

Microphytobenthos seasonality determines growth and reproduction in intertidal bivalves

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ABSTRACT: The annual cycles of phytoplanktonic and microphytobenthic biomasses as food sources for intertidal bivalves were investigated in a temperate estuarine muddy sandflat of Kwangyang Bay (Korea) from January to December 2002. Seasonal patterns in growth and reproductive activity of a suspension-feeder *Laternula marilina* and a deposit-feeder *Moerella rutila* were examined in order to assess their relationship with the annual cycles of pelagic and benthic microalgae, and were combined with analyses of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ of bivalve tissues and their potential food resources. Biomasses of phytoplankton and microphytobenthos peaked in late spring-summer, and demonstrated a unimodal pattern of seasonal variation. Photosynthetic pigment composition showed a predominance of diatom marker pigment, fucoxanthin, in the water and the sediments throughout the year. Similar patterns of annual cycles in pelagic and benthic microalgal biomasses and similarities in taxonomic composition indicated that resuspended microphytobenthos is an important contributor to the bay's phytoplankton component. This was supported by the $\delta^{13}\text{C}$ values of suspended particulate organic matter (POM) and physical characteristics of the bay. Synchrony in growth and reproductive activity was observed for both bivalves: their shell and tissue growth and gonadal development were achieved together during late spring and summer when chlorophyll *a* (chl *a*) concentrations were highest. This temporal coupling of macrofaunal and microalgal processes indicated that the activities of intertidal bivalves might depend largely on microphytobenthos seasonality. The isotopic signatures of the bivalve tissues demonstrated their dependence on organic matter of microphytobenthic source irrespective of season, despite a comparable contribution of phytoplankton to the diet of the suspension-feeding bivalve. These results highlight the importance of seasonal development of microphytobenthos as an available food source during the critical period of gamete production and growth for both suspension- and deposit-feeding bivalves.

KEY WORDS: Tidal flat · Algal pigment · Phytoplankton · Stable isotopes · Benthic bivalves · Annual cycle · Kwangyang Bay

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INTRODUCTION

Traditional ideas regarding food availability to bivalves have matched their growth and reproductive activities with the annual cycle of phytoplankton biomass in both deep environments and shallow coastal areas (Ansell et al. 1980, Hilbish & Zimmerman 1988, Boon et al. 1998). In a coastal zone with a Mediter-

ranean climate, rainfall and nutrient input are concentrated in winter, and phytoplankton biomass subsequently peaks in spring. Main growth and rapid gonad development of many bivalve species tends to match well with the spring phytoplankton bloom (see above references). In contrast, in a coastal region with a monsoonal climate as exists on the Korean peninsula, heavy rain and nutrient input are concentrated during

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the late spring-early summer and thereby phytoplankton blooms take place in summer (Kang et al. 2000, Park et al. 2001a). These authors showed that the primary growth and reproductive activities of many bivalve species are correlated with the summer phytoplankton blooms in Korean coastal waters.

However, recent stable isotope studies have demonstrated that locally produced organic matter, such as microphytobenthos and macroalgae as well as phytoplankton, support high growth rates, production and biomass of bivalves in coastal ecosystems (Duggins et al. 1989, Ruckelshaud et al. 1993, Sauriau & Kang 2000). For example, in bare and broad intertidal flats, although advection processes of particulate matter of marine and terrestrial origins are important during the submerged period of tidal cycles, microphytobenthos are recognized as important local primary producers. Biomass and production of microphytobenthos can equal or exceed those of phytoplankton in the overlying waters, and microphytobenthos can be resuspended with sediments, transported and deposited elsewhere (Baillie & Welsh 1980, Sullivan & Moncreiff 1988, de Jonge & van Beusekom 1992, Lucas et al. 2000). Their contribution to the carbon budgets in shallow-water systems has also been documented (Colijn & de Jonge 1984, Varela & Penas 1985, Sullivan & Moncreiff 1988, Cahoon & Cooke 1992). Previous isotope studies have also proven the importance of microphytobenthos production as a source of organic matter for benthic animals, as shown by the similarity in isotopic values between microphytobenthos and animals in intertidal and adjacent subtidal areas in estuarine bays (Riera & Richard 1996, Deegan & Garritt 1997, Kang et al. 2003).

Despite the evidence of important ecological linkages between intertidal bivalve production and microphytobenthic biomass, little attention has been paid to the relationship between the annual cycle of microphytobenthos biomass and benthic activities (i.e. growth and reproduction) of tidal flat macrobenthos (e.g. Sauriau & Kang 2000). Microphytobenthos biomass is variable with time and space in this dynamic system. In this respect, seasonal dynamic patterns of different algal components may provide a clue to the relative importance of advected particulate organic matter (POM: phytoplankton) and locally produced microphytobenthos to the benthic activities of bivalves.

Laternula marilina (Reeve, 1860) and *Moerella rutila* (Dunker, 1860) are common bivalve species on tidal flats from the south to the west coast of Korea. *Laternula marilina* is an infaunal suspension feeder, which obtains its nutrients by filtering fine particles suspended in the water, and *M. rutila* is an infaunal surface-deposit feeder, which obtains food from the surface of the sediment by using its siphon. Generally speaking, suspension feeders are exposed to greater

fluctuations in seston within its habitat on tidal flats than are deposit feeders, whereas deposit feeders are food-limited because sediments are mainly composed of inorganic matter (Lopez et al. 1989). However, it has been difficult to identify the actual food resources affecting the activities of intertidal animals. Accordingly, despite their high abundance in Korean tidal flats, information on the ecology of these 2 bivalve species has not previously been available.

This study tested the hypothesis that seasonal development of microphytobenthos can serve as a food source responsible for growth and reproductive activities of bivalves in the bare and broad muddy sandflats in Kwangyang Bay, Korea. To test this hypothesis, we compared the annual cycles of concentration and composition of microalgal groups with the benthic activities of 2 intertidal bivalve species, *Laternula marilina* and *Moerella rutila*. The photosynthetic pigment concentration and composition of sediments and seston were used as tracers of variations in food availability to bivalves, and the isotopic composition of potential food resources and bivalve tissues were used as tracers of actual food sources for bivalves.

MATERIALS AND METHODS

Study site. The study was conducted on the muddy sandflat at Seomjin River estuary in Kwangyang Bay (34° 56' N, 127° 48' E), situated on the south coast of Korea (Fig. 1). The total area of the bay is 230 km² at mean sea level. About 15 km² of intertidal flats north of Myo Island was reclaimed for Kwangyang Steel Mill Company (POSCO) in the early 1980s. An additional 6 square kilometers of intertidal flat east of the Works was reclaimed in 1988.

The bay is characterized by semidiurnal tides with a maximal range of about 3.4 m at spring tides, which induce strong currents (up to 50–80 cm s⁻¹ near the surface of the main channel at the mouth of the bay) and develop broad intertidal flats at the northern part (Kwon et al. 2001). Marine waters enter by a narrow southern channel (Yeosu Haeman) and freshwater enters mainly from Seomjin River at the northern end of the bay (mean flow 120 m³ s⁻¹ and annually 5.8 to 8.7 × 10⁸ t). Average salinities from 1989 to 2000 were 31.19 psu in the surface and 31.47 psu in the bottom waters, respectively, and from 1999 to 2000 ranged from 23.02 psu (after heavy rain) to 33.73 psu (in the dry season) (Lee et al. 2001a). Nutrient concentrations are relatively high compared to other coastal bays because of high levels of nutrient loading by discharge from Seomjin River and industrial wastewater. Phytoplankton concentrations range from 2.1–7.0 µg chlorophyll a (chl a) l⁻¹ in surface waters >20 psu to 27.8–34.2 µg

chl a l^{-1} in waters 4 to 7 psu along the salinity gradients in the Seomjin River estuary (Park et al. 2001b).

Dominant species of phytoplankton vary from cryptomonads (*Chroomonas* spp.), green algae (*Scenedesmus incrassatulus* and *Ulothrix tenerrima*), and freshwater diatoms (*Synedra affinis* and *Amphora* sp.) in the low salinity (<20 psu) region to marine diatoms (*Skeletonema costatum* etc.) in the high salinity (>20 psu) region (Kwon et al. 2001). The sediments consist of sand (ϕ 1 to 4, ~80, 75 and 74%), silt (ϕ 5 to 9, ~19, 23 and 24%) and clay (ϕ 9 to 11, ~1, 2 and 2%) at the upper, middle and lower intertidal levels (this study). The reed grass *Phragmites australis* forms large masses of vegetation at the edges of the Seomjin River estuary but most of the grass is confined to the supralittoral zone. *Zostera* sp. beds, which previously existed over the entire bay, vanished in the 1990s and have not recovered. Most of the sandflat is free of macrophytes so that benthic primary production is mainly due to microphytobenthos. The dominant microphytobenthos species were *Navicula* spp., *Cylinderotheca closterium*, *Pleurosigma angulatum*, *Achnathes* sp., *Rhizosolenia alata*, and *Melosira* sp. Detailed information on hydrobiology, sedimentary features, and macrobenthos distribution in the bay is given by Park et al. (1984) and Hong & Yoon (2004).

Sampling and processing. Macrobenthos and sediments were sampled at regular monthly intervals from January to December 2002. The lower portion of the tidal flat emerges twice a month during a spring low tide at about +30 cm of the local mean sea level. Sampling was conducted at midday low tide during spring tide periods. Three stations were fixed at the upper, middle and lower sites of the tidal flat, respectively. On each occasion, the upper site was exposed to the air for about 6 h and the middle and lower sites for about 4 h. Three replicate plots at each station were sampled randomly using a 0.25 m² aluminum corer to determine mean monthly abundance and size of bivalves. Sediments were washed through a 1 mm aperture mesh sieve and all recovered animals were stored in 4% buffered formalin and then transported to the laboratory. Additionally, 40 ind. of each bivalve species were also collected by hand at each sampling date and kept alive overnight in filtered seawater to evacuate gut content for microscopic examination of histological smears and stable isotope analysis.

At all stations, sediment samples for photosynthetic pigment analysis were collected using a 2.9 cm² core

(made with cut-off syringes), which was gently pushed by hand into the sediment. A total of 5 sediment cores were randomly sampled at each station at the end of the flood tide before the site was submerged. Samples were wrapped in aluminum foil, frozen in dry ice and kept dark until their immediate return to the laboratory. In the laboratory, sediments were separated into 5 slices of 0–5, 5–15, 15–25, 25–35 and 35–45 mm depths. Sediment samples were freeze-dried, ground and then kept in the refrigerator at –80°C until pigment extraction.

At the middle site of the tidal flat, water temperature and salinity data were obtained using the conductivity-temperature-depth (CTD) sensors (Sea-Bird Electronics). At the station, surface and bottom waters at a 0.5 m height above the sea-bottom sediments were sampled at midday high tide during spring tide periods with a pump system. The inlet at the end of polyethylene hoses was attached to the CTD frame. The other end of the tubes running to the ship's deck were connected to a membrane pump. About 20 l of water were pumped into glass tanks and prefiltered with a 200 μ m screen to remove any zooplankton and large particles.

For photosynthetic pigment analysis of water column seston, water samples were filtered through GF/F filters (Whatman, 47 mm diameter) immediately after collection. The filters were wrapped in aluminum foil and frozen in dry ice (–78°C); 2 filters were prepared from each collection. To determine the isotopic value of POM advected to the tidal flat through the central channel of the bay, seston was collected by filtering about 10 l of water on precombusted Whatmann GF/F filters, acidified with a drop of 10% HCl to remove carbonates, rinsed with Milli-Q water and then freeze-

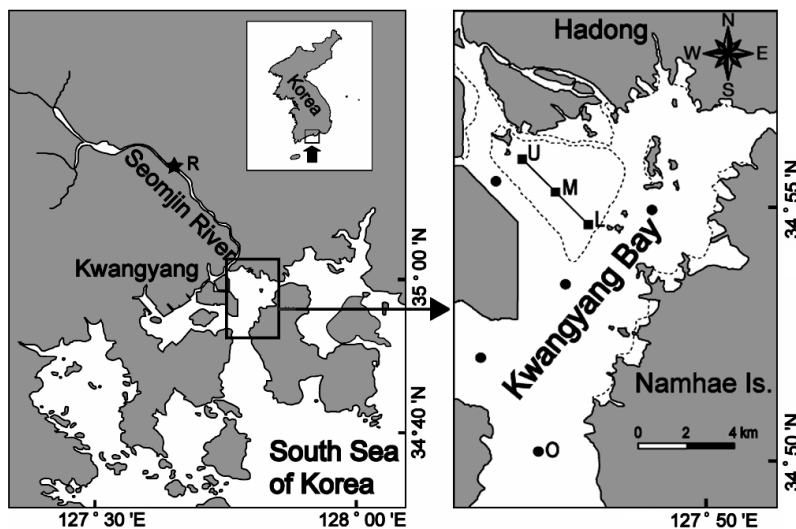


Fig. 1. Sampling sites on the muddy sandflats of Kwangyang Bay. R, riverine site; O, bay mouth site for offshore seston; U, M, L, upper, middle and lower sites on the intertidal flat; ●, subtidal sites for additional observation in 2004

dried (see Kang et al. 1999). Offshore water column seston were collected at the mouth of the bay at mid-day high tide during spring tide periods and prepared using the same methodology. The filters and dried seston samples were stored in a deep freezer (-80°C) until subsequent pigment and isotope analyses. On each sediment-sampling occasion, microphytobenthos were collected by scraping the visible mats of benthic diatoms on the sediment surface, then extracted and prepared for isotope analysis according to Couch's (1989) procedure as described by Riera & Richard (1996).

Pigment analyses of water column seston and sediments. Recent development in high performance liquid chromatography (HPLC) analysis of photosynthetic pigments make it possible to separate the different major chlorophylls, accessory pigments, and chlorophyll degradation pigments (Wright et al. 1991). Since many classes of pigments are specific to certain algal groups, the analysis of photosynthetic pigments allows us to identify different algal groups present in a microalgal community (Jeffrey et al. 1997). Therefore, the HPLC analysis of plant pigments is widely used to quantify microalgal biomass accurately and to identify the presence of major algal groups as taxonomic markers.

Filter samples were extracted with 100% acetone (5 ml) for 24 h in the dark (-20°C) and sonicated for 5 min. The filters were ground by a homogenizer (Glas-Col) to aid disruption of the algal cell. Before analysis, the extracted solution was centrifuged at $2016 \times g$ for 10 min to remove the particulate materials. Freeze-dried sediment samples were weighed (approximately 1 g) and extracted with 100% acetone (10 ml) for 24 h in the dark (-20°C) and sonicated for 5 min. The extracts were then filtered through a $0.20 \mu\text{m}$ Teflon filter and put in an autosampler. 0.25 ml of 1 M ammonium acetate was added to 0.75 ml of extract. After mixing thoroughly, phytopigment analyses on 1 ml of this solution were performed using reverse-phase HPLC (Waters system) as described by Wright et al. (1991). Canthaxanthin was used as an internal standard. Photosynthetic pigments were detected using the Waters 2487 absorbance detector (436 nm) and the Waters 474 Fluorescence detector (Excitation: 432, Emission: 650 nm). Quantification of the standard pigments was determined by spectrophotometer using published extinction coefficients (Jeffrey et al. 1997). Finally, pigment content g^{-1} dry sediment weight was transformed into mg m^{-2} by using the weight and surface area of the sediment sampled. In this study, the top 5 mm of the sediment was selected for the description of the microphytobenthos biomass.

Biometric and gonadosomic measurement of bivalves. Shell length, width and height for all individuals of the 2 bivalve species sorted—a suspension

feeder *Laternula marilina* and a deposit feeder *Moerella rutila*—were measured with Vernier calipers to the nearest 0.05 mm. The samples were then dissected and the dry tissue weight of each individual determined by subtracting water content from wet tissue after being kept for 2 d in a drying oven. Condition index was calculated from the dry weight of tissue and shell according to the formula: $\text{condition} = (\text{dry tissue weight in mg} \times \text{dry shell weight in mg}) \times 100$ (Lucas & Beninger 1985). Biometric measurements were taken from the 40 additional specimens of bivalves in the laboratory, and they were then carefully dissected. For 20 randomly selected specimens of each species, the gonads were fixed in Bouin's solution, embedded in paraffin, sectioned at $5 \mu\text{m}$, and stained with iron hematoxylin-eosin (Humason 1962). The gametogenic stage was classified and scored on a 0 to 4 scale according to Mann's (1979) scheme: Stage 0 = inactive, Stage 1 = early active, Stage 2 = late active, Stage 3 = ripe, Stage 4 = spent. The arithmetic means of the individual scores of the whole specimens was recorded as the gonadal maturity index (GMI) for each sampling date (see details in Dinamani 1987). The flesh of 20 specimens of each bivalve species was acidified (10% HCl) to remove carbonates and rinsed with distilled water, freeze-dried, and stored at -80°C for at least 48 h. The dry flesh was then ground to a fine powder with a mortar and pestle, and kept frozen (-80°C) until stable isotope analysis.

Stable isotope analysis. Stable isotope analysis was also applied to quantify the use of advected (phytoplankton) and locally produced (microphytobenthos) organic matter by the bivalves. Since stable isotope ratios of carbon ($^{13}\text{C}:^{12}\text{C}$) or nitrogen ($^{15}\text{N}:^{14}\text{N}$) in the tissue of an animal directly reflect those of food sources, primary sources of carbon and nitrogen used for animal production in aquatic ecosystems are commonly identified using carbon and nitrogen stable isotope analysis (Fry & Sherr 1984, Michener & Schell 1994). If there is sufficient discrimination in the isotope ratios among potential food sources, the isotope ratios of consumers will indicate which sources of organic matter have been incorporated into the animal tissue over time (DeNiro & Epstein 1979, 1981). In this study, seasonal variations in isotopic ratios of the bivalve tissues and their potential food sources were analyzed in order to confirm whether the bivalves used the major algal component for their growth and gametogenesis, because isotope ratios of algal components can exhibit seasonal variability (e.g. Goering et al. 1990, Cloern et al. 2002).

Dried and ground subsamples (0.5 to 1.5 g) were transferred into small tin capsules for isotope analysis. Samples wrapped in tin capsule were oxidized at high temperature (1030°C) in a Eurovector 3000 Series elemental analyzer (EA), and the resultant CO_2 and N_2

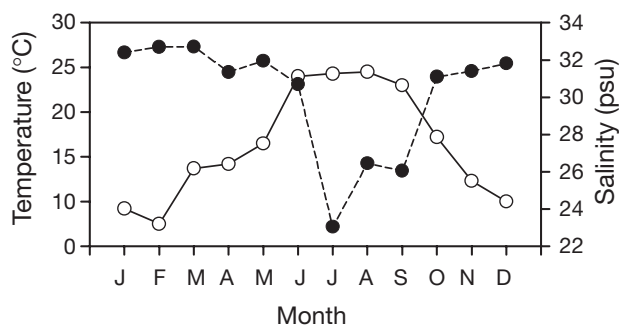


Fig. 2. Variation of temperature (○) and salinity (●) in the water column above the middle site on the intertidal bed in Kwangyang Bay

were analyzed for stable isotope ratios with a Micro-mass Isoprime continuous-flow isotope-ratio mass spectrometer (CF-IRMS). Stable isotope data are expressed as the relative difference between ratios of the sample and conventional standard gases (Pee Dee Belemnite [PDB] for carbon, and atmospheric N₂ for nitrogen). Delta (δ) notation is used to express these relative differences:

$$\delta x(\text{‰}) = [(R_{\text{sample}}/R_{\text{standard}}) - 1] \times 10^3$$

where x is ¹³C or ¹⁵N and R is the ¹³C:¹²C or ¹⁵N:¹⁴N ratio. A secondary standard of known relation to the international standard was used as a reference material. Measurement precision was approximately 0.1 and 0.3 ‰ for δ¹³C and δ¹⁵N, respectively.

Statistics. All data were reported as mean ± 1 SD. Statistical analyses were performed using a general linear model (SPSS). Data were tested for normality using the Shapiro-Wilk procedure. Homogeneity of variance among data was tested using Leven's test to meet the assumptions of parametric statistics before analysis. One-way ANOVAs and Tukey tests were conducted to test significant temporal and spatial differences in the concentrations of pigments and shell length and condition of bivalves. One-way ANOVAs were also performed to determine differences in isotopic values among potential food resources.

RESULTS

Temperature and salinity of the water column

Seasonal variations in temperature and salinity of the water column at the middle site of the tidal flat are shown in Fig. 2. Temperature showed a clear pattern of seasonal variation, with the lowest value (7.5°C) in February, a steep increase in spring, and maximum values in summer (June to September). The highest value (24.5°C) was recorded in August. Salinity

showed a reverse pattern of seasonal variation to temperature. Higher and rather constant values (>31 psu) were observed in winter-spring and fall, and minimum values (<26 psu) in summer (from July to September), following the characteristic summer rainfall in the monsoon climate region.

Algal pigments of the water column and sediments

Chl *a* concentrations from the surface and near-bottom waters were pooled and integrated with depth for the water column. The depth-integrated chl *a* concentrations within the water column at the middle part of the tidal flat displayed a unimodal pattern of seasonal variation with a range from 2.5 to 11.7 mg chl *a* m⁻² (annual mean concentration of 6.8 mg chl *a* m⁻², Fig. 3). Chl *a* concentration remained low during winter-early spring, increased from April, and reached a peak (11.7 to 11.4 mg chl *a* m⁻²) in July to August (1-way ANOVA, $p < 0.001$, compared to the other months). An annual maximum of chl *a* levels during summer was followed by a significant decrease in September to October, and

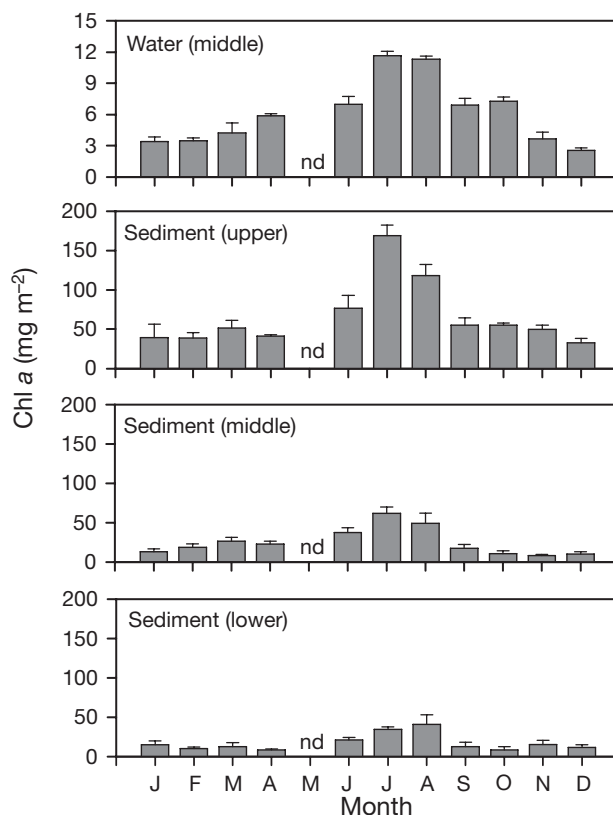


Fig. 3. Depth-integrated chl *a* concentration in the water column at the middle site and chl *a* contents in the top 5 mm of sediment in upper, middle and lower sites on the intertidal flat in Kwangyang Bay. Error bars: SD; nd: not determined

concentrations then remained low during late fall (November to December) ($p < 0.001$).

Chl *a* contents in the top 5 mm of the sediment at all 3 sampling sites varied from 8.3 ± 1.8 (at the middle site in November) to 168.9 ± 13.3 mg chl *a* m^{-2} (at the upper site in July) with an annual mean of 35.3 ± 33.5 mg chl *a* m^{-2} ; therefore, a clear seasonal variation in chl *a* concentration was evident as was also observed in the water column (Fig. 3). A significant increase in sediment chl *a* contents was observed in June at all 3 sites ($p < 0.01$ for all cases). At the upper site on the flat, the annual maximum of sediment chl *a* content was encountered in July (168.9 ± 13.3 mg chl *a* m^{-2}), the contents of June and August were intermediate, and the values of the other months were fairly constant at around 45.4 ± 11.2 mg chl *a* m^{-2} ($p < 0.01$). At the middle and lower sites, the highest contents of chl *a* were 39.4 ± 11.5 and 32.3 ± 11.1 mg chl *a* m^{-2} , respectively; these were recorded in June to August ($p < 0.01$ for both cases). When comparing sites, there were significant differences in sediment chl *a* contents among annual as well as monthly means associated with different submergence times. The chl *a* contents were generally higher ($p < 0.01$) at the upper site (annual mean 66.2 ± 41.6 mg chl *a* m^{-2}) than at the middle and lower sites (23.6 ± 12.9 and 19.0 ± 11.4 mg chl *a* m^{-2} , respectively).

Aside from chl *a* (the most dominant pigment in both the water and sediments), the concentrations of

the other algal pigments (chl *b* and *c*, carotenoids) were expressed as percentages of chl *a* to demonstrate the importance of the different pigment classes (Table 1). The dominant pigments found in the water column throughout the year were fucoxanthin and chl *c*. Though poor, diadinoxanthin was present in all water column samples. These 3 pigment classes identified are all present in diatoms (Stauber & Jeffrey 1988, Wright et al. 1991). Chl *b* and peridinin—marker pigments for chlorophytes/euglenoids and dinoflagellates, respectively (Jeffrey 1974, Wright et al. 1991)—were found in very small amounts. However, chl *b* was present in significant amounts in April to June and November to December, which probably indicated the existence of terrestrial plant detritus. Marker pigments for other taxa, such as 19'-butanoyloxyfucoxanthin, 19'-hexanoyloxyfucoxanthin, prasinoxanthin, violaxanthin, alloxanthin, diatoxanthin, lutein, zeaxanthin and β -carotene, were detected in much smaller proportions or below limits of detection.

The compositional proportions of algal pigments in the sediments were pooled for the 3 sites because their composition pattern did not show a large variation from the standard deviation values among sites (Table 1). The compositional pattern of algal pigment in the sediments was consistent with that in the water column, and demonstrated the concurrent presence of

Table 1. Algal pigments (ng l^{-1} , expressed as ratios of pigment:chl *a*) in water column seston at the middle site, and in sediments at the upper, middle and lower sites of the intertidal flat in Kwangyang Bay, determined by HPLC analysis. b-fuco-xanthin: 19'-butanoyloxyfucoxanthin; h-fuco-xanthin: 19'-hexanoyloxyfucoxanthin. SD shown in parantheses

	Chl <i>c</i>	Peridinin	b-fuco-xanthin	Fuco-xanthin	h-fuco-xanthin	Prasino-xanthin	Viola-xanthin	Diadino-xanthin	Allo-xanthin	Diato-xanthin	Lutein xanthin	Zea-xanthin	Chl <i>b</i>	β -carotene
Seston														
Jan	4.8	2.1	0.0	36.1	0.3	0.1	0.3	4.8	2.2	0.2	0.9	0.2	3.0	0.0
Feb	78.6	1.5	0.0	27.5	0.4	0.0	0.2	4.7	1.6	0.4	0.6	0.2	1.4	0.1
Mar	51.5	1.4	0.0	38.1	0.4	0.0	0.3	6.2	2.7	0.4	0.5	0.1	5.0	0.0
Apr	18.9	1.3	0.1	28.1	0.7	0.1	0.6	2.7	5.3	0.2	1.3	0.1	12.6	0.0
Jun	15.0	4.5	0.0	31.3	0.5	0.2	0.8	3.0	4.4	0.2	1.0	0.3	14.8	0.0
Jul	28.0	5.5	0.0	40.4	0.3	0.0	0.4	4.4	2.3	0.2	0.7	0.2	3.2	0.1
Aug	16.2	0.5	0.0	29.4	0.1	0.0	0.0	2.8	0.0	0.1	0.0	0.0	0.7	1.6
Sep	14.6	1.9	0.0	27.1	0.2	0.0	0.0	2.1	0.0	0.1	0.0	0.0	3.1	1.3
Oct	8.9	0.5	0.0	22.1	0.1	0.0	0.0	1.5	0.0	0.1	0.0	0.0	2.7	0.9
Nov	2.2	0.7	0.0	31.2	0.3	0.1	0.0	0.1	0.0	0.1	0.0	0.0	9.6	0.5
Dec	6.6	3.2	0.0	36.9	0.4	0.1	0.0	0.6	0.0	0.3	0.0	0.0	8.4	0.7
Sediment														
Jan	3.8 (2.6)	0.7 (0.4)	0.0 (0.0)	29.7 (6.2)	0.5 (0.1)	0.2 (0.1)	0.1 (0.0)	4.9 (1.6)	0.6 (0.1)	1.9 (0.4)	0.2 (0.1)	0.3 (0.0)	2.1 (0.8)	2.5 (1.0)
Feb	12.8 (14.0)	4.8 (2.3)	0.3 (0.2)	77.5 (8.1)	8.4 (8.0)	1.6 (1.7)	3.2 (5.1)	70.8 (46.6)	1.1 (0.7)	35.9 (15.9)	1.8 (1.4)	2.3 (1.5)	8.2 (5.7)	11.3 (5.3)
Mar	23.2 (14.5)	0.6 (0.4)	0.0 (0.0)	44.9 (20.6)	0.7 (0.2)	0.1 (0.0)	0.1 (0.1)	7.1 (3.4)	1.1 (0.4)	3.9 (2.1)	0.3 (0.3)	0.4 (0.4)	1.7 (1.6)	1.9 (1.6)
Apr	26.8 (7.6)	1.6 (1.9)	0.0 (0.0)	35.8 (1.1)	0.6 (0.2)	0.1 (0.0)	0.2 (0.1)	5.6 (0.1)	0.9 (0.3)	2.1 (0.2)	0.4 (0.4)	0.3 (0.1)	4.0 (0.5)	0.6 (0.3)
Jun	10.2 (5.5)	0.2 (0.1)	0.0 (0.0)	25.8 (1.9)	0.4 (0.0)	0.0 (0.0)	0.0 (0.0)	7.0 (1.6)	0.1 (0.1)	1.1 (0.2)	0.2 (0.1)	0.7 (0.4)	0.9 (0.1)	0.8 (1.1)
Jul	3.9 (2.0)	0.4 (0.3)	0.0 (0.0)	24.8 (1.0)	0.4 (0.1)	0.0 (0.0)	0.0 (0.0)	6.2 (0.6)	0.0 (0.0)	1.0 (0.2)	0.0 (0.0)	0.1 (0.1)	1.4 (1.2)	1.6 (0.5)
Aug	6.9 (4.1)	0.3 (0.2)	0.0 (0.0)	29.2 (4.1)	0.4 (0.1)	0.0 (0.0)	0.0 (0.0)	5.7 (0.3)	0.2 (0.2)	1.0 (0.2)	0.1 (0.1)	0.1 (0.1)	1.0 (1.0)	0.8 (0.7)
Sep	7.7 (2.3)	0.2 (0.2)	0.0 (0.0)	38.1 (15.7)	0.7 (0.4)	0.0 (0.0)	0.0 (0.0)	9.1 (3.0)	0.2 (0.1)	1.5 (0.5)	0.1 (0.1)	0.0 (0.0)	1.3 (0.7)	0.1 (0.1)
Oct	0.9 (0.2)	0.2 (0.1)	0.0 (0.0)	43.3 (1.6)	0.5 (0.2)	0.0 (0.0)	0.0 (0.0)	11.5 (2.0)	0.1 (0.1)	1.5 (0.2)	0.1 (0.1)	0.1 (0.0)	2.1 (0.5)	0.1 (0.2)
Nov	2.0 (1.3)	0.2 (0.1)	0.0 (0.0)	41.6 (0.8)	0.5 (0.1)	0.0 (0.0)	0.0 (0.0)	12.7 (2.7)	0.1 (0.0)	1.7 (0.2)	0.0 (0.0)	0.0 (0.0)	0.7 (0.5)	0.0 (0.0)
Dec	0.5 (0.1)	0.2 (0.0)	0.0 (0.0)	25.1 (6.2)	0.2 (0.1)	0.0 (0.0)	0.1 (0.0)	6.4 (2.1)	0.2 (0.1)	0.5 (0.2)	0.0 (0.0)	0.0 (0.0)	1.2 (0.4)	1.5 (0.5)

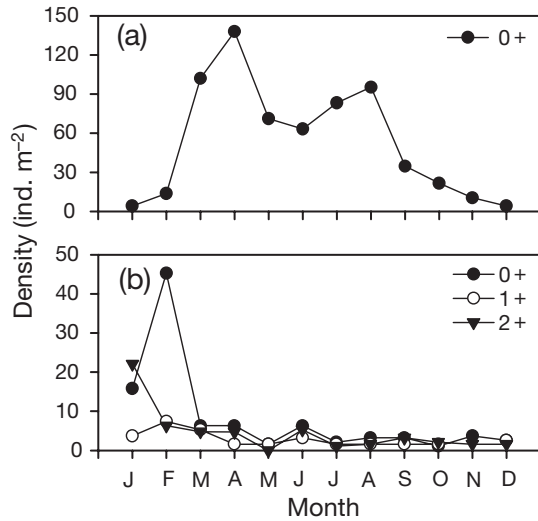


Fig. 4. (a) *Laternula marilina* and (b) *Moerella rutila*. Monthly variation of density (no. of ind. m⁻²) for each cohort

fucoxanthin, chl *c* and diadinoxanthin. Similarly to the water column, other pigment classes were nearly undetectable in the sediments except during February. However, the highest proportions of fucoxanthin, diadinoxanthin, diatoxanthin, and β -carotene in February indicated the predominance of diatoms (Cariou-Le Gall & Blanchard 1995).

Biometry and reproductive activities of bivalves

The mean annual densities of *Laternula marilina* and *Moerella rutila* were 54 (± 45) and 15 (± 17) ind. m⁻², respectively (Fig. 4). *L. marilina* solely contained the first year cohort, whereas *M. rutila* was comprised of different cohorts. Spat recruitment was initiated at the beginning of the investigation in January, but spat densities peaked in April (*L. marilina*) and in February (*M. rutila*), with 138 and 59 ind. m⁻², respectively. Thereafter, densities of both species decreased abruptly owing to high spat mortality. The density of *L. marilina* remained constant at around 80 ind. m⁻² from May to August, and then decreased rapidly until the end of December when few individuals were found. After February, the density of the *M. rutila* population remained constantly lower until the end of December, with the densities of all 3 cohorts being of equal proportion.

Seasonal trends in shell growth were similar between the 2 bivalve species (Fig. 5). Shell growth of *Laternula marilina* started in April and May and continued until August for the first year cohort (0+ year class). *Moerella rutila* also had fast growth during that period for the 3 cohorts, with a decreasing growth rate trend in the third year cohort.

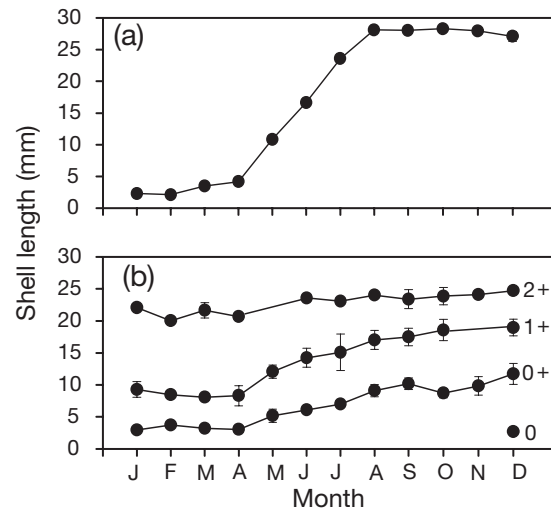


Fig. 5. (a) *Laternula marilina* and (b) *Moerella rutila*. Growth in shell length (mean \pm SD)

Seasonal variations in bivalve condition were very clear, with similar trends observed for both populations (Fig. 6). Condition started to improve in April and May for both species and continued until June to August for *L. marilina*, while the upward trend of *M. rutila* condition was prolonged until September to October. The condition maxima of the 2 populations were followed by a rapid decline in August to September and October to November, respectively.

In *Moerella rutila*, gonad production was observed only in the second and third year cohorts (1+ and 2+ year classes). The GMI data of the 2 bivalve populations showed that there was a good agreement in the gametogenic cycles between species except in November and December (Fig. 7). Gametogenesis of

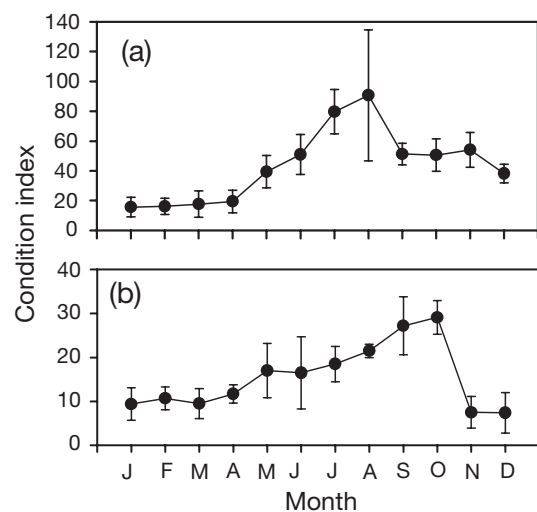


Fig. 6. (a) *Laternula marilina* and (b) *Moerella rutila*. Seasonal variation in condition index. Error bars are SD

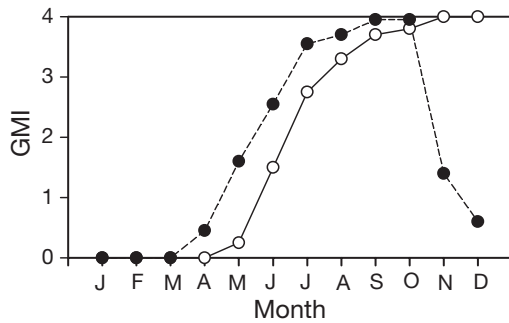


Fig. 7. *Laternula marilina* (○) and *Moerella rutila* (●). Monthly variation in gonadal maturity index (GMI)

Laternula marilina and *M. rutila* was initiated in May and April, and the GMI increased progressively until September and June to August, respectively. GMI values decreased rapidly in October to November for *M. rutila*, consistent with the rapid decline in condition. However, the mean *L. marilina* GMI displayed a maximum in November to December, without any sharp decline even after condition dropped in September.

Stable isotope ratios of bivalves and their potential food resources

Riverine POM had the lowest $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values among the 4 potential food resources measured, and varied between -26.6 and -25.0‰ with a mean of $-25.8 \pm 0.3\text{‰}$ ($n = 16$), and between 6.4 and 7.8‰ with a mean of $6.9 \pm 0.6\text{‰}$ ($n = 16$), respectively (Fig. 8). Values of sedimentary organic matter (SOM), macroalgae

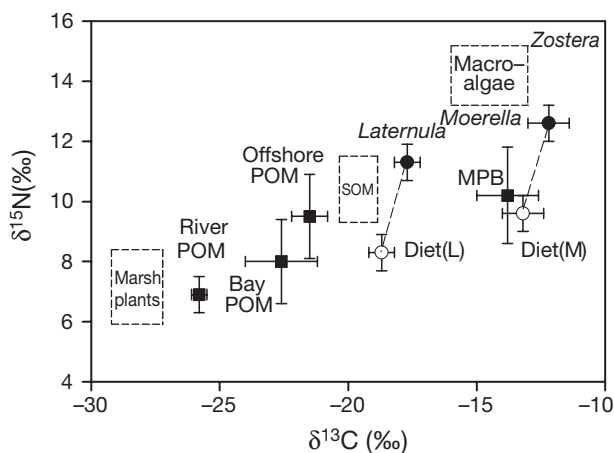


Fig. 8. *Laternula marilina* and *Moerella rutila*. Dual isotope plot of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ of bivalves and potential food sources. Points and bars = mean ± 1 SD; L, M, mean $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ of presumed diets of *L. marilina* and *M. rutila*, respectively, taking trophic enrichment (dashed line) into account; POM, suspended particulate organic matter; SOM, sedimentary organic matter; MPB, microphytobenthos

and *Zostera marina* previously reported in Kwangyang Bay by Kang et al. (2003) were close to values observed in marsh plants. Offshore sestonic POM had significantly higher $\delta^{13}\text{C}$ (-22.6 to -20.1‰ ; mean $-21.5 \pm 0.7\text{‰}$) and $\delta^{15}\text{N}$ (7.2 to 11.3‰ ; mean $9.6 \pm 1.4\text{‰}$) compared to riverine POM (1-way ANOVA, $p < 0.001$ for both cases, Fig. 8). $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values of bay channel POM were intermediate between these 2 components, and ranged from -25.2 to -20.5‰ (mean $-22.6 \pm 1.4\text{‰}$) and from 6.5 to 10.7 (mean $8.0 \pm 1.4\text{‰}$), respectively.

There was a significant seasonal variation in $\delta^{13}\text{C}$ in POM ($p < 0.001$, Fig. 9) in the bay channel, which exhibited the most negative values during the summer rainy season (June to July). Microphytobenthos had the highest $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values among the 4 potential food resources measured in this study, and ranged from -15.5 to -12.5‰ (mean $-13.8 \pm 1.2\text{‰}$) and from 8.1 to 12.3‰ (mean $-10.2 \pm 1.6\text{‰}$), respectively. Despite the statistically significant seasonal variations in the isotopic ratios of microphytobenthos ($p < 0.001$, Fig. 9), the ranges in isotopic values were quite narrow. There were clear differences in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values between microphytobenthos and bay channel POM advected to the tidal flat (2-sample t -test, $p < 0.001$ and $p = 0.008$ for C and N, respectively). The ranges of $\delta^{13}\text{C}$ from microphytobenthos and macroalgae overlapped. Both $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values of macroalgae and *Zostera marina* were previously reported to be highest among the POM sources in Kwangyang Bay (Kang et al. 2003) (Fig. 8). Therefore, a dual isotope plot of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ readily distinguishes potential food resources, and can exclude the isotopic ambiguities among them.

The monthly mean $\delta^{13}\text{C}$ values of *Laternula marilina* and *Moerella rutila* displayed no significant seasonal variations (1-way ANOVA, $p = 0.358$ and 0.331 , respectively), with relatively narrow ranges between -18.7 and -16.9‰ (mean $-17.7 \pm 0.5\text{‰}$) and between

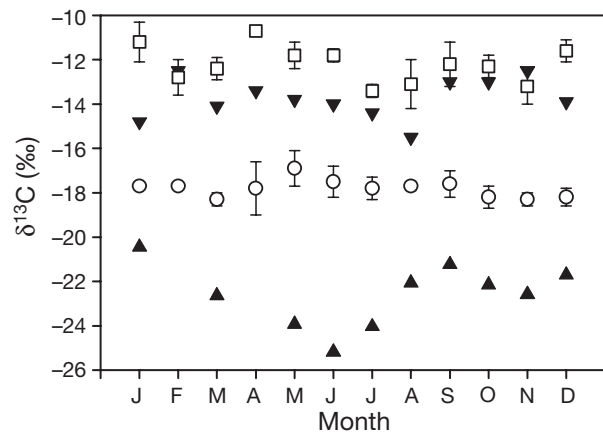


Fig. 9. Monthly variations of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values of bay channel POM (▲), microphytobenthos (▼), *Laternula marilina* (○) and *Moerella rutila* (□). Error bars are SD

-13.4 and -10.7‰ (mean $-12.2 \pm 0.8\%$), respectively (Fig. 9). Their mean $\delta^{15}\text{N}$ values ranged from 10.5 to 12.3‰ (mean $11.3 \pm 0.6\%$) and from 11.9 to 13.4‰ (mean $12.6 \pm 0.6\%$), respectively (monthly data not shown). There were significant differences in annual mean $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values between the 2 bivalve species (2-sample *t*-test, $p < 0.001$ for both cases, Fig. 8).

DISCUSSION

Distribution and seasonal development of microphytobenthos biomass

Microphytobenthos can be generally quantified by the chl *a* content. However, in earlier studies the chl *a* content was expressed using different estimates, often on a weight or areal basis for different sediment depths. Employment of different methodologies for chlorophyll analysis also makes comparisons among different investigations difficult (see MacIntyre et al. 1996). Sediment chl *a* contents measured in this study (range = 8.3 to 168.9, annual mean = $35.3 \pm 33.5 \text{ mg m}^{-2}$) were within the range of values reported from other intertidal flats (MacIntyre et al. 1996 and references therein).

Sediment chl *a* values varied greatly in space in our study area, and showed higher levels at the upper site than at the middle and lower sites (Fig. 3). Many abiotic and biotic controlling factors may result in such large spatial variability for intertidal microphytobenthos biomass (see Table 3 in MacIntyre et al. 1996, Cahoon & Safi 2002). While many studies have demonstrated that sandy sediments have lower microphytobenthos biomass than muddy sediments (de Jonge & de Jonge 1995, Montani et al. 2003), the reverse case has also reported (Miles & Sundback 2000, Cahoon & Safi 2002). Sediment variability was unlikely to be the cause of spatial variability in our study area because the sediments displayed a similar grain size at the upper, middle and lower sites, and were dominated by sand mixed with equal proportion of ~20% silt-clay ($>\phi 4$ or $<63 \mu\text{m}$). It is difficult to separately address the effects of other factors such as physical disturbance, nutrient availability, and macrofaunal grazing activity using our data. The most likely explanation for the variability may be the spatial difference in tidal elevation. Air exposure time is reduced at the lower intertidal site due to longer immersion time. This may result in reduced growth rates of microphytobenthos due to reduction of light penetration, elevation of sediment resuspension by wave activity, and elevated grazing pressure. Air exposure time might also influence the sediment surface temperature of intertidal flats, elevating temperature at the upper intertidal site, particu-

larly during spring-summer (Guarini et al. 1997), and thereby also the photosynthetic rate of benthic diatoms (Montani et al. 2003). A combination of these factors might support lower microphytobenthos biomass at the lower site.

In our study, microphytobenthos biomass was observed on a monthly basis for a whole year, and showed a strong seasonal variability at each sampling site with a uni-modal peak in summer (Fig. 3). Spatial differences in seasonal trends in the development of microphytobenthic assemblages have been previously reported (Wolfstein et al. 2000). Several authors have reported summer peaks similar to our study (de Jonge & de Jonge 1995, Montani et al. 2003). In contrast, others observed spring-early summer peaks and a subsequent decline in June or August due to grazing of benthic invertebrates and resuspension events (Cariou-Le Gall & Blanchard 1995, Wolfstein et al. 2000). Late winter peaks, as reported for other tidal flats (Montani et al. 2003), were not observed in our study.

Montani et al. (2003) reported a strong interannual variability in an estuarine sandflat of the Seto Inland Sea (Japan) and demonstrated that a seasonal decrease in microphytobenthos biomass in summer might be attributed to a seasonal reduction in irradiance and rainfall events. In our study, a peak in summer biomass made it difficult to infer the effects of grazing by benthic invertebrates and flushing by rainfall on microphytobenthos biomass during that time. Seasonal variability in microphytobenthos biomass on the tidal flat of Kwangyang Bay might be explained by the seasonal growth rate of microphytobenthos and the erosion of sediments. Increased growth rate due to high production can support the peak in summer biomass. Higher primary production in spring and summer than in fall and winter is very common for microphytobenthos, and is related to high daily quantum irradiance and to high temperature (Montani et al. 2003 and references therein). It is also assumed that extensive nutrient input through Seomjin River during the low-saline summer season affected primary production of microphytobenthos (Lee et al. 2001a).

A sharp decline in microphytobenthic biomass in September could be related to erosion due to typhoons and heavy rainfall in late August. The effect of sediment resuspension on microphytobenthos biomass has been pointed out (de Jonge & van Beusekom 1992, Cariou-Le Gall & Blanchard 1995). Due to its nearly closed geomorphology, local waves created in Kwangyang Bay play a minor role in sorting sediments (Park et al. 1984). However, the surface sediments were subjected to abrupt erosion by occasional typhoons and heavy rainfall resulting in a coarse-silt dominated texture in summer (Ryu et al. 2003). Based on a 2 yr data set, these authors showed that typhoons and heavy

rainfall accelerated erosion and coarseness of intertidal surface sediments in late August 2002. In contrast, the tidal flats of Kwangyang Bay receive a thin surface layer whose texture becomes finer due to a predominance of sedimentation, even during winter when the open-type tidal flats are exposed to large waves and thus greater erosion. Therefore, relatively low and constant microphytobenthos biomass in the bay after October was probably due to generally lowered daily quantum irradiance and temperature, rather than increased erosion of surface sediments during fall and winter.

Importance of resuspended microphytobenthos to sestonic POM pool

The seasonal variation of chl *a* concentration in the Kwangyang Bay water column conforms to a seasonal cycle typical of temperate zones, with an initial spring diatom bloom that occurs in April (Fig. 3). However, the chl *a* concentration in the water column did not display a minimum throughout the summer; rather, it continued to increase during spring and peaked in July to August. Chl *a* levels in the water column decreased at the end of summer and then remained low during late fall. An interesting question is whether the summer peak of chl *a* concentration in the water column comes from local production of phytoplankton in the water column, from advection of phytoplankton through tidal exchange with the adjacent open-sea area, or from the contribution of microphytobenthos via resuspension of sediments.

Despite large freshwater discharge through Seomjin River, the input of suspended particulate matter (SPM) from terrestrial sources is negligible except during the summer monsoon period so that SPM concentration is relatively low in the Seomjin River estuary (Kwon et al. 2002). Chl *a* concentration peaks at low salinity (5 to 15 psu) zone and thus production of living phytoplankton is primarily responsible for distribution of SPM along the salinity gradient in the estuary. However, SPM concentration is very high during the low saline summer monsoon period due to the enormous discharge of freshwater containing high concentrations of SPM. During that period, phytoplankton production is limited by increased light attenuation due to a seasonal maximum turbidity, and thus chl *a* concentration in the water column of the estuary did not peak even at the low salinity (5 to 15 psu) zone (Kwon et al. 2002). This phenomenon is likely to prohibit a high contribution of 'true' phytoplankton to the summer chl *a* peak in the water column. Reduced photosynthetic production in the water column due to light-limitation in turbid estuaries is a well-known

phenomenon (Shaffer & Sullivan 1988, de Jonge & van Beusekom 1992, Kromkamp et al. 1995).

An important feature of the bay is the large tidal prism volume relative to the bay volume. Due to the effect of large tidal exchange, the annual cycle of chl *a* inside the bay may be a reflection of that occurring in the adjacent area of the South Sea of Korea. Concurrent with summer chl *a* peaks in the bay, extensive blooms of dinoflagellates *Cochlodinium polykrikoides* have been observed outside the bay in the South Sea of Korea (Lee et al. 2001b, Kim et al. 2004). In this study, HPLC pigment analyses in water samples showed very low ratios of the dinoflagellate marker pigment, peridinin, to chl *a* (average 0.021 ± 0.017 , Table 1), indicating a very low relative proportion of microflagellate species in the waters of Kwangyang Bay. High ratios of diatom marker pigment, fucoxanthin, to chl *a* (average 0.316 ± 0.056) suggested that diatoms were dominant in the water column of the bay throughout the year (Table 1). This is an indication that annual cycles of phytoplankton development inside and outside the bay may be uncoupled. Differences in succession of phytoplankton between inner bay systems and coastal waters in the South Sea of Korea have long been observed (Kim et al. 2004). In addition, in 2002, frequent diatom blooms occurred in the inner bays while dinoflagellate blooms occurred in the coastal waters of the South Sea of Korea.

An alternative explanation for the summer chl *a* peak in the water column involves the importance of resuspension events of microphytobenthos along with surface sediments. The taxonomic composition of phytoplankton in Kwangyang Bay, which was dominated by diatoms as indicated by HPLC pigment analysis, was very similar to that of microphytobenthos on the bay sediments (Table 1). The most dominant phytoplankton taxa were including *Thalassiosira* spp., *Skeletonema costatum*, *Navicula* spp. and *Cylindrotheca closterium* (authors' unpubl. data). Two different concepts can be proposed to explain such a similarity in taxonomic compositions between the phytoplankton and microphytobenthos at shallow estuarine and intertidal locations (Cahoon & Safi 2002 and references therein). These authors assumed that sedimentary pigments accumulate from sinking phytoplankton. However, they also assumed that the dynamic physical processes at work in shallow estuaries might limit net deposition rate. Instead, they pointed out the contrasting possibility that resuspended microphytobenthos may make a considerable contribution to phytoplankton biomass in the water column in shallow estuaries. Indeed, several field and laboratory studies have clearly highlighted the fact that highly dynamic physical processes may result in the greater resuspension flux of microphytobenthos than the sedimen-

tation flux by sinking of phytoplankton (see references in 'Introduction').

An isotopic data set of POM, measured at 5 additional subtidal locations in Kwangyang Bay in 2004, appears to verify this hypothesis (authors' unpubl. data). A concurrent change in $\delta^{13}\text{C}$ of POM and salinity was noticeable at these sites (Fig. 10). Relatively negative values in POM $\delta^{13}\text{C}$ at the time of the abrupt decline in salinity indicated the input of a large quantity of riverine POM to the bay POM pool, probably due to high freshwater discharge. A high contribution of terrestrial POM was limited only to the short-term period of heavy rainfall in June and August. Except during the short-term heavy rainfall period, $\delta^{13}\text{C}$ values of bay POM were more positive than those of POM in the mouth of the bay ($-21.5 \pm 0.7\text{‰}$) during the late spring-summer period from 25 May to 14 September. POM in this estuarine bay is expected to be a mixture of riverine POM ($-25.8 \pm 0.3\text{‰}$) and coastal phytoplankton advected through the mouth of the bay (Fig. 11). However, a mixture of these 2 sources could not account for the unexpected ^{13}C -enriched values of POM in the late spring-summer.

A possible explanation for the ^{13}C enrichment of POM may be either the high productivity of phytoplankton or high abundance of ^{13}C -enriched phytoplankton due to a change in species composition (Tan & Strain 1983, Goering et al. 1990). However, phytoplankton productivity tends to be light-limited due to high SPM concentration during the summer monsoon (Kwon et al. 2002). Our pigment data also showed the consistent predominance of diatoms in the phytoplankton population throughout the year (Table 1). Therefore, changes in productivity and species composition of phytoplankton could not account for the ^{13}C enrichment of POM during the late spring-summer period. Another explanation for the ^{13}C enrichment of POM may be the existence of other sources of organic matter

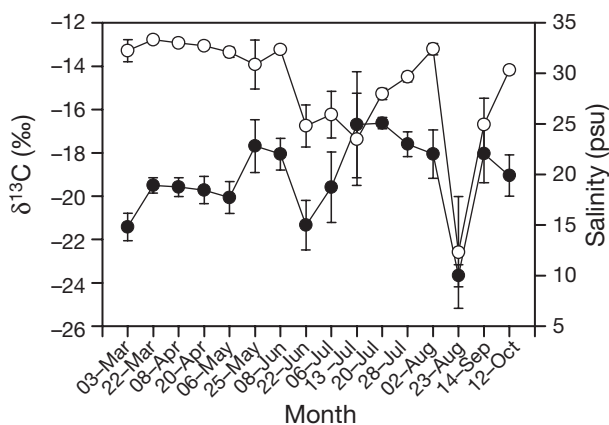


Fig. 10. Variation in salinity (○) and $\delta^{13}\text{C}$ of POM (●) measured for 5 additional channel sites from March to October 2004. Error bars are SD

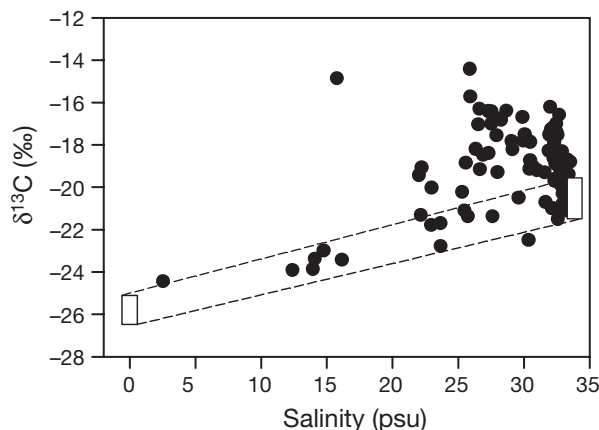


Fig. 11. Salinity versus $\delta^{13}\text{C}$ of POM measured for 5 additional channel sites from March to October 2004. Dashed lines indicate mixing line between riverine POM and marine POM (advected through the mouth of the bay)

such as macroalgae (around -14.2‰) and microphytobenthos (around -13.8‰ , Fig. 8), which have relatively high $\delta^{13}\text{C}$. Broad intertidal flats in Kwangyang Bay are indeed bare and are characterized by a large quantity of microphytobenthos (see above). Therefore, the isotopic evidence strongly suggests that the ^{13}C enrichment of POM reflects a considerable contribution of resuspended microphytobenthos to the bay POM pool.

Relationship between annual cycle of microphytobenthos biomass and bivalve activities

Laternula marilina and *Moerella rutila* at the intertidal site of Kwangyang Bay exhibited similar patterns of seasonal cycles in benthic activities. Our results clearly showed that shell and tissue growth (expressed by condition) and gonadal development in both species are achieved together during late spring and summer. This indicates that such a distinct pattern of seasonal cycle in their tissue growth was closely related to seasonal reproductive activity. Seasonal patterns in growth and reproductive activity of *L. marilina* and *M. rutila* are unknown as yet. Therefore, it was difficult for us to compare growth and reproductive activity of these bivalves with those in other locations.

The temporal overlap in tissue growth and gonadal development suggests that *Moerella rutila* can be considered an 'opportunistic' species where gonadal development depends directly on the quantity and quality of ingested food, rather than a 'conservative' species where gonadal development is indirectly dependent on nutrient stores accumulated during previous growth (Bayne 1976, see also Navarro et al. 1989). On the other hand, Hsueh (2003) showed that the population cycle of *Laternula marilina* is initiated

by larval settlement occurring as early as late March and culminates in a die-off at the beginning of December of the same year. He estimated a maximum life span of 9 mo. Our result is also in good agreement with this seasonal cycle. In our study, recruitment of *L. marilina* reached a maximum in April and growth continued until August. A mass of *L. marilina* spawns in summer (August to September). Most of the individuals die off immediately after spawning and so individual numbers dropped abruptly after September. The GMI maximum in November to December appeared to reflect this phenomenon.

Seasonal changes in temperature and food availability have been considered the main exogenous factors determining growth, reproductive activities and survivorship of marine bivalves (Bayne 1976, Mann 1979, Widdows et al. 1979). In the tidal flat of Kwangyang Bay, both factors showed nearly identical patterns of seasonal variation. For both species, the initiation of gametogenesis was coincident with spring temperature increase. This supports the idea that the seasonal fluctuation of temperature has an important effect on bivalve reproduction.

High food availability during the critical periods of macrobenthos life cycles is crucial in maintaining the structure and activity of the intertidal community. Temporal and spatial differences in the gametogenic cycle of many bivalves are attributable to temporal differences in food availability among locations (Newell 1982, Navarro et al. 1989). Knowledge of the relative importance of various algal components to bivalve activities is important in examining the factors that contribute to food availability during growth and gametogenesis of bivalves. For most marine bivalves, including those studied here, the spring bloom phytoplankton has been considered as a major algal component responsible for the growth and reproductive activities of marine bivalves.

In general, bivalves directly assimilate bloom phytoplankton as a means of energy acquisition for gamete production or for nutrient storage during the bloom for posterior gametogenesis. However, in the intertidal habitat of Kwangyang Bay, chl *a* concentrations increased during spring and peaked in summer both in the sediments and water column, as did activities of growth and gonadal development in bivalves. The results of this study demonstrated that the maximized activities of *Laternula marilina* and *Moerella rutila* are linked to sufficient food supply during late spring-summer. In particular, due to the recruitment pattern of *L. marilina*, their growth season tends to be related to seasonal food availability. Such high food availability may result from a high contribution of microphytobenthos, whose biomass peaks in late spring-summer. Resuspended microphytobenthos were likely to

account for a considerable part of microalgae present in the water column during that period, as discussed earlier. This implies that the summer bloom of microphytobenthos may be an important food source for growth and gonadal production of both deposit- and suspension-feeding bivalves, along with the availability of phytoplankton to the suspension feeder.

The food preference of suspension-feeding bivalves for fresh microalgal component has been well established (Kjørboe & Mohlenberg 1981, Prins et al. 1991). Kang et al. (1999) concluded that the relative dependence on benthic versus pelagic food resources during the growing season of the cockle *Cerastoderma edule* is primarily determined by temporal changes in the availability of these food resources. Selective feeding has also been reported for deposit-feeding bivalves (Hylleberg & Gallucci 1975). Field observation of Kamermans (1994) for 5 species of intertidal bivalves in the Wadden Sea indicated that deposit and suspension feeders could use the same food source. She found a similarity in algal composition in the stomachs of both groups, and a higher proportion of benthic algae in the stomachs compared to the algal composition of the water column sample 1 cm above the bottom. Lucas et al. (2000) demonstrated that resuspension-deposition processes of the microphytobenthos have important implications for benthic suspension feeders and reduction of microphytobenthos abundance through the grazing pressure of deposit feeders. Although a clear relationship between seasonal cycles in the bivalve activities and the microphytobenthos development was observed in the Kwangyang Bay tidal flat, simultaneous peaks in chl *a* concentrations in the water column and sediments still mask the relative contribution of these microalgal sources to the bivalve diets. Accordingly, the food sources actually assimilated by the bivalves were assessed by stable isotope composition of bivalves and their potential food resources.

The isotope signatures of potential food resources were well distinguishable by a dual isotope plot of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ (Fig. 8). Taking into consideration the documented enrichment of about 1‰ for carbon and 3.5‰ for nitrogen per trophic level (DeNiro & Epstein 1978, 1981, Minagawa & Wada 1984), $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values of the presumed diet would be ca. -18.7 and 7.8‰ for *Laternula marilina*, and ca. -13.2 and 9.1‰ for *Moerella rutila*, respectively. These values for *L. marilina* fell between those of the bay channel POM and microphytobenthos, which indicated the utilization of both of these as dominant food sources. Further, the isotopic values of the presumed diet of *M. rutila* clustered well with microphytobenthos, indicating exclusive use of microphytobenthos.

Other potential food resources were expected to play only a minor role in the diets of these bivalves. This

result may explain a closer link of deposit-feeding *Moerella rutila* to microphytobenthos compared with suspension-feeding *Laternula marilina*. On the other hand, while a distinct seasonal variation was detected in $\delta^{13}\text{C}$ of bay channel POM, a small but significant variation was observed in $\delta^{13}\text{C}$ of microphytobenthos (Fig. 9). Several studies have suggested a seasonal dietary shift as determined from concurrent changes in the isotope ratios of bivalve tissues and POM (Riera & Richard 1997, Kasai et al. 2004). However, despite the large seasonal variation in $\delta^{13}\text{C}$ of POM, little temporal change was detected in the $\delta^{13}\text{C}$ values of tissue from either bivalve in this study (Fig. 9).

As roughly estimated using a simple 2-source mixing model with $\delta^{13}\text{C}$ values of the bay POM and microphytobenthos as end-members (Fry & Sherr 1984), over 50% and nearly 100% of organic carbon incorporated into *Laternula marilina* and *Moerella rutila* tissues, respectively, was derived from microphytobenthos, irrespective of season. Such a dietary shift between feeding groups of bivalves have been reported (Herman et al. 2000). However, the relative dependence of the suspension feeder *L. marilina* on microphytobenthos in this study was higher than that cited by Herman et al. (2000). Page & Lastra (2003) found that intertidal suspension feeding bivalves primarily use resuspended microphytobenthos for much of the year. Our finding is consistent with that of Page & Lastra (2003). They also documented that the relative importance of phytoplankton and microphytobenthos sources to the bivalve diet may vary within and among estuaries. Our results indicated that although the bivalves utilize slightly different dietary sources depending on their feeding strategies, resuspended microphytobenthos are available even for suspension feeders most of the time in the Kwangyang Bay intertidal habitat. Further, the isotopic signatures of the bivalve tissues, compared with those of potential food resources, confirm the great dependence of intertidal bivalves on organic matter of a microphytobenthic source, despite a comparable contribution of phytoplankton to the diet of the suspension-feeding bivalves.

CONCLUSIONS

Both the suspension-feeding *Laternula marilina* and the deposit-feeding *Moerella rutila* showed very similar seasonal cycles in growth and reproductive activity when raised in a common habitat. Their peak shell and tissue growth and gonadal development being achieved together during late spring and summer. Seasonal patterns of growth and reproductive cycle similar to those found here for *L. marilina* and *M. rutila* have also been reported for the filter-feeding bivalves

Scapharca subcrenata (Lee 1998), *Tegillarca granosa* (Lee 1997), and *Mya arenaria* (Kim & Yoo 2001) inhabiting the adjacent intertidal flats. However, these authors were not able to compare bivalve activities to the annual cycle of food availability. Monthly observations of the quantity and composition of algal pigments, combined with analyses of stable isotope ratios from bivalve tissues and their potential food resources, allowed us to assess the means by which sufficient food is supplied for benthic activities of bivalves. Our results indicated that strong trophic links exist between microphytobenthos and intertidal bivalves throughout the year, depending on their feeding physiology and trophic/hydrodynamic characteristics, as previously reported for other tidal flats (Riera & Richard 1996, Kang et al. 1999, Herman et al. 2000). Therefore, rapid growth and gamete production of the bivalves during the late spring-summer in our study reflected the seasonal development of microphytobenthos available to the bivalves. Synchrony in growth and gonadal development of different bivalve species at the same place supports this hypothesis. Sauriau & Kang (2000) proposed the important contribution of microphytobenthos to the growth and secondary production of intertidal suspension-feeding bivalves, in contradiction to the general paradigm that marine phytoplankton is a major component of their diet. To our knowledge, our study is the first attempt to elucidate the relationship between seasonal development of microphytobenthos and growth and reproductive activities of intertidal bivalves. This study highlighted the importance of microphytobenthos as an available food source during the critical period in which gamete production and growth of both suspension- and deposit-feeding bivalves are concentrated.

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