *G. dominans* on *P. donghaiense* was 1.48 ng C predator^{-1} d^{-1} (9.3 cells predator^{-1} d^{-1}). The K_{IR} was 80.1 ng C mL^{-1} (501 cells mL^{-1}).

Growth and ingestion rates of *Gyrodinium dominans* on all *Prorocentrum* spp. at single prey concentrations

At single high mean prey concentrations of 2,450-2,779 ng C mL^{-1}, the specific growth rates of *G. dominans* on *P. donghaiense*, *P. triestinum*, and *P. cordatum* were 0.871, 0.850, and 0.759 d^{-1}, respectively; those on *P. rhathymum*, *P. micans*, and *P. hoffmannianum* were 0.499, 0.206, and 0.153 d^{-1}, respectively, but that on *P. lima* was -0.193 d^{-1} (Fig. 4A). The specific growth rates of *G. dominans* on the seven *Prorocentrum* spp. at single high mean prey concentrations were significantly different (ANOVA, $F_{6, 14}$ = 108.14, $p < 0.001$) (Fig. 4A), and Tukey's HSD test revealed that they were divided into four groups.

At single high mean prey concentrations of 2,450-2,779 ng C mL^{-1}, the ingestion rates of *G. dominans* on *P. rha-*

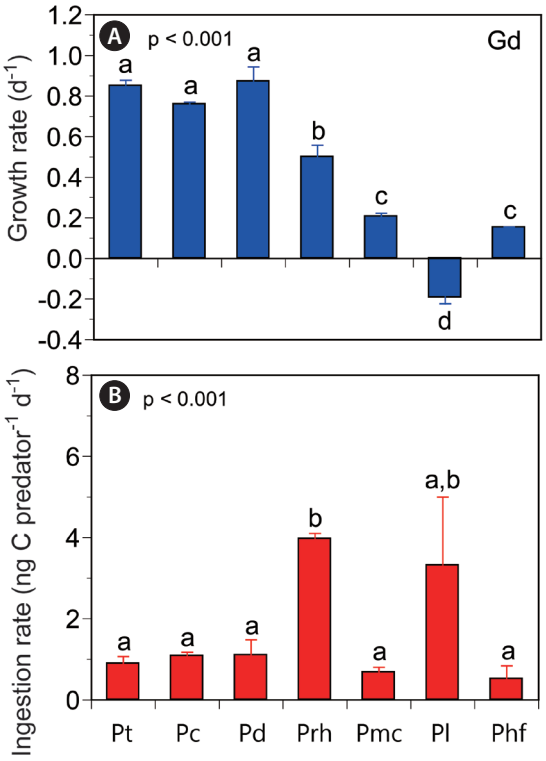

Fig. 4. Growth (A) and ingestion (B) rates of *Gyrodinium dominans* (Gd) on the seven *Prorocentrum* spp. at single high mean prey concentrations. Symbols represent treatment means ± standard error. The different lowercase alphabetical letters above each bar indicate significantly different groups after *post-hoc* tests. Pt, *Prorocentrum triestinum*; Pc, *Prorocentrum cordatum*; Pd, *Prorocentrum donghaiense*; Prh, *Prorocentrum rhathymum*; Pmc, *Prorocentrum micans*; Pl, *Prorocentrum lima*; Phf, *Prorocentrum hoffmannianum*.

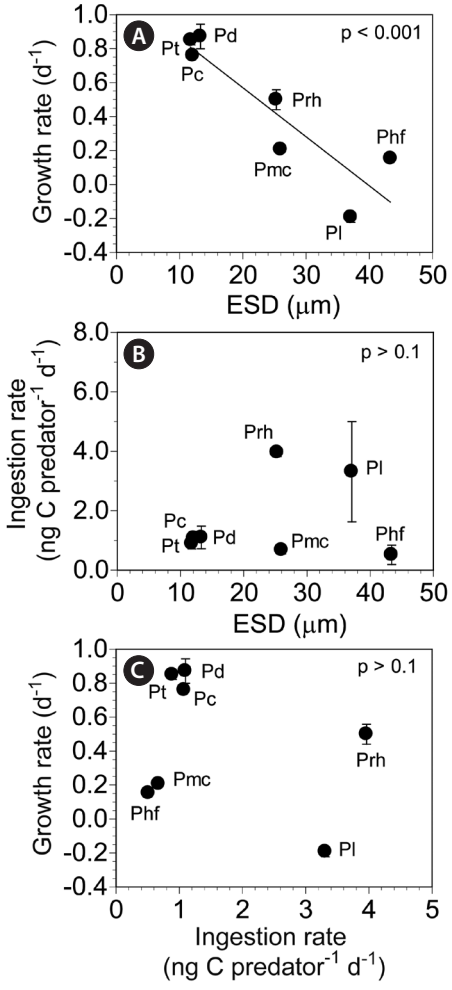

Fig. 5. Growth (GR) (A) and ingestion (B) rates of *Gyrodinium dominans* on seven *Prorocentrum* spp. as functions of prey size (ESD, μm; equivalent spherical diameter). Growth rates of *G. dominans* on seven *Prorocentrum* prey species as functions of the ingestion rates (C). The data were obtained from Fig. 4. Symbols represent treatment means ± standard error. The equation of the linear regression in (A) is as follows: GR (d^{-1}) = -0.029 (ESD) + 1.147, r^2 = 0.785. Pc, *Prorocentrum cordatum*; Pd, *Prorocentrum donghaiense*; Phf, *Prorocentrum hoffmannianum*; Pl, *Prorocentrum lima*; Pmc, *Prorocentrum micans*; Prh, *Prorocentrum rhathymum*; Pt, *Prorocentrum triestinum*.

thymum and *P. lima* were 4.0 and 3.3 ng C predator^{-1} d^{-1}, respectively, and those on *P. donghaiense* and *P. cordatum* were both 1.1 ng C predator^{-1} d^{-1}; however, those on *P. triestinum*, *P. micans*, and *P. hoffmannianum* were 0.9, 0.7, and 0.5 ng C predator^{-1} d^{-1}, respectively (Fig. 4B). The ingestion rates of *G. dominans* feeding on the seven *Prorocentrum* spp. were also significantly different (Welch's ANOVA, $F_{6, 6.08} = 43.24$, $p < 0.001$) (Fig. 4B), and the Games-Howell *post-hoc* test revealed that they were divided into two groups.

The specific growth rates of *G. dominans* on the seven *Prorocentrum* spp. were significantly correlated with prey sizes (linear regression, F-test, F = 69.46, r^2 = 0.785, $p < 0.001$), but the ingestion rates were not significantly correlated with prey sizes (linear regression, F-test, F = 0.59, r^2 = 0.030, p = 0.451) (Fig. 5A & B). Furthermore, the growth rates of *G. dominans* on the seven *Prorocentrum* spp. were not significantly correlated with the ingestion rates (linear regression, F-test, F = 1.35, r^2 = 0.066, p = 0.261) (Fig. 5C).

Growth and ingestion rates of *Polykrikos kofoidii* on *Prorocentrum hoffmannianum* as a function of prey concentration

The specific growth rates of *P. kofoidii* on *P. hoffmannianum* ranged from -0.325 to 0.165 d^{-1} (Fig. 6). The specific growth rates of *P. kofoidii* on *P. hoffmannianum* at mean prey concentrations of 4-577 ng C mL^{-1} (2-254 cells mL^{-1}) were not significantly different from zero ($p > 0.1$, two-tailed *t*-test). However, the specific growth rate of *P. kofoidii* on *P. hoffmannianum* at 1,518 ng C mL^{-1} (669

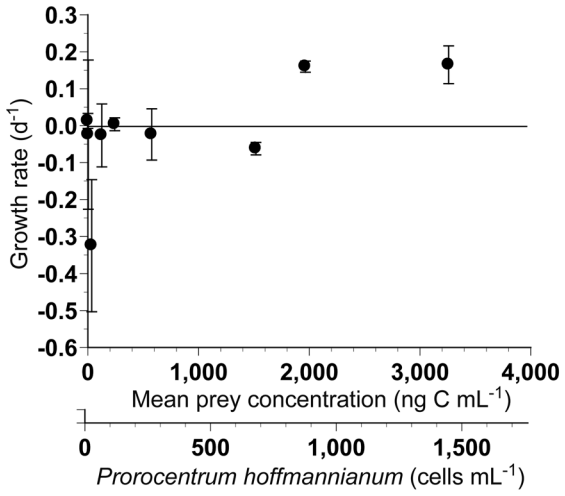

Fig. 6. Specific growth rate of *Polykrikos kofoidii* on *Prorocentrum hoffmannianum* as a function of the mean prey concentration (*x*). Symbols represent treatment means ± standard error.

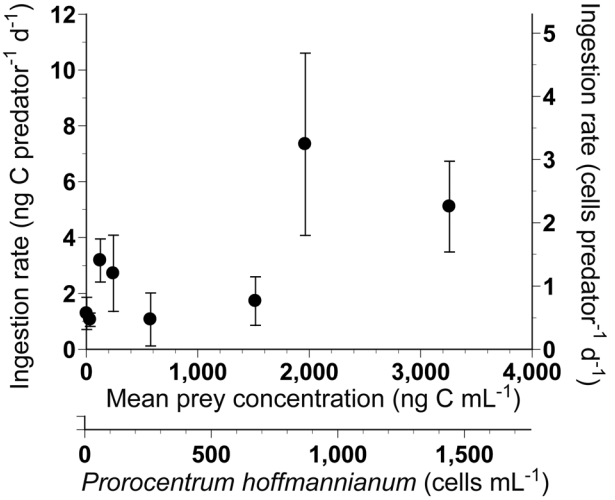

Fig. 7. Ingestion rate of *Polykrikos kofoidii* on *Prorocentrum hoffmannianum* as a function of the mean prey concentration (*x*). Symbols represent treatment means ± standard error.

cells mL^{-1}) was significantly lower than zero ($p < 0.05$, one-tailed *t*-test), whereas those at 1,965-3,259 ng C mL^{-1} (866-1,436 cells mL^{-1}) were significantly greater than zero ($p < 0.05$ or $p < 0.005$, one-tailed *t*-test) (Fig. 6).

The ingestion rates of *P. kofoidii* on *P. hoffmannianum* ranged from 1.1 to 7.3 ng C predator^{-1} d^{-1} (Fig. 7). The ingestion rates of *P. kofoidii* on *P. hoffmannianum* at mean prey concentrations of 4-126 ng C mL^{-1} (2-55 cells mL^{-1}) and 1,965-3,259 ng C mL^{-1} (866-1,436 cells mL^{-1}) were significantly greater than zero ($p < 0.05$ or 0.01, one-tailed *t*-test); however, the rest were not significantly different from zero at the other mean prey concentrations ($p > 0.1$, two-tailed *t*-test).

Growth and ingestion rates of *Polykrikos kofoidii* on all *Prorocentrum* spp. at single prey concentrations

At single high mean prey concentrations of 1,442-1,965 ng C mL^{-1}, the specific growth rate of *P. kofoidii* on *P. hoffmannianum* was 0.160 d^{-1}, but those on *P. donghaiense*, *P. lima*, *P. rhathymum*, and *P. triestinum* ranged from -0.272 to -0.071 d^{-1} (Fig. 8A). The specific growth rates of *P. kofoidii* on *P. cordatum* and *P. micans* at 1,467-2,303 ng C mL^{-1} were -0.363 and -0.042 d^{-1}, respectively (Kim and Jeong 2004). The specific growth rates of *P. kofoidii* on the seven *Prorocentrum* spp. at single high mean prey concentrations were significantly different (ANOVA, $F_{6, 14} = 6.32$, $p = 0.002$) (Fig. 8A), and Tukey's HSD test revealed that they were divided into two groups.

At single high mean prey concentrations of 1,442-1,965 ng C mL^{-1}, the ingestion rate of *P. kofoidii* on *P. hoffmannianum* was 7.3 ng C predator^{-1} d^{-1}, those on *P. lima* and *P. rhathymum* were 4.2-4.7 ng C predator^{-1} d^{-1}, and those on *P. donghaiense* and *P. triestinum* were 0.8-1.0 ng C predator^{-1} d^{-1} (Fig. 8B). The ingestion rates of *P. kofoidii* on *P. cordatum* and *P. micans* at 1,467-2,303 ng C mL^{-1} were 0.4 and 4.1 ng C predator^{-1} d^{-1}, respectively (Kim and Jeong 2004). The ingestion rates of *P. kofoidii* on the seven *Prorocentrum* spp. at single high mean prey concentrations were significantly different (Kruskal-Wallis test, $H_6 = 12.98$, $p = 0.043$) (Fig. 8B); however, they were not divided into different groups as per the Mann-Whitney *U* test with Bonferroni correction.

The specific growth rates of *P. kofoidii* on the seven *Prorocentrum* spp. were significantly correlated with prey sizes (linear regression, F-test, $F = 9.04$, $r^2 = 0.322$, $p = 0.007$), and the ingestion rates were also significantly correlated with prey sizes (linear regression, F-test, $F = 18.74$, $r^2 = 0.497$, $p < 0.001$) (Fig. 9A & B). Furthermore, the growth rates of *P. kofoidii* on the seven *Prorocentrum* spp. were significantly correlated with the ingestion rates (linear regression, F-test, $F = 8.31$, $r^2 = 0.304$, $p = 0.010$) (Fig. 9C).

The specific growth rates of *G. dominans* and *P. kofoidii* were significantly differently affected by the seven *Prorocentrum* spp. at single high mean prey concentrations (MANOVA, Pillai's trace = 1.69, $F_{12, 28} = 12.65$, $p < 0.001$). Moreover, the ingestion rates of *G. dominans* and *P. kofoidii* were significantly differently affected by the seven

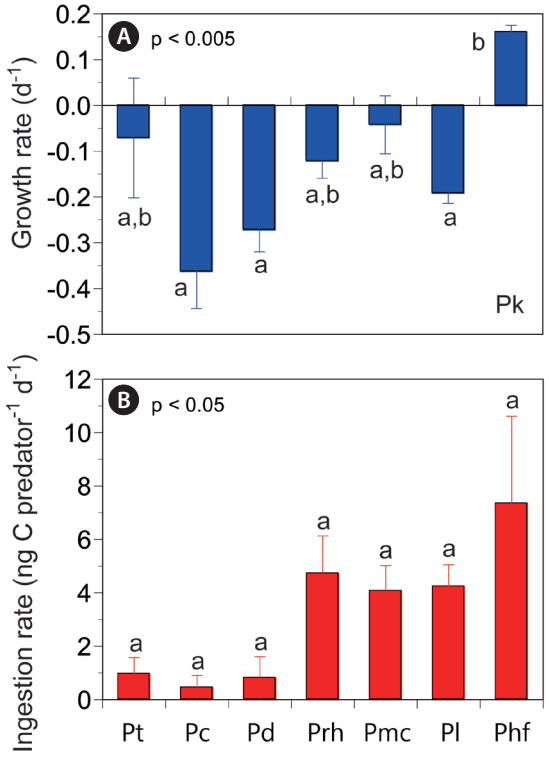

Fig. 8. Growth (A) and ingestion (B) rates of *Polykrikos kofoidii* (Pk) on the seven *Prorocentrum* spp. at single high mean prey concentrations. Symbols represent treatment means ± standard error. The different lowercase alphabetical letters above each bar indicate significantly different groups after *post-hoc* tests. Pt, *Prorocentrum triestinum*; Pc, *Prorocentrum cordatum*; Pd, *Prorocentrum donghaiense*; Prh, *Prorocentrum rhathymum*; Pmc, *Prorocentrum micans*; Pl, *Prorocentrum lima*; Phf, *Prorocentrum hoffmannianum*.

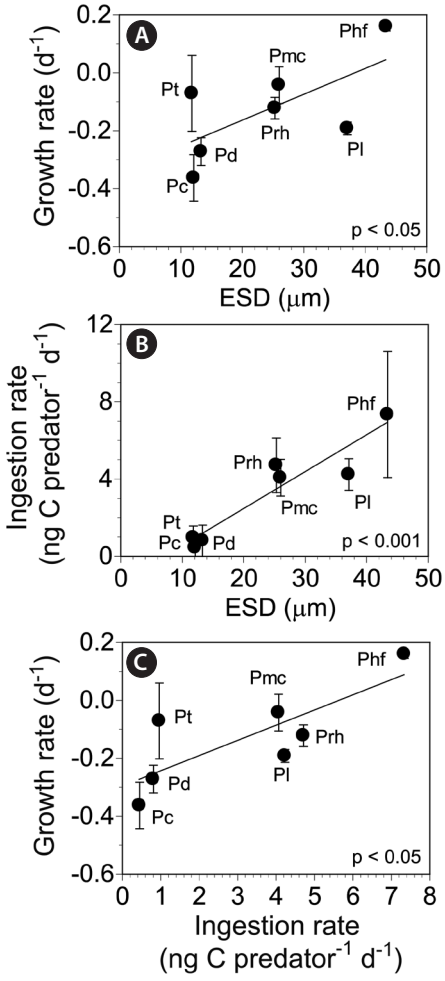

Fig. 9. Growth (GR) (A) and ingestion (B) rates of *Polykrikos kofoidii* on seven *Prorocentrum* spp. as functions of prey size (ESD, μm; equivalent spherical diameter). Growth rates of *P. kofoidii* on seven *Prorocentrum* spp. as a function of the ingestion rates (IRs) (C). The data were obtained from Fig. 8. Symbols represent treatment means ± standard error. The equations of the linear regressions are followings: (A) GR (d^{-1}) = 0.009 (ESD) - 0.345, r^2 = 0.322; (B) IR (ng C $predator^{-1}$ d^{-1}) = 0.191 (ESD) - 1.389, r^2 = 0.496; (C) GR (d^{-1}) = 0.032 (IR) - 0.232, r^2 = 0.307. Pc, *Prorocentrum cordatum*; Pd, *Prorocentrum donghaiense*; Phf, *Prorocentrum hoffmannianum*; Pl, *Prorocentrum lima*; Pmc, *Prorocentrum micans*; Prh, *Prorocentrum rhathymum*; Pt, *Prorocentrum triestinum*.

Prorocentrum spp. at single high mean prey concentrations (MANOVA, Pillai's trace = 1.20, $F_{12, 28}$ = 3.48, p = 0.003).

Swimming speed

The average (± standard error [SE], n = 20) and maximum swimming speeds of *P. rhathymum*, *P. lima*, and *P. hoffmannianum* were 254 (± 18.43) and 420, 78 (± 6.47) and 160, and 82 (± 3.42) and 120 μm s^{-1}, respectively.

The maximum swimming speeds of the seven *Prorocentrum* spp. were not significantly positively correlated with cell sizes (linear regression, F-test, F = 0.261, r^2= 0.050, p = 0.631) when the data on the maximum swimming speeds of *P. cordatum* (194 μm s^{-1}), *P. triestinum* (175 μm s^{-1}), *P. donghaiense* (280 μm s^{-1}), and *P. micans* (380 μm s^{-1}) obtained from Jeong et al. (1999*a*, 2015) and Berdalet et al. (2007) were used.

DISCUSSION

Feeding by heterotrophic protists on each *Prorocentrum* species

The seven *Prorocentrum* spp. used in this study are known to cause red tides or algal blooms on the surface or bottom of water bodies (Labib 1996, Ismael and Aida 1997, Faust 2009, Ingarao et al. 2009, Li et al. 2011, Kang et al. 2013). Of these, *Prorocentrum cordatum*, *P. micans*,

and *P. triestinum* have been reported to cause red tides in many countries, while *P. donghaiense*, *P. rhathymum*, *P. hoffmannianum*, and *P. lima* have been reported to cause red tides in only some countries (Labib 1996, Ismael and Aida 1997, Hajdu et al. 2000, Hernández-Becerril et al. 2000, Tango et al. 2005, Vila and Masó 2005, Faust 2009, Ingarao et al. 2009, Li et al. 2011, Kang et al. 2013, Jeong et al. 2017, Roselli et al. 2019). However, to understand the population dynamics of each of these species, their growth and mortality rates as a result of predation should be determined. To determine the mortality rate of each *Prorocentrum* sp. as a result of predation, the kind of predators should be first identified. We tested six heterotrophic protist predators that are commonly found in many marine environments; thus, there is a high possibility that any of these predators could encounter any of the seven *Prorocentrum* spp. Prior to this study, the type of heterotrophic protist predators that are able to feed on *P. triestinum*, *P. rhathymum*, and *P. hoffmannianum* had not been reported. This study showed that *Gyrodinium dominans*, *Oxyrrhis marina*, *Oblea rotunda*, *Polykrikos kofoidii*, and *Strombidinopsis* sp. are predators of *P. triestinum* and *P. rhathymum*, while *G. dominans*, *P. kofoidii*, and *Strombidinopsis* sp. are predators of *P. hoffmannianum*. Furthermore, *Pfiesteria piscicida* was able to feed on motionless cells of *P. triestinum*, *P. rhathymum*, and *P. hoffmannianum*, but not moving cells (Table 4).

Prior to this study, *P. micans* was reported to fed upon by *G. dominans*, *O. rotunda*, and *P. kofoidii* (Strom and Buskey 1993, Nakamura et al. 1995, Jeong et al. 2001*b*). Thus, this study extends the list of protist predators of *P. micans* to include *O. marina* and *Strombidinopsis* sp. Meanwhile, *P. donghaiense* was reported to be eaten by *O. marina* (An et al. 2016). Therefore, this study extends the list of protist predators of *P. donghaiense* to include *G. dominans*, *O. rotunda*, *P. kofoidii*, and *Strombidinopsis* sp. Lim et al. (2017) reported a high abundance of *G. dominans* following a red tide dominated by *P. donghaiense* in the South Sea of Korea in 2014. Thus, there is a high possibility that *G. dominans* fed actively on *P. donghaiense* in those waters. In addition, *P. lima* was reported to be eaten by *P. kofoidii* (Matsuoka et al. 2000). Thus, this study extends the list of protist predators of *P. lima* to include *G. dominans* and *Strombidinopsis* sp.

Based on their feeding on seven *Prorocentrum* spp., these six protist predator species can be categorized into three groups: the first group comprises *G. dominans*, *P. kofoidii*, and *Strombidinopsis* sp. that are able to feed on all seven *Prorocentrum* spp. (group I); the second group comprises *O. marina* and *O. rotunda* that are able to feed on some *Prorocentrum* spp., but not others (group II); and the third group comprises *Pfiesteria piscicida* that

Table 4. Feeding occurrence by six heterotrophic predators on each *Prorocentrum* sp.

Prey species	Habitat	ESD	IPC	Om	Gd	Pk	Pp	Or	Stm	Reference
Prorocentrum triestinum	PLK	11.8	10-20	O	O	O	O[b]	O	O	This study
P. cordatum	PLK	12.1	10-20	O[a]	O[a]	O[a]	O[b]	O[a]	O[a]	Strom and Buskey (1993), Lee (1998), Jeong et al. (1999*b*, 2001*b*, 2006), Kim and Jeong (2004), This study
P. donghaiense	PLK	13.3	10-20	O[a]	O	O	O[b]	O	O	An et al. (2016), This study
P. rhathymum	PLK	25.3	5	O	O	O	O[b]	O	O	This study
P. micans	PLK	26.6	5	O	O[a]	O[a]	O[b]	O[a]	O	Strom and Buskey (1993), Nakamura et al. (1995), Jeong et al. (2001*b*, 2006), This study
P. lima	BEN	37.1	0.8	X	O	O[a]	O[b]	X	O	Matsuoka et al. (2000), This study
P. hoffmannianum	BEN	43.4	0.8	X	O	O	O[b]	X	O	This study

Initial predator concentrations (cells mL^{-1}) were 1,000 for *O. marina*, 450-1,000 for *G. dominans*, 100 for *P. kofoidii*, 2,000 for *P. piscicida*, 300 for *O. rotunda*, and 5 for *Strombidinopsis* sp.

ESD, equivalent spherical diameter (μm); IPC, target initial prey concentration ($\times 10^3$ cells mL^{-1}); Om, *Oxyrrhis marina*; Gd, *Gyrodinium dominans*; Pk, *Polykrikos kofoidii*; Pp, *Pfiesteria piscicida*; Or, *Oblea rotunda*; Stm, *Strombidinopsis* sp.; O or O[a], the predator was observed to feed on the target prey cell; X, the predator was not observed to feed on the target prey cell; O or X, tested in this study; O[a], cited from other studies; O[b], the predator fed on motionless cells of the target *Prorocentrum* sp., but did not feed on moving cells. PLK, planktonic *Prorocentrum* spp.; BEN, benthic *Prorocentrum* spp.

is able to feed on motionless cells of all seven *Prorocentrum* spp., but not moving cells (group III). *G. dominans*, *Polykrikos kofoidii*, and *Strombidinopsis* sp. succeeded in engulfing whole cells of the two largest *Prorocentrum* spp., *P. lima* and *P. hoffmannianum*, while *O. marina* tried (and failed) to engulf whole cells of these two *Prorocentrum* spp. In addition, *O. rotunda* failed to deploy a pallium although it did succeed in anchoring a prey cell using a tow filament. Thus, *P. lima* and *P. hoffmannianum* are likely too large for *O. marina* and *O. rotunda* to feed on. Moreover, Jeong et al. (2006) reported that *Pfiesteria piscicida* fed on naked dinoflagellates, but not on living cells of large mixotrophic thecate dinoflagellates of ESD >12 µm. However, *P. piscicida* was able to feed on the dead cells of these large mixotrophic thecate dinoflagellates. Similarly, Kim et al. (2019) reported that *P. piscicida* did not feed on actively swimming cells of *Scrippsiella acuminata* (Ehrenberg) Kretschmann, Elbrächter, Zinssmeister, S. Soehner, Kirsch, Kusber & Gottschling; *S. lachrymosa* J. Lewis ex Head; *S. donghaiensis* H. Gu; and *S. masanensis* S. Y. Lee, H. J. Jeong, S. J. Kim, K. H. Lee & S. H. Jang, but did feed on motionless cells of these species. This study confirms this pattern of only feeding on the motionless cells of large thecate dinoflagellates. Meanwhile, *P. piscicida* did not feed on *Yihiella yeosuensis* S. H. Jang, H. J. Jeong, Ø. Moestrup & N. S. Kang, although it is a naked dinoflagellate (Jeong et al. 2018). The fast jumping behavior of *Y. yeosuensis* was suggested to be responsible for *P. piscicida* not feeding, because heat-killed cells of *Y. yeosuensis* were consumed. *P. piscicida* only feeding on heat-killed prey cells was also found for the actively swimming heterotrophic nanoflagellate *Katablepharis japonica* N. Okamoto & I. Inouye (Kim et al. 2017). Thus, with regard to feeding by *P. piscicida*, the presence and absence of thecate and fast and actively swimming behaviors in prey species are critical.

The maximum swimming speeds of *G. dominans* (2,533 µm s^{-1}), *O. marina* (700 µm s^{-1}), *Pfiesteria piscicida* (670 µm s^{-1}), *Polykrikos kofoidii* (911 µm s^{-1}), and *Strombidinopsis* sp. (1,740 µm s^{-1}) are much greater than those of the seven *Prorocentrum* spp. (120-420 µm s^{-1}), although the maximum swimming speed of *O. rotunda* (420 µm s^{-1}) is comparable to that of *P. rhathymum*, the fastest species among the seven *Prorocentrum* spp. (Cosson et al. 1988, Buskey et al. 1993, Burkholder and Glasgow 1997, Jeong et al. 1999*a*, 1999*b*, 2001*b*, 2002, Kim and Jeong 2004). Therefore, these predators may not have difficulty in catching any of these seven *Prorocentrum* prey species.

Growth and ingestion rates of *Gyrodinium dominans* on *Prorocentrum* spp.

At single high mean prey concentrations, all the *Prorocentrum* spp. tested in this study, except for *P. lima*, supported a positive growth rate of *G. dominans*. At single high mean prey concentrations, the growth rate of *G. dominans* on *P. donghaiense* was the highest (0.871 d^{-1}), followed by *P. triestinum* and *P. cordatum*. The growth rates of *G. dominans* on the seven *Prorocentrum* spp. were significantly correlated with prey size. However, the growth rate of *G. dominans* on *P. lima* was negative, whereas that on the larger *P. hoffmannianum* was positive, although the size of *P. lima* (37.1 µm) is smaller than that of *P. hoffmannianum* (43.4 µm). Meanwhile, the ingestion rate of *G. dominans* on *P. lima* was considerably higher than that on *P. hoffmannianum* at similar mean prey concentrations. Both *P. lima* and *P. hoffmannianum* are known to produce diarrhetic shellfish poisoning toxins such as okadaic acid (OA) and OA analogs (dinophysis toxins) (e.g., Hu et al. 2010). In addition, they produce some polyketides without OA carbon framework; *P. lima* produces prorocentrolide and prorocentin, while *P. hoffmannianum* produces hoffmanniolide and prorocentrol (Torigoe et al. 1988, Hu et al. 1999, Lu et al. 2005, Sugahara et al. 2011). Thus, the prorocentrolide and prorocentin produced by *P. lima* may be partially responsible for this negative growth, whereas the hoffmanniolide and prorocentrol produced by *P. hoffmannianum* may not cause negative growth.

The calculated maximum growth rate (μ_{max}) of *G. dominans* on *P. donghaiense*, when the data were fitted to Eq. (2), was 1.62 d^{-1}, whereas the highest growth rate at the given prey concentration was 1.13 d^{-1} because the growth rates increased slowly but continuously. In the study of Kim and Jeong (2004), the calculated μ_{max} of *G. dominans* on *P. cordatum* and the highest growth rate at the given prey concentration were both 1.13 d^{-1}. Furthermore, at the single high prey concentration, the growth rate of *G. dominans* on *P. donghaiense* was slightly higher than that on *P. cordatum*. Thus, *P. donghaiense* may be the optimal *Prorocentrum* prey species for *G. dominans*, or at least be equally optimal to *P. cordatum*. Moreover, the calculated μ_{max} of *G. dominans* on *P. donghaiense* is greater than that on any other algal prey species (Nakamura et al. 1995, Kim and Jeong 2004, Yoo et al. 2010, Jeong et al. 2011, 2014, Lee et al. 2014, Anderson and Menden-Deuer 2017, Kim et al. 2019). The abundance of *G. dominans* is expected to be high during or after red tides dominated by *P. donghaiense*.

Growth and ingestion rates of *Polykrikos kofoidii* on *Prorocentrum* spp.

At single high mean prey concentrations, *P. hoffmannianum* supported the positive growth of *P. kofoidii*, but the six other *Prorocentrum* spp. caused negative growth. The ingestion rate of *P. kofoidii* on *P. hoffmannianum* was 7.3 ng C predator^{-1} d^{-1}. Thus, a *P. kofoidii* cell acquired 174% of its own body carbon from *P. hoffmannianum* daily. The ingestion rates of *P. kofoidii* on *P. rhathymum*, *P. lima*, and *P. micans* were 4.1-4.7 ng C predator^{-1} d^{-1}. Thus, a *P. kofoidii* cell acquired 98-112% of its own body carbon from *P. rhathymum*, *P. lima*, and *P. micans* daily. Differences in the ingestion rates between *P. hoffmannianum* and these three other *Prorocentrum* spp. may be responsible for the positive and negative growth rates of *P. kofoidii*. *P. hoffmannianum* is known to live attached to macroalgae or floating detritus (Faust 1990, 2009). Strong winds may detach *P. hoffmannianum* cells, allowing *P. kofoidii* to encounter them in the water column (Faust 2009). These detached *P. hoffmannianum* cells can be an excellent prey item for *P. kofoidii*.

The highest growth rate of *P. kofoidii* on *P. hoffmannianum* at the mean prey concentration given in this study (0.165 d^{-1}) was slightly higher than those of the phototrophic dinoflagellates *Effrenium voratum* (H. J. Jeong, S. Y. Lee, N. S. Kang & LaJeunesse) LaJeunesse & H. J. Jeong, *Gymnodinium impudicum* (S. Fraga & I. Bravo) Gert Hansen & Moestrup, *Amphidinium carterae*, and *Gymnodinium aureolum* (E. M. Hulburt) Gert Hansen (0.04-0.11 d^{-1}), but much lower than the μ_{max} of *P. kofoidii* on *Alexandrium tamarense* (Lebour) Balech, *Gymnodinium catenatum* H. W. Graham, *Lingulodinium polyedra* (F. Stein) J. D. Dodge, and *Scrippsiella acuminata* (0.83-1.12 d^{-1}) (Jeong et al. 2001*b*, 2013, 2014, Yoo et al. 2010, Kang et al. 2018). The highest ingestion rate of *P. kofoidii* on *P. hoffmannianum* at the mean prey concentration given in this study (7.3 ng C predator^{-1} d^{-1}) is lower than those on *A. tamarense*, *G. catenatum*, *L. polyedra*, and *S. acuminata* (16.6-26.2 ng C predator^{-1} d^{-1}) (Jeong et al. 2001*b*, Kang et al. 2018). Thus, the low ingestion rate of *P. kofoidii* on *P. hoffmannianum* is partially responsible for its lower growth rate on the prey than those on the common red-tide dinoflagellates *A. tamarense*, *G. catenatum*, *L. polyedra*, and *S. acuminata*.

Conclusively, among the seven *Prorocentrum* spp., *G. dominans* showed the top three highest growth rates when it fed on the three smallest *Prorocentrum* spp., which were *P. triestinum*, *P. cordatum*, and *P. donghaiense*; however, *P. kofoidii* had negative growth rates when fed on these three prey species. Thus, during red tides dominated by *P. triestinum*, *P. cordatum*, and *P. donghaiense*, *G. dominans* is likely to be abundant, while *P. kofoidii* may be rare or absent. In contrast, *P. kofoidii* had a positive growth rate only when fed on *P. hoffmannianum*. Thus, when *P. hoffmannianum* is abundant, *P. kofoidii* may co-occur and *G. dominans* may be absent. This differential feeding on *Prorocentrum* spp. between *G. dominans* and *P. kofoidii* may provide different ecological niches and reduce competition between these two common heterotrophic protist predators.

ACKNOWLEDGEMENTS

We thank Sung Yeon Lee, Kyung Ha Lee, Se Hyeon Jang, Soo Yoon Kim, and Se Hee Eom for technical supports. This research was supported by the Useful Dinoflagellate program of Korea Institute of Marine Science and Technology Promotion (KIMST) funded by the Ministry of Oceans and Fisheries (MOF) and the National Research Foundation (NRF) funded by the Ministry of Science and ICT (NRF-2015M1A5A1041806; NRF-2017R1E1A1A01074419) award to HJJ.

REFERENCES

An, X., Li, X. & Li, Z. 2016. Effects of concentrations of *Prorocentrum donghaiense* and *Oxyrrhis marina* on the feeding behaviour of *Oithona brevicornis*. Nat. Environ. Pollut. Technol. 15:1195-1198.

Anderson, S. R. & Menden-Deuer, S. 2017. Growth, grazing, and starvation survival in three heterotrophic dinoflagellate species. J. Eukaryot. Microbiol. 64:213-225.

Azanza, R. V., Fukuyo, Y., Yap, L. G. & Takayama, H. 2005. *Prorocentrum minimum* bloom and its possible link to a massive fish kill in Bolinao, Pangasinan, Northern Philippines. Harmful Algae 4:519-524.

Backer, L. C. & McGillicuddy, D. J. Jr. 2006. Harmful algal blooms: at the interface between coastal oceanography and human health. Oceanography 19:94-106.

Barker, H. A. 1935. The culture and physiology of the marine dinoflagellates. Arch. Mikrobiol. 6:157-181.

Berdalet, E., Peters, F., Koumandou, V. L., Roldán, C., Guadayol, Ò. & Estrada, M. 2007. Species-specific physiological response of dinoflagellates to quantified small-scale turbulence. J. Phycol. 43:965-977.

Burkholder, J. M. & Glasgow, H. B. Jr. 1997. *Pfiesteria piscicida* and other *Pfiesreria*-like dinoflagellates: behavior,

impacts, and environmental controls. Limnol. Oceanogr. 42:1052-1075.

Buskey, E. J., Coulter, C. & Strom, S. 1993. Locomotory patterns of microzooplankton: potential effects on food selectivity of larval fish. Bull. Mar. Sci. 53:29-43.

Calbet, A., Isari, S., Martínez, R. A., Saiz, E., Garrido, S., Peters, J., Borrat, R. M. & Alcaraz, M. 2013. Adaptations to feast and famine in different strains of the marine heterotrophic dinoflagellates *Gyrodinium dominans* and *Oxyrrhis marina*. Mar. Ecol. Prog. Ser. 483:67-84.

Chen, H. L., Lu, S. H., Zhang, C. S. & Zhu, D. D. 2006. A survey on the red tide of *Prorocentrum donghaiense* in East China Sea, 2004. Ecol. Sci. 25:226-230.

Claessens, M., Wickham, S. A., Post, A. F. & Reuter, M. 2008. Ciliate community in the oligotrophic Gulf of Aqaba, Red Sea. Aquat. Microb. Ecol. 53:181-190.

Cosson, J., Cachon, M., Cachon, J. & Cosson, M. -P. 1988. Swimming behaviour of the unicellular biflagellate *Oxyrrhis marina*: *in vivo* and *in vitro* movement of the two flagella. Biol. Cell 63:117-126.

Coyne, K. J., Hutchins, D. A., Hare, C. E. & Cary, S. C. 2001. Assessing temporal and spatial variability in *Pfiesteria piscicida* distributions using molecular probing techniques. Aquat. Microb. Ecol. 24:275-285.

Dam, H. G. & Colin, S. P. 2005. *Prorocentrum minimum* (clone Exuv) is nutritionally insufficient, but not toxic to the copepod *Acartia tonsa*. Harmful Algae 4:575-584.

Faust, M. A. 1990. Morphologic details of six benthic species of *Prorocentrum* (Pyrrophyta) from a mangrove island, twin cays, belize, including two new species. J. Phycol. 26:548-558.

Faust, M. A. 2009. Ciguatera-causing dinoflagellates in a coral-reef mangrove ecosystem, Belize. Atoll Res. Bull. 569:1-30.

Franklin, D. J., Brussaard, C. P. D. & Berges, J. A. 2006. What is the role and nature of programmed cell death in phytoplankton ecology? Eur. J. Phycol. 41:1-14.

Frost, B. W. 1972. Effects of size and concentration of food particles on the feeding behavior of the marine planktonic copepod *Calanus pacificus*. Limnol. Oceanogr. 17:805-815.

Gaines, G. & Taylor, F. J. R. 1984. Extracellular digestion in marine dinoflagellates. J. Plankton Res. 6:1057-1061.

Guillard, R. R. L. & Hargraves, P. E. 1993. *Stichochrysis immobilis* is a diatom, not a chrysophyte. Phycologia 32:234-236.

Guillard, R. R. L. & Ryther, J. H. 1962. Studies of marine planktonic diatoms: I. *Cyclotella nana* Hustedt, and *Detonula confervacea* (Cleve) Gran. Can. J. Microbiol. 8:229-239.

Hajdu, S., Edler, L., Olenina, I. & Witek, B. 2000. Spreading and establishment of the potentially toxic dinoflagellate *Prorocentrum minimum* in the Baltic Sea. Int. Rev. Hydrobiol. 85:561-575.

Hallegraeff, G. M. 1992. Harmful algal blooms in the Australian region. Mar. Pollut. Bull. 25:186-190.

Hansen, P. J. 1991. Quantitative importance and trophic role of heterotrophic dinoflagellates in a coastal pelagial food web. Mar. Ecol. Prog. Ser. 73:253-261.

Hansen, P. J. 1992. Prey size selection, feeding rates and growth dynamics of heterotrophic dinoflagellates with special emphasis on *Gyrodinium spirale*. Mar. Biol. 114:327-334.

Heil, C. A., Glibert, P. M. & Fan, C. 2005. *Prorocentrum minimum* (Pavillard) Schiller: a review of a harmful algal bloom species of growing worldwide importance. Harmful Algae 4:449-470.

Heinbokel, J. F. 1978. Studies on the functional role of tintinnids in the Southern California Bight. I. Grazing and growth rates in laboratory cultures. Mar. Biol. 47:177-189.

Hernández-Becerril, D. U., Altamirano, R. C. & Alonso, R. R. 2000. The dinoflagellate genus *Prorocentrum* along the coasts of the Mexican Pacific. Hydrobiologia 418:111-121.

Hoppenrath, M., Chomérat, N., Horiguchi, T., Schweikert, M., Nagahama, Y. & Murray, S. 2013. Taxonomy and phylogeny of the benthic *Prorocentrum* species (Dinophyceae): a proposal and review. Harmful Algae 27:1-28.

Hu, T., Curtis, J. M., Walter, J. A. & Wright, J. L. C. 1999. Hoffmanniolide: a novel macrolide from *Prorocentrum hoffmannianum*. Tetrahedron Lett. 40:3977-3980.

Hu, W., Xu, J., Sinkkonen, J. & Wu, J. 2010. Polyketides from marine dinoflagellates of the genus *Prorocentrum*, biosynthetic origin and bioactivity of their okadaic acid analogues. Mini-Rev. Med. Chem. 10:51-61.

Ingarao, C., Lanciani, G., Verri, C. & Pagliani, T. 2009. First record of *Prorocentrum lima* (Dinophyceae) inside harbor areas and along the Abruzzo region coast, W Adriatic. Mar. Pollut. Bull. 58:596-600.

Ismael, G. I. & Aida, M. L. 1997. First record of a *Prorocentrum rhathymum* (Prorocentraceae) red tide in the Gulf of California. Rev. Biol. Trop. 45:1263-1271.

Jeong, H. J., Ha, J. H., Park, J. Y., Kim, J. H., Kang, N. S., Kim, S., Kim, J. S., Yoo, Y. D. & Yih, W. H. 2006. Distribution of the heterotrophic dinoflagellate *Pfiesteria piscicida* in Korean waters and its consumption of mixotrophic dinoflagellates, raphidophytes and fish blood cells. Aquat. Microb. Ecol. 44:263-278.

Jeong, H. J., Kang, H., Shim, J. H., Park, J. K., Kim, J. S., Song, J. Y. & Choi, H. -J. 2001*a*. Interactions among the toxic

dinoflagellate *Amphidinium carterae*, the heterotrophic dinoflagellate *Oxyrrhis marina*, and the calanoid copepods *Acartia* spp. Mar. Ecol. Prog. Ser. 218:77-86.

Jeong, H. J., Kang, H. C., You, J. H. & Jang, S. H. 2018. Interactions between the newly described small- and fast-swimming mixotrophic dinoflagellate *Yihiella yeosuensis* and common heterotrophic protists. J. Eukaryot. Microbiol. 65:612-626.

Jeong, H. J., Kim, J. S., Song, J. Y., Kim, J. H., Kim, T. H., Kim, S. K. & Kang, N. S. 2007. Feeding by protists and copepods on the heterotrophic dinoflagellates *Pfiesteria piscicida*, *Stoeckeria algicida*, and *Luciella masanensis*. Mar. Ecol. Prog. Ser. 349:199-211.

Jeong, H. J., Kim, S. K., Kim, J. S., Kim, S. T., Yoo, Y. D. & Yoon, J. Y. 2001*b*. Growth and grazing rates of the heterotrophic dinoflagellate *Polykrikos kofoidii* on red-tide and toxic dinoflagellates. J. Eukaryot. Microbiol. 48:298-308.

Jeong, H. J., Kim, T. H., Yoo, Y. D., Yoon, E. Y., Kim, J. S., Seong, K. A., Kim, K. Y. & Park, J. Y. 2011. Grazing impact of heterotrophic dinoflagellates and ciliates on common red-tide euglenophyte *Eutreptiella gymnastica* in Masan Bay, Korea. Harmful Algae 10:576-588.

Jeong, H. J., Lim, A. S., Franks, P. J. S., Lee, K. H., Kim, J. H., Kang, N. S., Lee, M. J., Jang, S. H., Lee, S. Y., Yoon, E. Y., Park, J. Y., Yoo, Y. D., Seong, K. A., Kwon, J. E. & Jang, T. Y. 2015. A hierarchy of conceptual models of red-tide generation: nutrition, behavior, and biological interactions. Harmful Algae 47:97-115.

Jeong, H. J., Lim, A. S., Lee, K., Lee, M. J., Seong, K. A., Kang, N. S., Jang, S. H., Lee, K. H., Lee, S. Y., Kim, M. O., Kim, J. H., Kwon, J. E., Kang, H. C., Kim, J. S., Yih, W., Shin, K., Jang, P. K., Ryu, J. -H., Kim, S. Y., Park, J. Y. & Kim, K. Y. 2017. Ichthyotoxic *Cochlodinium polykrikoides* red tides offshore in the South Sea, Korea in 2014: I. Temporal variations in three-dimensional distributions of red-tide organisms and environmental factors. Algae 32:101-130.

Jeong, H. J., Lim, A. S., Yoo, Y. D., Lee, M. J., Lee, K. H., Jang, T. Y. & Lee, K. 2014. Feeding by heterotrophic dinoflagellates and ciliates on the free-living dinoflagellate *Symbiodinium* sp. (Clade E). J. Eukaryot. Microbiol. 61:27-41.

Jeong, H. J., Shim, J. H., Kim, J. S., Park, J. Y., Lee, C. W. & Lee, Y. 1999*a*. Feeding by the mixotrophic thecate dinoflagellate *Fragilidium* cf. *mexicanum* on red-tide and toxic dinoflagellates. Mar. Ecol. Prog. Ser. 176:263-277.

Jeong, H. J., Shim, J. H., Lee, C. W., Kim, J. S. & Koh, S. M. 1999*b*. Growth and grazing rates of the marine planktonic ciliate *Strombidinopsis* sp. on red-tide and toxic dinoflagellates. J. Eukaryot. Microbiol. 46:69-76.

Jeong, H. J., Yoo, Y. D., Kim, J. S., Seong, K. A., Kang, N. S. & Kim, T. H. 2010. Growth, feeding and ecological roles of the mixotrophic and heterotrophic dinoflagellates in marine planktonic food webs. Ocean Sci. J. 45:65-91.

Jeong, H. J., Yoo, Y. D., Lee, K. H., Kim, T. H., Seong, K. A., Kang, N. S., Lee, S. Y., Kim, J. S., Kim, S. & Yih, W. H. 2013. Red tides in Masan Bay, Korea in 2004–2005: I. Daily variations in the abundance of red-tide organisms and environmental factors. Harmful Algae 30(Supp. 1):S75-S88.

Jeong, H. J., Yoon, J. Y., Kim, J. S., Yoo, Y. D. & Seong, K. A. 2002. Growth and grazing rates of the prostomatid ciliate *Tiarina fusus* on red-tide and toxic algae. Aquat. Microb. Ecol. 28:289-297.

Kang, H. C., Jeong, H. J., Jang, S. H. & Lee, K. H. 2019. Feeding by common heterotrophic protists on the phototrophic dinoflagellate *Biecheleriopsis adriatica* (Suessiaceae) compared to that of other suessioid dinoflagellates. Algae 34:127-140.

Kang, H. C., Jeong, H. J., Kim, S. J., You, J. H. & Ok, J. H. 2018. Differential feeding by common heterotrophic protists on 12 different *Alexandrium* species. Harmful Algae 78:106-117.

Kang, N. S., Lee, K. H., Jeong, H. J., Yoo, Y. D., Seong, K. A., Potvin, E., Hwang, Y. J. & Yoon, E. Y. 2013. Red tides in Shiwha Bay, western Korea: a huge dike and tidal power plant established in a semi-enclosed embayment system. Harmful Algae 30(Suppl. 1):S114-S130.

Khan, S., Arakawa, O. & Onoue, Y. 1997. Neurotoxins in a toxic red tide of *Heterosigma akashiwo* (Raphidophyceae) in Kagoshima Bay, Japan. Aquac. Res. 28:9-14.

Kim, J. S. & Jeong, H. J. 2004. Feeding by the heterotrophic dinoflagellates *Gyrodinium dominans* and *G. spirale* on the red-tide dinoflagellate *Prorocentrum minimum*. Mar. Ecol. Prog. Ser. 280:85-94.

Kim, S. J., Jeong, H. J., Jang, S. H., Lee, S. Y. & Park, T. G. 2017. Interactions between the voracious heterotrophic nanoflagellate *Katablepharis japonica* and common heterotrophic protists. Algae 32:309-324.

Kim, S. J., Jeong, H. J., Kang, H. C., You, J. H. & Ok, J. H. 2019. Differential feeding by common heterotrophic protists on four *Scrippsiella* species of similar size. J. Phycol. 55:868-881.

Kudela, R. M. & Gobler, C. J. 2012. Harmful dinoflagellate blooms caused by *Cochlodinium* sp.: global expansion and ecological strategies facilitating bloom formation. Harmful Algae 14:71-86.

Labib, W. 1996. Water discoloration in Alexandria, Egypt, April 1993. I. Occurrence of *Prorocentrum triestinum* Schiffer (red tide) bloom and associated physical and chemical conditions. Chem. Ecol. 12:163-170.

Lee, C. W. 1998. Growth and grazing rates of the heterotrophic dinoflagellate *Oxyrrhis marina* and the ciliate *Stormbidinopsis* sp. on *Prorocentrum* spp. M.S. thesis, Kunsan National University, Gunsan, 36 pp. (in Korean with English abstract).

Lee, K. H., Jeong, H. J., Kang, H. C., Ok, J. H., You, J. H. & Park, S. A. 2019*a*. Growth rates and nitrate uptake of co-occurring red-tide dinoflagellates *Alexandrium affine* and *A. fraterculus* as a function of nitrate concentration under light-dark and continuous light conditions. Algae 34:237-251.

Lee, K. H., Jeong, H. J., Yoon, E. Y., Jang, S. H., Kim, H. S. & Yih, W. 2014. Feeding by common heterotrophic dinoflagellates and a ciliate on the red-tide ciliate *Mesodinium rubrum*. Algae 29:153-163.

Lee, S. Y., Jeong, H. J., Kwon, J. E., You, J. H., Kim, S. J., Ok, J. H., Kang, H. C. & Park, J. Y. 2019*b*. First report of the photosynthetic dinoflagellate *Heterocapsa minima* in the Pacific Ocean: morphological and genetic characterizations and the nationwide distribution in Korea. Algae 34:7-21.

Lee, S. Y., Jeong, H. J., You, J. H. & Kim, S. J. 2018. Morphological and genetic characterization and the nationwide distribution of the phototrophic dinoflagellate *Scrippsiella lachrymosa* in the Korean waters. Algae 33:21-35.

Leles, S. G., Mitra, A., Flynn, K. J., Tillmann, U., Stoecker, D., Jeong, H. J., Burkholder, J., Hansen, P. J., Caron, D., Glibert, P., Hallegraef, G., Raven, J. A., Sanders, R. W. & Zubkov, M. 2019. Sampling bias misrepresents the biogeographical significance of constitutive mixotrophs across global oceans. Glob. Ecol. Biogeogr. 28:418-428.

Lewis, J. 1990. The cyst-theca relationship of *Oblea rotunda* (Diplopsalidaceae, Dinophyceae). Br. Phycol. J. 25:339-351.

Li, Y., Lü, S., Jiang, T., Xiao, Y. & You, S. 2011. Environmental factors and seasonal dynamics of *Prorocentrum* populations in Nanji Islands National Nature Reserve, East China Sea. Harmful Algae 10:426-432.

Lim, A. S., Jeong, H. J., Jang, T. Y., Kang, N. S., Lee, S. Y., Yoo, Y. D. & Kim, H. S. 2013. Morphology and molecular characterization of the epiphytic dinoflagellate *Prorocentrum* cf. *rhathymum* in temperate waters off Jeju Island, Korea. Ocean Sci. J. 48:1-17.

Lim, A. S., Jeong, H. J., Seong, K. A., Lee, M. J., Kang, N. S., Jang, S. H., Lee, K. H., Park, J. Y., Jang, T. Y. & Yoo, Y. D. 2017. Ichthyotoxic *Cochlodinium polykrikoides* red tides offshore in the South Sea, Korea in 2014: II. Heterotrophic protists and their grazing impacts on red-tide organisms. Algae 32:199-222.

Lowe, C. D., Day, A., Kemp, S. J. & Montagnes, D. J. S. 2005. There are high levels of functional and genetic diversity in *Oxyrrhis marina*. J. Eukaryot. Microbiol. 52:250-257.

Lu, C. K., Chou, H. N., Lee, C. K. & Li, Z. -H. 2005. Prorocentin, a new polyketide from the marine dinoflagellate *Prorocentrum lima*. Org. Lett. 7:3893-3896.

Mallin, M. A., Paerl, H. W. & Rudek, J. 1991. Seasonal phytoplankton composition, productivity and biomass in the Neuse River estuary, North Carolina. Estuar. Coast. Shelf Sci. 32:609-623.

Matsubara, T., Nagasoe, S., Yamasaki, Y., Shikata, T., Shimasaki, Y., Oshima, Y. & Honjo, T. 2007. Effects of temperature, salinity, and irradiance on the growth of the dinoflagellate *Akashiwo sanguinea*. J. Exp. Mar. Biol. Ecol. 342:226-230.

Matsuoka, K., Cho, H. -J. & Jacobson, D. M. 2000. Observations of the feeding behavior and growth rates of the heterotrophic dinoflagellate *Polykrikos kofoidii* (Polykrikaceae, Dinophyceae). Phycologia 39:82-86.

Morton, S. L., Bomber, J. W. & Tindall, P. M. 1994. Environmental effects on the production of okadaic acid from *Prorocentrum hoffmannianum* Faust I. temperature, light, and salinity. J. Exp. Mar. Biol. Ecol. 178:67-77.

Murakami, Y., Oshima, Y. & Yasumoto, T. 1982. Identification of okadaic acid as a toxic component of a marine dinoflagellate *Prorocentrum lima*. Bull. Jpn. Soc. Sci. Fish. 48:69-72.

Nagahama, Y., Murray, S., Tomaru, A. & Fukuyo, Y. 2011. Species boundaries in the toxic dinoflagellate *Prorocentrum lima* (Dinophyceae, Prorocentrales), based on morphological and phylogenetic characters. J. Phycol. 47:178-189.

Nakamura, Y., Suzuki, S. -Y. & Hiromi, J. 1995. Growth and grazing of a naked heterotrophic dinoflagellate, *Gyrodinium dominans*. Aquat. Microb. Ecol. 9:157-164.

Ok, J. H., Jeong, H. J., Lim, A. S. & Lee, K. H. 2017. Interactions between the mixotrophic dinoflagellate *Takayama helix* and common heterotrophic protists. Harmful Algae 68:178-191.

Pearce, I., Handlinger, J. H. & Hallegraeff, G. M. 2005. Histopathology in Pacific oyster (*Crassostrea gigas*) spat caused by the dinoflagellate *Prorocentrum rhathymum*. Harmful Algae 4:61-74.

Psarra, S., Tselepides, A. & Ignatiades, L. 2000. Primary productivity in the oligotrophic Cretan Sea (NE Mediterranean): seasonal and interannual variability. Prog. Oceanogr. 46:187-204.

Roselli, L., Vadrucci, M. R., Fanelli, F., Ungaro, N. & Caroppo, C. 2019. First bloom event of the small dinoflagellate *Prorocentrum shikokuense* in the Mediterranean Sea: cryptogenic or introduced? Mar. Pollut. Bull. 139:197-

204.

Scheiner, S. M. 1993. MANOVA: multiple response variables and multispecies interactions. *In* Scheiner, S. M. & Gurevitch, J. (Eds.) *Design and Analysis of Ecological Experiments*. Chapman and Hall, New York, pp. 94-112.

Sierra-Beltrán, A. P., Cortés-Altamirano, R. & Cortés-Lara, M. C. 2005. Occurrences of *Prorocentrum minimum* (Pavillard) in México. Harmful Algae 4:507-517.

Smayda, T. J. 1997. Harmful algal blooms: their ecophysiology and general relevance to phytoplankton blooms in the sea. Limnol. Oceanogr. 42:1137-1153.

Strathmann, R. R. 1967. Estimating the organic carbon content of phytoplankton from cell volume or plasma volume. Limnol. Oceanogr. 12:411-418.

Strom, S. L. & Buskey, E. J. 1993. Feeding, growth, and behavior of the thecate heterotrophic dinoflagellate *Oblea rotunda*. Limnol. Oceanogr. 38:965-977.

Sugahara, K., Kitamura, Y., Murata, M., Satake, M. & Tachibana, K. 2011. Prorocentrol, a polyoxy linear carbon chain compound isolated from the toxic dinoflagellate *Prorocentrum hoffmannianum*. J. Org. Chem. 76:3131-3138.

Tango, P. J., Magnien, R., Butler, W., Luckett, C., Luckenbach, M., Lacouture, R. & Poukish, C. 2005. Impacts and potential effects due to *Prorocentrum minimum* blooms in Chesapeake Bay. Harmful Algae 4:525-531.

Tillmann, U. & Hoppenrath, M. 2013. Life cycle of the pseudocolonial dinoflagellate *Polykrikos kofoidii* (Gymnodiniales, Dinoflagellata). J. Phycol. 49:298-317.

Torigoe, K., Murata, M., Yasumoto, T. & Iwashita, T. 1988. Prorocentrolide, a toxic nitrogenous macrocycle from a marine dinoflagellate, *Prorocentrum lima*. J. Am. Chem. Soc. 110:7876-7877.

Verni, F. & Gualtieri, P. 1997. Feeding behaviour in ciliated protists. Micron 28:487-504.

Vila, M. & Masó, M. 2005. Phytoplankton functional groups and harmful algae species in anthropogenically impacted waters of the NW Mediterranean Sea. Sci. Mar. 69:31-45.

Watts, P. C., Martin, L. E., Kimmance, S. A., Montagnes, D. J. S. & Lowe, C. D. 2010. The distribution of *Oxyrrhis marina*: a global disperser or poorly characterized endemic? J. Plankton Res. 33:579-589.

Westfall, J. A., Bradbury, P. C. & Townsend, J. W. 1983. Ultrastructure of the dinoflagellate *Polykrikos*. I. Development of the nematocyst-taeniocyst complex and morphology of the site for extrusion. J. Cell Sci. 63:245-261.

Wyatt, T. & Zingone, A. 2014. Population dynamics of red tide dinoflagellates. Deep Sea Res. Part II: Top. Stud. Oceanogr. 101:231-236.

Yasumoto, T., Seino, N., Murakami, Y. & Murata, M. 1987. Toxins produced by benthic dinoflagellates. Biol. Bull. 172:128-131.

Yoo, Y. D., Jeong, H. J., Kang, N. S., Kim, J. S., Kim, T. H. & Yoon, E. Y. 2010. Ecology of *Gymnodinium aureolum*. II. Predation by common heterotrophic dinoflagellates and a ciliate. Aquat. Microb. Ecol. 59:257-272.

Zingone, A., Siano, R., D'Alelio, D. & Sarno, D. 2006. Potentially toxic and harmful microalgae from coastal waters of the Campania region (Tyrrhenian Sea, Mediterranean Sea). Harmful Algae 5:321-337.